

National Agricultural and Food Centre Slovak University of Agriculture Nitra NutriVet, Ltd. Pohořelice

# 17<sup>th</sup> INTERNATIONAL CONFERENCE

# FORAGE CONSERVATION



27<sup>th</sup> – 29<sup>th</sup> September, 2016 Horný Smokovec, Slovak Republic

# FORAGE CONSERVATION

Proceedings of 17th International Conference

27<sup>th</sup> – 29<sup>th</sup> September, 2016

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Edited by

Ľubica Rajčáková

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27<sup>th</sup> – 29<sup>th</sup> September, 2016 Slovak Republic

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# Foreword

We cordially welcome you at the 17th International Conference Forage Conservation in the beautiful mountain region of High Tatras, September 27-29.

For the National Agricultural and Food Centre – Research Institute for Animal Production Nitra, as well as the other organizers of the conference, it is a privilege to have been able to organize this event. We are pleased by you interest, thanks to which we are able to continue the long tradition of the silage conference in Slovakia. We believe you will be comfortable here and that we will be able to create an atmosphere, which will contribute to finding new professional and personal contacts, which will form the foundation of your future research and project cooperation.

Although the current situation in the Slovak agricultural industry is difficult, cattle husbandry and ruminant nutrition are the keystones of almost every farm's existence. The conditions in our country make year-round grazing impossible. Therefore, feeding the animals silages is inevitable during the winter season, even in the extensive husbandry. For summer seasons in the latest years periods of drought, whether short or long, have been characteristic, and those affect negatively the forage yield and no less also the quality of the forage. Most farms which are intensely engaged in cattle husbandry, and in particular the husbandry of dairy cows, have as a result turned to year-long silage feeding without grazing. Such husbandry system ensures stable feed doses, which positively affect the metabolism and health of the animals as well as the milk and meat production. The production of silages with high nutritional value and hygienic quality is therefore a priority for farmers. We believe exactly there is the space for researchers and scientists but also the experts from the sphere of commerce. The primary production companies needs the help and support from experts. Our mission is to explore new biological, chemical, and technological options and opportunities, which would lead to a higher quality and efficiency of produced feed.

We are honoured that over 87 participants from 16 countries of the world have registered for our conference. Participation of outstanding native and foreign specialists is a guarantee that the conference will contribute not only to development of new knowledge but it will contribute also to more effective transfer of scientific and research information into the user's sphere of service and primary production. These proceedings include 4 plenary papers and 51 papers, which will be presented at the conference in four sessions as an oral presentation or as a poster. The sessions cover the core areas of silage research from microbiology of ensilaging and feed safety to ensilaging technology and management.

We hope the 17<sup>th</sup> International Conference Forage Conservation will contribute to the implementation of new knowledge from the area of silage production and feed conservation into practice. We believe the issues connected to quality feed production will be intensely discussed among the attendees and your participation at the conference will be inspiring for your further work as well. We wish your time in Slovakia is well-spent as well as pleasant and that you return in the future.

Allow me to end with a respectful mention of our late colleage Prof. Ing. Alexander Sommer, DrSc. In these days we commemorate the 10<sup>th</sup> anniversary of his death as well as his 80<sup>th</sup> birthday. He was a man in a thousand and a prominent expert on animal nutrition. His scientific works dealt mainly with problems of nutrients digestion, proteosynthesis and proteolysis in ruminants, and ileal digestibility of amino acids in pigs, but he also left significant contributions in the fields of protein degradability, genetically modified feeds and relations between crude proteins and energy in nutrition of animals, as well as utilization of stimulatory substances and the ecological aspects of animal production.

There remains a dignified monument in the form of his lifetime-work and human activity, and he remains in our minds and in our hearts.

On behalf of the organisers

Ľubica Rajčáková Chair of the 17<sup>th</sup> ICFC **Man of Science** 

Homage to the Memory

of Alexander SOMMER

Research Institute for Animal Production Nitra – National Agricultural and Food Centre



To the bright memory of Professor Dipl. Ing. Alexander Sommer, DSc.

In 2016, Professor Dipl. Ing. Alexander Sommer, DSc. would be celebrating his 80<sup>th</sup> birthday. Instead, we are commemorating the 10<sup>th</sup> anniversary of his untimely departure from this world.

Prof. Sommer, who devoted his energy to the Research Institute for Animal Production in Nitra, spent his entire working life engaged in the sphere of agricultural science and animal nutrition. For many years he was the head of the Institute of Animal Nutrition within RIAP Nitra, a department he built and developed. He played an important role in the development of first Czechoslovakian and later Slovak agricultural science. In the world as well as in his home country, he was a well-known and well-respected researcher. Prof. Sommer together with Dr. Miroslav Škultéty, substantially contributed to the establishment of the workplace on forage conservation in Nitra. They also generated new tradition of organizing regular symposia on forage conservation. He remains an influence on and an inspiration to the field of animal nutrition today.

However creative, talented and filled with genuine enthusiasm for his work, Prof. Sommer was also a principled and disciplined man with a drive towards perfection. His work was recognized at home and abroad, and led to many professional successes. He was both a scientist and an educator, whose influence on the researchers he supervised shapes the face of Slovak animal nutrition research even today. His intent was never simply to do science for the science's sake; all his efforts in research as well as management aimed towards science for the welfare of humanity. He was also an ambassador of RIAP Nitra for the world. As a guest lecturer, he visited universities not only at home but also abroad and participated in numerous conferences, and was involved in international cooperation, including the participation in and coordination of international research projects and experiments.

Prof. Sommer was one of those people who devote themselves to their profession wholeheartedly. Science was for him not a work but a calling. Retirement was a distant future he would postpone indefinitely every time it approached as there was always a project requiring his attention. His mind could simply not suffer even the idea of idleness. Therefore, he left us too early, with unrealized plans and intentions.

At the Institute for Nutrition within the Research Institute for Animal Production Nitra, however, we continue to build onto the foundation laid by his work and his legacy, and his memory, stays with us and lives on in our effort and successes.

Nitra, September 2016

Research team of Institute for Nutrition

# PLENARY SESSION – INVITED PAPERS

<u>Utilization of perennial fodder crops for production of high quality silages</u> Hakl, J., Šantrůček, J.

Volatile organic compounds (VOC) in silages – effects of management factors on their formation Waiss K

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Fibre and protein quality of silages and their effects on ruminant performance Nadeau E., Richardt W., Nørgaard P.

<u>A review on additives for grain silages</u> Morais, G., Daniel, J.L.P., Siqueira, G.R., Silva, N.C., Schonell, E.P., Nussio, L.G.

# Utilization of Perennial Fodder Crops for Production of high Quality Silages

**BACK** 

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# INTRODUCTION

Perennial fodder crops such as legumes or grasses play an important role in ruminant nutrition all over the world. In the regions where area of permanent grassland is limited represent the very important protein and digestible fiber source. Among these crops, forage legumes such as lucerne or red clover represent a major protein source for ruminant nutrition in Europe (Krawutschke *et al.*, 2013). For successful forage production, high forage value of stand is generally required. This value consists of two parts: forage yield and quality. In spite of forage quality evaluating by a range of parameters, forage dry matter yield is only one-dimensional variable. It must be remembered yield is a key factor in forage production, especially for economic efficiency in relation to cost per hectare. There is a range of factors influencing both forage yield and quality. The aim of this review is not list all of them but provide basic knowledge as well as trends at these with the highest practical impact.

# SUITABLE SPECIES/CULTIVARS SELECTION

Effective forage production cannot be naturally the same across different environments. In this regard, species or cultivars selection generally represents simple but highly effective tool for adaptation of feedstuff production to environment conditions. For example, in temperate zones, biennial or perennial legume or grass species are the most common whilst annual species can be in the first place in regions with intensive drought or frost period. For condition of the Europe, the most important traditional forage legume crops are lucerne (*Medicago sativa*) and red clover (*Trifolium pratense*). These two crops have complementary production responses to climatic conditions, where lucerne is high yielding in dry whilst red clover in wet conditions (Peterson *et al.*, 1992). Regrading to grasses in this zone, the most important species are perennial and Italian ryegrass (*Lolium perenne* and *multiflorum*), meadow and tall fescue (*Festuca pratensis* and *arundinacea*), timothy (*Phleum pratense*) and orchard grass (*Dactylis glomerata*). There is a range of other grass species important for specific environment. Properties of all mentioned species are described in detail for example by Frame *et al.* (1997). Selection of proper species or composition of mixture of species remains the basic tool for successful start of forage production.

Breeding process is intended to improve the properties of the selected species important for human civilization. Yield improvement in forage crops during last century has lagged far behind that of annual grain crops (Brummer, 1999) because breeders changed rather harvest index than total biomass production, which is explanation for low yield progress of perennial forage crops. Lamb *et al.* (2006) concluded that evidence for changes in lucerne forage yield for cultivars released between 1940 and 1995 was environmentally depend. In environments where conditions lead to plant stand losses, recently released cultivars with multiple disease resistance had a yield advantage over older cultivars, but in environments where no differences in plant density occurred, older cultivars yielded the same as the improved new cultivars.

Except of this slow but continuous improving of yield or quality, there were also some milestones in this process at perennial forage crops. The following is an example with high practical impact on forage production. It has long been a goal for forage breeders to combine the stress tolerant characteristics of *Festuca* species with the earliness and high nutritive value of *Lolium* species. Some breeding programs have been designed to transfer *Festuca* genes into *Lolium*, and as a result some *Festulolium* cultivars have been developed in Europe and in the USA (Humphreys *et al.*, 2003). *Festulolium* provides specialist function and novel alternatives to existing grass species/cultivars that may lack resilience against abiotic or biotic stresses.

Following example represent a case of high perspective breeding method for improving forage quality. Lignin is defined as a complex organic compound that binds cellulose fibers and hardens and strengthens the cell walls of plants. This process accelerates as plants mature and gives structural support to the plants as they become taller. Regarding to animal nutrition, lignin is a well-known as highly important anti-nutritive substance which is still essential for plant functions. Due to its negative effect during digestion, experiments with genetically reduced lignin synthesis have been made in various plant species. According to Shadle *et al.* (2007), analysis of lucerne forage quality parameters showed strong reductions of neutral- and acid-detergent fibre in the down-regulated lines, in parallel with large increases (up to 20%) in dry matter forage digestibility. Reduction of HCT enzyme activity in these lines was from at least 15–50%. The most severely down-regulated lines exhibited significant stunting, reduction of biomass yield and delayed flowering. Vascular structure was impaired in the most strongly down-regulated lines. Although manipulation of lignin biosynthesis can greatly improve forage digestibility, accompanying effects on plant development need to be better understood. In spite of these distresses, there was released first low lignin lucerne cultivar for the commercial utilization by Aflorex Seed Company. In these "Hi-Gest" cultivars, content of lignin is reduced by 7 - 10% without declared

negative impact to agronomic traits. The grower has two general harvest options available when growing lucerne with this new technology: (1) harvesting fields on a normal  $\sim 28$  day cutting schedule to produce a high quality forage that has increased fiber digestibility and higher animal intake; (2) extend the peak harvest date by up to 7 days to  $\sim 35$  days versus 28 days. This option utilizes the low lignin trait as a means of increasing yield without sacrificing forage quality. If a field is ready to be cut but rainy weather is forecast by delaying harvest up to 7 days to avoid rained-on forage. This flexibility at harvest time helps the producer minimize the effect of improper weather and reduced forage quality. Synthetic cultivars harvested at the later date would have lower forage quality due to its maturity and higher lignin content.

# HARVEST FREQUENCY

Optimal stand utilization is very important in terms of both yield and forage quality. It is well known higher cut frequency improves nutritive value of harvested forage because of reduced stem weight proportion and its better digestibility in relation to lower lignification. However, it must be remembered that more intensive cut regime reduces stand yield and persistence. Regarding to yield, our results show that four-cut regime obtained significantly lower yield than three-cut in Central Europe region but this reduction was represented only 4 - 5% (Hakl et al., 2011). For this environment, it seems that one cut over standard intensity of utilization only slightly reduces yield but provides high potential for improving forage quality. The adverse effect of intensity of lucerne harvesting on persistence and the following spring regrowth has been historically attributed to a reduction in the concentrations of organic reserves, especially total non-structural carbohydrates. For this purpose, it must be carefully distinguished between effect of number of cut per year and their schedule over year. The regrowth interval between the last summer harvest and the autumn harvest is the major determinant of lucerne persistence and spring regrowth (Dhont et al., 2004). In Central Europe, this interval was traditionally expressed as number of days when should be at least 50 days. The accumulation of growing degree days > 5° after the last summer harvest has been proposed as a criterion to estimate the duration of this interval (Bélanger et al., 1992). For investigation this matter in Europe condition, the field experiment was conducted in Central Bohemia in 2002-2004. In this experiment, the interval between summer and last autumn harvest was 40–50 days or 60–70 days, respectively. These intervals were expressed as cumulative growing degree-days (GDD) where GDD values ranged from 540 to 905 over three years period. The plants were sampled in each autumn with four replicates for each variant; the average depth of sampling was 150 mm. The weight of roots, amount of starch, and water soluble saccharides (WSC) per m<sup>2</sup> was determined. The total accumulation of root reserve saccharides was determined mainly by conditions over growing period in particular year. The length of the interval or cumulative GDD influenced only variation of this basic amount. It was documented by significant differences among evaluated years in dependence on weather condition and following stand development. In the Table 1, you can see reduction of starch concentration and amount of all reserves at early harvest interval in 2002. GDD was very high at both intervals in 2003 which resulted in no significantly different amount of root reserves between intervals. In 2004, higher GDD value was obtained at early interval in spite of lower number of days which resulted in significantly higher concentration of starch. Total amount of root and root reserves was not affected by length of the interval. In Central Bohemia condition, the GDD around 600-700 °C was preliminary determined for maximal accumulation of root reserve saccharides. The GDD above this level did not significantly increase the root reserve accumulation.

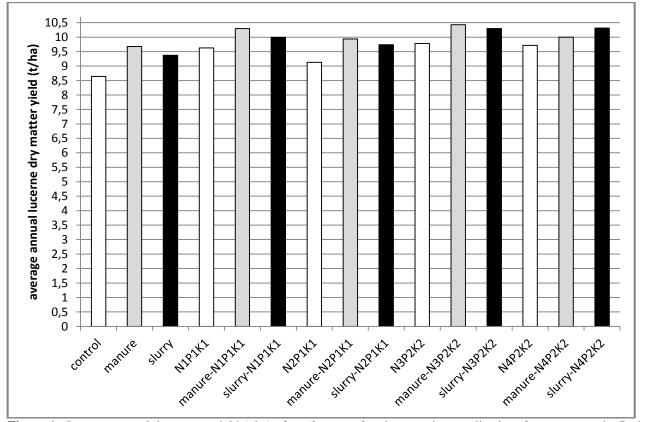
2002					2003			2004		
	early	late	Р	early	late	Р	early	late	Р	
Interval:GDD	540	850		693	905		734	621		
Interval: days	43	72		42	67		54	63		
concentration (g.kg <sup>-1</sup> )										
starch	104	126	0.0045	174	153	0.0022	199	142	0.0049	
WSC	144	147	0.5766	193	188	0.3492	141	150	0.6663	
amount (g.m <sup>-2</sup> )										
starch	17	33	0.0004	22	22	0.9090	38	30	0.5263	
WSC	23	39	0.0016	24	27	0.3782	27	31	0.4543	
total	40	72	0.0008	46	49	0.5884	65	61	0.5841	
root	158	261	0.0006	125	144	0.2686	190	212	0.6367	

 Table 1
 Effect of length of regrowth interval between the summer and autumn harvest on concentration and amount of starch and water soluble saccharides (WSC) in lucerne roots over three year period (site Červený Újezd).

P = probability of F test, different letters document statistical differences in each column (Tukey HSD,  $\alpha = 0.05$ )

# FERTILIZATION

Intensive agricultural cropping system requires large quantities of plant nutrients (Lloveras et al. 2012) which highlight importance of suitable fertilization management. It can be shortly assumed that lack of nutrients in the soil reduced significantly forage yield. The impact of forage legumes fertilization has been traditionally focused on effects of direct application of phosphorus (P) and/or potassium (K) in various combinations (Macolino et al. 2013) whereas direct application of nitrogen (N) is not usually effective due to N fixation by legumes. In spite of intensive previous research about lucerne fertilization, there is a lack of long-term studies investigating indirect effect of organic and N fertilization on yield within applied crop rotation. In present, we can investigate differences in forage yield under different combination of mineral (6 treatments) and organic (3 treatments) fertilization in long-term experiment conducted since 1955 in Ruzyně. For more detail about site and experiment description see Hakl et al. (2016b). Longterm absence of fertilization provided average annual dry matter yield 8.64 t/ha which is presented in Figure 1. Indirect application of mere manure or slurry significantly increased yield to 9.68 and 9.37 t/ha, respectively. The highest values of DMY over 10 t/ha were observed at treatments, where organic fertilizers were applied at N3P2K2 and N4P2K2 treatment, however the same value was also observed at application of manure under N1P1K1 treatment. These results reveal that not only direct but also indirect fertilization substantially influenced lucerne DMY (Hakl et al., 2016b). Effect of fertilization is generally more obvious for yield than forage quality and there is only few studies about effect of fertilization on nutritive value of perennial fodder legumes. According to Lissbrant et al. (2009), low P and K soil fertility reduced fibre concentrations in the lucerne forage. This is in line our preliminary results from long-term experiment in Ruzyně, where variable fertilization resulted in different stand structure. The highest plant density was observed in control, slurry or manure treatments. Increasing rate of N reduced plant density but maintained stem density up to N3 level. Intensive fertilization also increased stand height which was in line with lower leaf weight ratio. These investigations suggest explanation for reduced forage nutritive value under higher nutrient supply described by Lissbrant et al. (2009). Further research is warranted to identify the influence by which long-term fertilization management affects lucerne yield components, nutrients content and digestibility within separate lucerne leaves and stems.

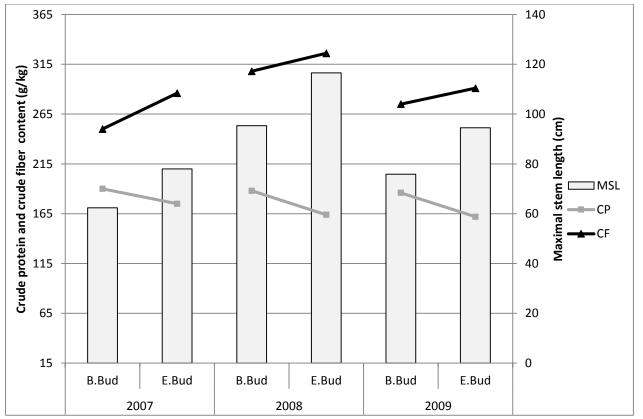


**Figure 1** Lucerne annual dry matter yield (t/ha) after 60 years of various nutrient application (8-year mean, site Praha – Ruzyně)

# FORAGE QUALITY PREDICTION

Timing of the forage harvest is critical for obtaining optimal quality for animal production. For forage crops that serves as the primary fiber source in the diet, NDF is the principal forage quality variable of concern (Parsons *et al.*,

2006a). Some predictive equations can be used to estimate the forage quality, assisting the producers in decision making at harvest time. Parsons et al. (2006b) described an ideal method for estimating quality in the field as a harvest decision aid must be quick, simple, inexpensive, and consistent across all harvests during the season and across a wide range of environments. The most widely used of these are the predictive equations for alfalfa quality (PEAQ). This method is based on the length of the tallest stem and the stage of the most mature stem in the sample (Hintz and Albrecht, 1991). These equations have been developed for many regions of the USA. Results indicated some bias in using the equations outside the state of development; however, the prediction errors have been sufficiently low to suggest the PEAQ equations are robust over a wide range of environments (Parsons et al., 2006a). GDD are a temperature-derived index representing the amount of heat to which plants are exposed. It was used similarly to assessment of length of interval between harvests which was mentioned above. This method has been used with mixed success with the perennial types of forage (Sulc et al., 1999). In the Czech Republic, these methods have not been tested for any perennial forage crops; therefore Hakl et al. (2010) have tested their accuracy and suitability for lucerne prediction within the first cut period in Central Bohemia. Their results revealed higher accuracy for PEAQ in comparison with GDD. Suitability of PEAQ method was later reported by Anderzejewska et al. (2013) also for northern Europe. Further our research has shown that the developmental stage was not suitable indicator for forage quality in year with untypical weather condition (see Figure 2). The best solution was a combination with stem length with clear relation to crude fiber content whilst a lower relation was observed to crude protein content. For optimal lucerne quality, the term of first harvest should be in a bud stage when the stem length is to 60 - 65 cm. Recent investigations in this research area has shown that canopy reflectance (i.e., remotely sensed) data may allow rapid assessment of nutritive values, such as total N, neutral detergent fiber (NDF), and acid detergent fiber (ADF) of lucerne. The remotely sensed based prediction equations explained from 78 to 83% of the variation in measured total N, NDF, and ADF, correctly predicted about 78% of the measured TDN/CP ratios. This technology could help improve profit margins by timing the cutting or harvesting of alfalfa, in rapid assessment of nutritive values over large areas devoted to growing alfalfa, and assessing nutritive quality in real time (Starks et al. 2016).



**Figure 2** Lucerne maximal stem length (cm), crude protein and fiber content (g/kg) in the first cut (3-year period, site Červený Újezd).

## FORAGE CONSERVATION

Forage conservation cannot be excluded from group of highly important factors affecting quality of feed for animals; however its impact is strongly limited in terms of forage quality before conservation. In this case, we properly cannot talk about increasing of quality but only about decreasing nutrients or quality losses during conservation process. Despite this limitation, the conservation of forage crops is one of the most risk-intensive processes undertaken by farm managers. From the time of harvest until it is used as feed, it is subject to significant losses both in quantity and quality. These losses occur during harvesting and field operations, and later during storage and handling of the product. To minimize the risk associated with forage conservation it is important to understand these processes, how they interact with one another, and how their effects can be mitigated through various management practices. These management tools were summarily described for example by Moore and Peterson (1995). However, this topic is the subject of the next conference section.

#### **PROTEIN UTILIZATION**

Contrast to previous topics, this theme covers area about specific evaluation of legume forage quality in connection with animal utilization. According to our opinion, it is very hot and highly important topic therefore was included in this review. Protein degradability in forage legumes is of global importance because utilization efficiency of forage has economic and environmental consequences. Rumen protein degradation and the resulting imbalance between carbohydrate and protein supply leads to lower N-use efficiency by ruminants (Broderick, 1995). Increasing the amount of protein that escapes from the rumen could benefit ruminant nutrition and improve the economics of the dairy industry (Chen et al., 2009). The most commonly studied factors affecting protein fractions include plant species and harvest maturity (Kirchhof et al., 2010; Krawutschke et al., 2013). Previous studies (e.g. Lemaire et al., 2005) have shown that N concentration in forage is closely related to plant morphology in the lucerne stand. In spite of it, in almost all published studies that investigated CP fraction of legumes, information on stand traits was not presented (e.g., Kirchhof et al., 2010; Krawutschke et al., 2013). Therefore, we hypothesize that changes in plant morphology within a dense canopy could also be connected to variation in CP fractions. Within two year period, lucerne leaf and stem samples were taken in the three cuts and plant density, stem density, maximal stem length and leaf weight ratio were assessed. All dried stem and leaf samples were milled to pass through a 1 mm screen and analysed for CP fractions according to Licitra et al. (1996) where protein content was fractionated into A and B<sub>1</sub> (soluble fractions), B<sub>2</sub>, and insoluble fractions B3 and C. Recently published results of Hakl et al. (2016a) suggest that stand traits make an important contribution, accounting for about 75% of CP fraction variability. Above all, maximal stem length is a variable that can be easily assessed for individual plants and has a strong negative correlation with leaf weight ratio, which is assessed as less easily than maximal stem length. The findings of this research indicate that plant morphology should be considered, particularly when evaluating the genetic variability of the CP fraction within legume species (Tremblay et al., 2003) or measuring protein composition among lucerne cultivars (Chen et al., 2009).

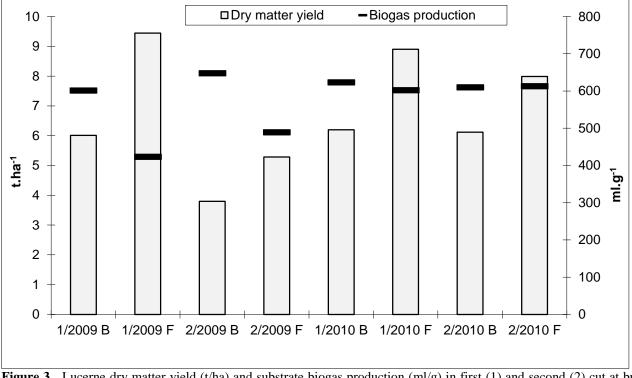
# ALTERNATIVE FORAGE UTILIZATION

Traditional utilization of forage biomass is connected with ruminant nutrition. In many European countries, decrease of number of cattle units in connection with recently low milk production profitability make an issue with utilization of produced forage (e.g. Stypinsky *et al.* 2009). This is key problem for permanent grassland because grassland area cannot be reduced due to environmental impact in landscape. In the arable land, perennial fodder crops simply are not included in a crop rotation. However, absence of these crops together with lower production of organic fertilizers has negative impact on soil fertility and balance of organic matter. From these reason, we are looking for alternative utilization of these crops for various purposes. For example, there is a tendency for utilization of forage legumes as a protein source for monogastric animals, pharmacy or human nutrition. In spite of these minor possibilities, the major activity is energy production from forage biomass because generation of energy from biomass has a key role in current EU strategies to enhance energy security. In present, biogas production from energy crops in the arable land is mainly based on the anaerobic digestion of maize. The maize achieved the highest methane yield per hectare in comparison with cereal or sunflower (Amon *et al.* 2007). On the other hand, it must be note that maize growing is limited in some areas and can have some negative impact on environment as higher pesticide and fertilizes requirements. Maize fields are, in general, relatively vulnerable to both water and wind erosion (Graebig *et al.* 2010).

Unlike maize, biogas production from lucerne or clover forage is not common practice. Legume crops could be also suitable source for biogas production and it is generally accepted that their growing significantly improve soil fertility in contrast to maize growing. According to Walla and Schneeberger (2006), lucerne grass mixture is more efficient energy crop than silage maize on organic farms. Forage legume stands seem to be a suitable biomass source because of its persistency, high productivity, self-sufficiency of  $N_2$  and positive impact on soil fertility. According to Amon *et al.* (2007), specific harvest and processing technologies are required when crops are used as a renewable energy source compared to growing them as a forage source for ruminants. The traditional harvest management for livestock feed recommend the cut term in the bud stage in relation to high quality of forage (Hakl *et al.* 2010). In contrast to it, the suitable harvest management system produced more total forage than a three- or four- cut management system harvested at early bud (Lamb et al. 2003). The impact of changes in lucerne biomass quantity and quality under different harvest management could be different for biogas production in comparison with animal utilization.

For clarifying these relationships, we tested biogas production from lucerne biomass over two year in field plot experiment. Biomass was tested in 120 ml bottles in five replications for each variant. After basic homogenization and grinding of fresh matter, two grams of tested biomass and 80 ml of inoculum were dosed into fermentors. Active mesophile anaerobic sediment from biogas plant was used as the inoculum. Cultivation took place in thermo box at 40 °C for a period of 40 days. Production of biogas in laboratory tests of biomass was evaluated once a day, using gas-

metric burette. In figure 3, values of substrate biogas yield were in wide range of 423 to 648 L/kg DM. When 10% as average ash content in lucerne forage and 60% methane content in biogas is considered, methane yield from 280 to 430 L CH<sub>4</sub>/kg<sub>OM</sub> could be obtained. This range corresponds with results published by Amon et al. (2007) about methane yield from other energy crops. The average methane yield 398 L CH<sub>4</sub>/kg<sub>OM</sub> was obtained from maize silage whilst from wheat ranged between 140 and 343, from sunflower between 154 and 454, and from grassland between 128 and 392 L CH<sub>4</sub>/kg<sub>OM</sub>. As was noted by Prochnow et al. (2009), the aim of energy crop for biogas production is to achieve the highest possible methane yields per hectare. Results show that area biogas yield from lucerne forage could be significantly increased by change in harvest management towards to delayed cuts. It is in accordance with Lamb et al. (2003), that harvesting twice per season at a later maturity stage would be an effective management strategy for maximizing yield in a lucerne biomass energy production system. In our study with biogas production, the average increase of yield in late flower stage was relatively stable across year and achieved approximately 50 and 35% in the first and second cut, respectively. In spite of substrate biogas yield higher about 25% in the bud stage in 2009, the higher area biogas yield was produced in late bloom stage. These result about increasing area biogas yield in spite of decrease substrate biogas yield support idea, that requirements on the biomass quality are different when crops are anaerobically digested in biogas plants compared to being fed to cattle. The reason could be that the digester at the biogas plant offers more time to degrade the organic substance than the rumen does. Another important point could be a different micro-organism population in the digester (Amon et al. 2007) or fact that higher proportion of NDF in the forage does not result in lower dry matter intake in the case of biogas plant. In this experiment, lucerne reached lower methane yield per hectare in comparison with maize and probably would not play a dominant role in biogas production from crops growing on arable land. Nevertheless, the methane yield of lucerne seems to be higher or comparable with other crops as cereal or sunflower and lucerne growing could be a suitable supplement for biogas production due to lucerne no-productive function with positive impact on soil fertility and reduction of soil erosion.



**Figure 3** Lucerne dry matter yield (t/ha) and substrate biogas production (ml/g) in first (1) and second (2) cut at bud (B) and flower (F) stages in 2009 – 2010.

### CONCLUSION

Successful forage production through perennial fodder crops depends on achieved biomass yield and high forage quality. In this review, we would like to highlight the importance of agronomic decisions on harvested forage quality before start of conservation process. Although forage yield at the time of animal feeding is aloof interest, contributed substantially to economic of animal production.

### ACKNOWLEDGEMENT

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# Volatile Organic Compounds (VOC) in Silages – Effects of Management Factors on their Formation

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# INTRODUCTION

Based on empirical observations from commercial farms that well preserved, but odd-smelling maize silages may cause problems regarding feed intake and milk yield by dairy cows, volatile organic compounds (VOC) were analyzed in recent studies (Weiss et al., 2009a, b). Ethyl and propyl esters of lactate and acetate have been found in farm silages (Weiss, 2009a, Weiss et al. 2015). According to Kriszan et al. (2007), Raun and Kristensen (2010) and Gerlach et al. (2013), feed intake was negatively correlated with the concentration of some VOC. The knowledge on the effects of specifically VOCs on feed intake by ruminants is still very limited. In addition, those substances have been discussed in relation to climate-damaging ozone formation, and it was reported that silages on dairy farms may be a significant source of VOC emission (Mitloehner et al., 2009).

Correlations were found in maize silages between ensiling conditions, type of silage additive as well as ethanol content and the concentrations of the ethyl esters – ethyl acetate (EA) and ethyl lactate (EL) (Weiss et al., 2016). Ester and ethanol levels were highest in silages stored under strict anaerobic conditions. Elevated levels of ethanol and the corresponding esters EA and EL were not only detected in maize silages, but also in silages from grass, grass-legumemixtures, legumes, whole-crop cereals and sorghum (Weiss and Auerbach, 2012a, b, Weiss and Kalzendorf, 2016). Regardless of silage type, silage additive and ensiling conditions, in the most cases there was found a strong correlation between ethanol and ester concentrations, highlighting the prominent role of alcohol in the ester formation. Therefore, any measure that reduces ethanol will restrict ester content.

# OCCURRENCE OF VOLATILE COMPOUNDS IN SILAGES

# VOC in maize silages

On the basis of the results from investigations in farm silages with well preserved, but odd-smelling maize silages the first lab scale ensiling experiments were carried out with maize (Table 1). Elevated levels of ethanol, EA and EL were detected in maize silages, but also in silages from whole-crop wheat and high-moisture corn (Table 2). Ester and ethanol levels were highest in silages stored under strict anaerobic conditions. It was also shown that esters remain detectable in silages for a few days after opening of the silos under aerobic conditions (Weiss et al., 2011). Data from farm silages presented in Table 2 (whole-crop maize 6 and 7) also showed high ethanol and ester concentrations. These silages were well fermented and highly compacted. In 7 out of 14 silages biological additives were used.

Results of ensiling experiments concerning effect of storage period on fermentation pattern indicates that concentration of ethanol strongly effected formation of esters during fermentation process. Weiss et al. (2009b) found increasing contents of ethanol and especially lactic acid over 90 days, whereas the corresponding ethyl esters increased during the first 30 days. This findings is in line with results from Gerlach et al. (2015) which investigated the effect of storage length of different maize silage varieties.

Lab-scale ensiling exp Whole-crop maize 1 Whole-crop maize 2 Whole-crop maize 3	<b>periments</b> 310 316 349 332	60 30 180	60, 90 2,14, 28, 49, 90	biological, chemical (Weiss et al., 2009a) biological (Weiss et al., 2009b)
Whole-crop maize 2 Whole-crop maize 3	316 349	30	2,14, 28, 49, 90	<b>C</b>
Whole-crop maize 3	349			biological (Weiss et al. 2009b)
1		180		
	222		2,14, 28, 49, 90	biological, chemical *)
Whole-crop maize 4	332	12	90	chemical (Weiss and Auerbach, 2012b)
Whole-crop maize 5	315 - 513	79	112 anaerobic, 0-8 aerobic	without (Gerlach et al., 2013)
Whole-crop wheat	276	34	60, 90	biological *)
High-moisture corn	635	30	97	biological, chemical (Auerbach and Weiss, 2011)
Commercial farm sila	ages			
Whole-crop maize 6	254 - 322	3	approx. 90	without (Weiss et al., 2009a)
Whole-crop maize 7	299 - 403	11	approx. 90 - 180	biological, Weiss et al., 2016)
				* (Weiss and Auerbach, unpublished)

Table 1. Characterization of the data set of maize silages (ensiling experiments, n= 439)

**BACK** 

Type of Silage	Lactic acid	Acetic acid	Ethanol	Ethyl	Ethyl	Regression
				acetate	lactate (EL)	EA+EL(y),
				(EA)		Ethanol (x)
	g/kg DM	g/kg DM	g/kg DM	mg/kg DM	mg/kg DM	$y = ax + b$ $R^2$
Whole-crop maize 1	6.9 - 74.8	5.8 - 79.4	0.9 - 51.7	12 - 284	16 - 379	12.50x+ 91.2 0.70
Whole-crop maize 2	32.5 - 119.8	8.6 - 25.8	3.2 - 28.3	55 - 343	30 - 683	26.47x+121.5 0.65
Whole-crop maize 3	13.7 - 67.4	0.5 - 26.7	3.3 - 20,1	38 - 639	0 - 224	18.10x+ 91.7 0.20
Whole-crop maize 4	73.8 - 124.6	5.3 - 29.2	6.2 - 50.8	116 - 262	156 - 661	11.55x+266.0 0.93
Whole-crop maize 5	0 - 75.5	0 - 36.6	0 - 36.9	0-1109	0 - 986	52.51x+ 0.2 0.88
Whole-crop wheat	20.7 - 99.9	9.1 - 42.4	21.9-121.8	84 - 951	309 - 1277	6.76x+684.0 0.24
High-moisture corn	6.1 - 20.7	1.0 - 14.5	0.2 - 7.6	0 - 107	0-47	17.62x+ 0.3 0.78
Whole-crop maize 6	11.3 - 70.8	25.8 - 48.7	21.0 - 64.0	357 - 789	118 -1263	
Whole-crop maize 7	37.2 - 86.9	10.4 - 28.3	1.1 - 24.1	12 - 64	47 -1305	

**Table 2.** Contents of volatile organic compounds (VOC), especially esters and their correlation, in different maize silages (Weiss and Auerbach, 2012a)

## VOC in grass silages

Extensive literature search yielded one study only by Krizsan et al. (2007), who detected variable concentrations of esters in grass silage, but the mean content never exceeded 30 mg/kg DM. Therefore, the aim of investigations with grass silages (Table 3) was to determine the incidence of VOC in grass silages, particularly ethanol and the ethyl esters of lactic and acetic acids.

Grass silages contained high ethanol and ester concentrations, particularly in those from trials 1 and 2 (Table 4). This may be attributed to the lower storage temperature, which promotes ester formation. Weiss et al. (2009a) observed that maize silages stored at 20 °C had higher ester contents than were detected at 35 °C. The correlation coefficients presented in Table 4 varied widely between 0.35 and 0.85, depending on the trial.

Table 3.	Characterization	of the data	a set from grass	s silages (	(lab-scale	ensiling ex	periments, n= 620	9
T 1	$0.1 \dots DM$		Q4	0.1	1.1.4	4	• • • • • • • • • • • • • • • • • • • •	

Trial	Silage DM	n	Storage length	Silage additive type used in experiment
	(g/kg)		(days)	
1	211 - 438	213	252 - 266	biological, chemical, molasses (Lengyel et al., 2012)
2	191 - 464	209	252 - 266	biological, chemical, molasses (Lengyel, unpublished data)
3	230 - 318	49	81	biological, chemical (Nadeau, unpublished data)
4	318 - 383	12	91	biological (Nadeau, unpublished data)
5	223 - 299	45	90	biological, chemical (Kalzendorf, unpublished data)
6	274 - 357	17	142	biological (Nadeau, unpublished data)
7	283 - 373	12	270	chemical (Nadeau, unpublished data)
8	202 - 219	21	131	biological, chemical (Kalzendorf, unpublished data)
9	223 - 240	21	121	biological, chemical (Kalzendorf, unpublished data)
10	243 - 268	21	139	biological, chemical (Kalzendorf, unpublished data)

Table 4. Fermentation	products, pH and ester concentratio	ons in grass silages (n= 620) (Weiss and Auerbach, 2013)

Trial	pН	Lactic acid	Acetic acid	Ethanol	Total esters*	Corre	elation**
		(g/kg DM)	(g/kg DM)	(g/kg DM)	(mg/kg DM)	r <sub>s</sub>	P value
1	3.7 - 6.7	0 - 99.5	1.5 - 62.8	0.7 - 39.6	0 - 3540	0.35	< 0.001
2	3.6 - 5.8	0 - 89.8	2.0 - 46.7	0 - 35.3	0 - 3995	0.37	< 0.001
3	4.0 - 4.5	60.6 - 117.5	11.1 - 36.5	2.2 - 18.7	0 - 359	0.91	< 0.001
4	3.8 - 4.2	42.7 - 81.8	13.2 - 35.4	6.7 - 12.0	216 - 455	0.52	ns
5	3.8 - 4.5	32.3 - 89.2	14.2 - 76.7	1.6 - 13.1	73 - 378	0.64	< 0.001
6	4.2 - 4.9	30.0 - 116.7	19.7 - 49.3	2.4 - 7.8	0		-
7	4.3 - 4.7	36.6 - 86.5	7.5 - 13.3	2.1 - 19.9	0 - 161	0.65	< 0.05
8	3.8 - 4.2	42.6 - 105.1	8.4 - 19.9	0.9 - 15.1	0 - 378	0.84	< 0.001
9	3.9 - 4.3	49.9 - 110.6	1.6 - 13.9	1.0 - 14.1	0 - 189	0.85	< 0.001
10	4.0 - 4.7	24.0 - 76.2	14.0 - 31.5	3.9 - 12.3	62 - 272	0.85	< 0.001

\*sum of ethyl acetate and ethyl lactate, \*\* correlation between ethanol and total ester concentrations, r<sub>s</sub> Spearman rank correlation coefficient, ns not significant

The pH of the silages had a pronounced effect on ester levels (Table 5). Strong relationships ( $r_s>0.50$ ) were mostly observed when the pH of the silages did not exceed the value of 4.25. This is in line with observations by Hangx et al. (2001) who found ester reactions be stimulated by low pH in the environment.

pH class	n	Total esters*	Ethanol	Corre	lation**
		(mg/kg DM)	(g/kg DM)	r <sub>s</sub>	P value
> 3.50 - 3.75	19	482 - 3995	0 - 35	0.60	< 0.01
> 3.75 - 4.00	126	0 -1856	1 - 40	0.72	< 0.001
> 4.00 - 4.25	176	0 - 920	1 - 25	0.55	< 0.001
> 4.25 - 4.50	131	0 - 762	1 - 18	0.21	< 0.05
> 4.50 - 4.75	81	0 - 550	1 - 24	0.26	< 0.05
> 4.75 - 5.00	42	0 - 384	0 - 38	0.49	< 0.001
> 5.00 - 5.25	26	0 - 255	1 - 37	0.49	< 0.05
> 5.25 - 5.50	10	63 - 211	4 - 28	-0.35	ns
> 5.50	9	0 - 171	3 - 24	0.25	ns

**Table 5.** Relationship between ethanol and ester contents in grass silages (n= 620) as affected by pH (Weiss and Auerbach, 2013)

\*sum of ethyl acetate and ethyl lactate, \*\* correlation between ethanol and total ester concentrations, r<sub>s</sub> Spearman rank correlation coefficient, ns not significant

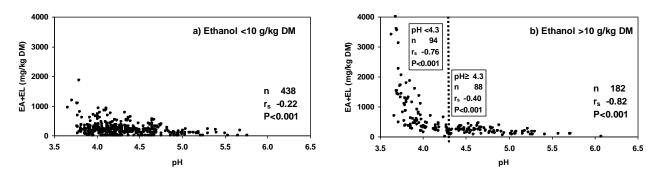
The allocation of the grass silages to different ethanol classes was done as described by Weiss and Auerbach (2012a) and showed clear effects of ethanol content on the relationship between pH and total ester concentration (Table 6). Within each ethanol class, a great variation in ester concentration was observed. The detected ester levels in grass silages were extremely high compared with those reported by Weiss and Auerbach (2012b) for maize silages.

**Table 6.** Relationship between pH and ester content in grass silages (n = 620) as affected by ethanol (Weiss and Auerbach, 2013)

Ethanol class	n	Total esters*	pH	Correlation**		
(g/kg DM)		(mg/kg DM)	range	rs	P value	
≤ 5	257	0 - 1180	3.7 - 5.8	-0.12	ns	
> 5 - 10	181	0 - 1856	3.8 - 6.7	-0.46	< 0.001	
> 10 - 15	100	0 - 1147	3.7 - 5.7	-0.66	< 0.001	
> 15 - 20	39	87 - 3116	3.7 - 5.7	-0.88	< 0.001	
> 20 - 25	21	0 - 3540	3.6 - 6.1	-0.93	< 0.001	
> 25 - 30	12	63 - 3589	3.7 - 5.3	-0.83	< 0.001	
> 30 - 35	5	274 - 2054	3.8 - 4.8	-0.60	ns	
> 35 - 40	5	182 - 3995	3.7 - 5.2	-0.90	< 0.05	

\*sum of ethyl acetate and ethyl lactate, \*\* correlation between pH and total ester concentration, r<sub>s</sub> Spearman rank correlation coefficient, ns not significant

As shown in figure 1a, the correlation between total ester content and pH in grass silages was very weak ( $r_s$ =-0.22; *P*<0.001) up to an ethanol content of 10 g/kg DM, whereas a very strong negative relationship was found ( $r_s$ =-0.82; *P*<0.001) at higher ethanol levels (Figure 1b). The least correlation existed if silage pH exceeded the threshold value of pH 4.3.



**Figure 1.** Total ester concentrations as affected by ethanol class a)  $\leq 10$  g/kg DM, b) > 10 g/kg DM (Weiss and Auerbach, 2013)

In summary it can be stated that grass silages may also contain ethyl esters. However, the relationship between ethanol and ethyl esters in grass silages seems to be not as close as that for maize silages. This can be explained by the fact that the intensity of ester reactions is affected by the pH of the silage and grass silages often have pH values above 4.0. As a

consequence, the correlation coefficients decrease with increasing pH. In conclusion it can be stated that the ester concentrations are strongly correlated with the ethanol concentration and the silage pH.

# VOC in sugar cane silages

In tropical areas ensiled sugarcane are an important forage with more than 400 g/kg DM water-soluble carbohydrates, which are substrates for intensive fermentation (Daniel et al., 2013a). Ethanol is the main fermentation end product in sugar cane silages (Kung and Stanley, 1982). Concerning feed intake Daniel et al. (2013b,c) reported no difference when fresh sugar cane silages was compared with oven-dried material resulting in the loss of volatiles, which was reconstituted with water before feeding. On the one hand ethanol has been correlated with esters and other volatile organic compounds (Weiss et al., 2009a) and, on the other, Kriszan et al. (2007) and Gerlach et al. (2013) observed negative correlations between some VOC and feed intake.

Daniel et al. (2013a) found that the volatile organic compounds comprised up to 22% of the sugarcane dry matter. Table 7 contains data concerning the occurrence of VOC in sugarcane silages, without additives, with sodium benzoate, and with *Lactobacillus buchneri*. In addition to high contents of ethanol, acetic acid, and lactic acid, 1,2-propanediol, ethyl lactate, acetone, 2,3-butanediol, propionic acid, n-butyric acid, ethyl acetate, 2-butanol, methanol, propanol, and isobutyric acid were found (Daniel et al., 2013a).

Common name	Mean	SD <sup>a</sup>	Min.	Max.
	g/kg			
DM oven <sup>b</sup>	28.3	4.0	22.2	34.9
DM corr <sup>c</sup>	31.1	3.1	26.7	36.5
	g/kg DM			
Ethanol	54.2	48.1	5.0	154.5
Acetic acid	32.8	11.5	14.3	53.5
Lactic acid	26.0	20.9	6.5	60.4
	mg/kg DN	M d		
Propane-1,2-diol	1532	2348	<100	12186
Ethyl lactate	697	799	132	2401
Acetone	573	527	<5	2072
Butane-2,3-diol	358	250	<100	905
Propionic acid	284	350	<100	1107
n-Butyric acid	273	369	<100	1383
Ethyl acetate	167	174	<5	597
2-Butanol	135	194	<5	538
Methanol	133	359	<100	1555
Propanol	123	81	<5	290
iso-Butyric acid	<100	55	<100	274

Table 7. Concentrations of fermentation	products in sugarcane silages	(n = 33). Daniel et al. (2013a)

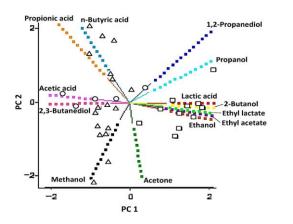
<sup>a</sup> Standard deviation.

<sup>b</sup> Dry matter determined by oven drying (predrying at 55 °C for 72 h followed by drying at 105 °C for 12 h).

<sup>c</sup> Dry matter corrected for volatile compounds (Weissbach, 2009).

<sup>d</sup> iso-valeric acid, n-valeric acid, and caproic acid were below the limit of detection of 100 mg/kg DM; 1-butanol was below the limit of detection of 5 mg/kg DM.

Daniel et al. (2013a) performed a statistical calculation with principal component analysis using PRINCOMP procedure of SAS (Figure 2). They postulated some functional relationships among the fermentation end-products in sugarcane silages. Ethanol was negatively associated with acetic acid and 2,3-butanediol, but positively correlated with lactic acid and esters.



**Figure 2.** Principal components analysis of volatile organic compounds in sugarcane silages. PC 1, first principal component (0.48); PC 2, second principal component (0.15). Silages were untreated ( $\Box$ ), treated with sodium benzoate ( $\Delta$ ) and inoculated with Lactobacillus buchneri (O) (Daniel et al., 2013a)

# EFFECTS OF SILAGE ADDITIVES ON FORMATION OF VOC

#### Whole crop maize silage

The findings that volatile organic compounds (VOC) are frequently found in silages and may detrimentally affect feed intake by dairy cattle (Weiss et al., 2009; Weiss and Auerbach, 2012a) have initiated more research with focus on the use of silage additives to reduce ethanol and ester formation. It is well known that silage additives can alter ethanol contents thereby exerting an effect on ethyl ester production. Weiss and Auerbach (2012b) tested the effects of chemical silage additives on fermentation pattern, production of VOC and aerobic stability of maize silage. They found that treatment had significant effects on all parameters tested, except pH, which was very low in all silages (Table 8). DM losses were highest in acid treatments, whereas a significant reduction in DM loss was found by liquid mixture of sodium benzoate and potassium sorbate (KST). These observations can be explained by differences in ethanol concentrations, whose formation always results in  $CO_2$  release, which escapes from the silo. The most significant reduction in ethanol was caused by the chemical additive KST, whereas acid additives (GSP, PROM) stimulated ethanol production.

Parameter	Treatment				SED	Signi-
	CON	KST <sup>5</sup>	GSP <sup>6</sup>	PROM <sup>7</sup>		ficance
DM loss (%)	6.5 <sup>b</sup>	4.3 <sup>a</sup>	7.5°	7.5°	0.31	***
WSC <sup>1,2</sup>	13.9 <sup>a</sup>	17.3 <sup>ab</sup>	20.5 <sup>b</sup>	19.2 <sup>b</sup>	1.28	**
рН	3.65	3.53	3.63	3.63	0.05	*
NH <sub>3</sub> -N (g/kg total N)	108 <sup>a</sup>	106 <sup>a</sup>	90 <sup>b</sup>	87 <sup>b</sup>	3.28	***
Lactic acid <sup>2</sup>	86.5ª	118.8 <sup>b</sup>	84.6 <sup>a</sup>	78.2 <sup>a</sup>	7.31	**
Acetic acid <sup>2</sup>	22.2 <sup>b</sup>	13.1ª	5.7ª	5.7ª	2.53	***
Propionic acid <sup>2</sup>	0.5ª	0.3ª	1.2 <sup>b</sup>	1.7°	0.08	***
Ethanol <sup>2</sup>	23.2 <sup>b</sup>	6.5 <sup>a</sup>	49.1°	46.5 <sup>c</sup>	1.57	***
1,2-propanediol <sup>2</sup>	0.3 <sup>b</sup>	0.4 <sup>b</sup>	$0^{\mathrm{a}}$	$0^{a}$	0.08	***
Ethyl lactate <sup>3</sup>	398 <sup>b</sup>	166 <sup>a</sup>	617°	612°	39.3	***
Ethyl acetate <sup>3</sup>	223 <sup>b</sup>	123 <sup>a</sup>	189 <sup>b</sup>	184 <sup>b</sup>	16.1	**
Total ethyl esters <sup>3</sup>	621 <sup>b</sup>	289ª	806°	795°	31.3	***
ASTA <sup>4</sup> (days)	5.9ª	12.7 <sup>b</sup>	14.0 <sup>b</sup>	14.0 <sup>b</sup>	1.17	***

**Table 8.** Effects of silage additives on DM losses, fermentation pattern, volatile organic compounds and aerobic stability of whole-crop maize silage (DM 332 g/kg); Weiss and Auerbach (2012b)

<sup>1</sup>water-soluble carbohydrates; <sup>2</sup>g/kg DM; <sup>3</sup>mg/kg DM; <sup>4</sup>aerobic stability; means in columns with unlike superscripts differ significantly at *P*<0.05 (Tukey test); <sup>5</sup>liquid mixture of 21.9% sodium benzoate and 13.2% potassium sorbate, 2 l/t (KOFASIL STABIL, ADDCON EUROPE GmbH, Germany); <sup>6</sup>liquid mixture of 35 % formic and 12% propionic acids, 25.5 % sodium formate, 1.5 % sodium benzoate, 4 l/t (GrasAAT SP, ADDCON NORDIC AS, Norway); <sup>7</sup>liquid mixture of 48.8 % formic acid/ formate, 18.4 % propionic acid/propionate, 6.1 % sodium, 4 l/t (PROMYR XR 680, Perstorp AB, Sweden).

The concentration of ethyl esters in this study were also clearly affected by the concentration of ethanol and the respective organic acids. In general, lactate content was high, and KST increased the concentration of this fermentation acid over that of silages of all other treatments. Hafner et al. (2014, 2015) confirm this results. They postulated that especially potassium sorbate is an effective additive for reducing production of ethanol and ethyl esters in corn silage. Acetic acid concentration was reduced by all used additives. As contents of lactic acid were higher than those of acetic acid, the formation of ethyl lactate was also more pronounced than that of ethyl acetate. KST decreased contents of

ethanol, ethyl lactate (EL) and ethyl acetate (EA). GSP and PROM stimulated the production of ethanol and EL, whereas no effect was found on EA. By using all experimental data from all individual silages of all treatments (n=12), a very high correlation was found between ethanol and total ester concentrations ( $R^2$ =0.985). Elevated ethanol production in anaerobic conditions can be attributed to the activity of yeasts, which may have been present at high numbers during the initial stages of fermentation but died off during later storage.

Further investigations of Weiss et al. (2015b, 2016) with maize confirm that silage additives containing sodium benzoate, calcium propionate and potassium sorbate were superior to other treatments regarding suppression of ethanol and ester formation as well as improvement of aerobic stability, with and without air ingress.

#### Sorghum silages

A study of Auerbach and Weiss (2012) with sorghum silages aimed at testing the effects of different silage additives on dry matter (DM) losses, fermentation pattern, VOC production and aerobic stability of this type of silages. Sorghum was chosen as silage type because it represents an important forage source for ruminants in semi-arid regions, and its production often bears the risk of excessive ethanol fermentation so that high concentrations of VOC are to be expected. Lactic and acetic acids were affected by variety and treatment, and an interaction was determined between the two factors for lactic acid (Table 9). Ethanol was reduced by *Lactobacillus buchneri* (LB) at all inoculation rates, and the lowest levels were consistently found if a mixture of sodium benzoate and potassium sorbate (BS) were used. The use of *Lactobacillus plantarum* (LP) alone or in combination with LB1 did not affect ethanol production when compared with control silages. The concentrations of reaction products of ethanol and organic acids – ethyl lactate and ethyl acetate – were affected by variety and treatment. Application of BS and LB (regardless of inoculation rate) caused the lowest ester contents, and no differences between CON, LP and LP+LB1 were found.

Treatment		Lactic acid		Acet	Acetic acid		Ethanol		esters <sup>1)</sup>
		(g /kg	, DM)	(g /k	g DM)	(g /kg	DM)	(mg/k	(g DM)
Variety		Goliath	Maya	Goliath	Maya	Goliath	Maya	Goliath	Maya
CON <sup>3</sup>		92.0 <sup>bA</sup>	40.3 <sup>cB</sup>	24.9 <sup>abA</sup>	27.3 <sup>bcA</sup>	31.7 <sup>cA</sup>	34.2 <sup>cdA</sup>	381 <sup>dA</sup>	587 <sup>dB</sup>
$LP^4$		90.2 <sup>bA</sup>	38.3 <sup>bcB</sup>	20.2 <sup>aA</sup>	22.0 <sup>aA</sup>	34.9 <sup>cA</sup>	28.8 <sup>cA</sup>	506 <sup>dA</sup>	586 <sup>cdA</sup>
$LB^{5}1$		24.3 <sup>aA</sup>	22.8 <sup>bA</sup>	$47.4^{bcB}$	$37.0^{abdA}$	19.9 <sup>bA</sup>	19.5 <sup>bA</sup>	251 <sup>cA</sup>	414 <sup>bcB</sup>
$LB^{5}2$		22.3 <sup>aA</sup>	24.6 <sup>bA</sup>	51.6 <sup>cB</sup>	$45.5^{cdA}$	18.9 <sup>bA</sup>	18.3 <sup>bA</sup>	$245^{bcA}$	365 <sup>abcB</sup>
LB <sup>5</sup> 3		19.9 <sup>aA</sup>	17.8 <sup>aA</sup>	53.5 <sup>cA</sup>	51.8 <sup>dA</sup>	17.3 <sup>bA</sup>	19.6 <sup>bA</sup>	$214^{abA}$	299 <sup>aB</sup>
$LP+LB1^{6}$		103.6 <sup>bA</sup>	$26.9^{abcB}$	$24.3^{abA}$	$22.8^{abA}$	34.2 <sup>cA</sup>	39.5 <sup>dA</sup>	$460^{dA}$	$559^{dB}$
$\mathbf{BS}^7$		95.4 <sup>bA</sup>	24.4 <sup>bB</sup>	$27.6^{abA}$	27.9 <sup>bA</sup>	6.9 <sup>aA</sup>	$7.7^{aA}$	131 <sup>aA</sup>	$239^{abA}$
SEM		8.18	1.83	3.05	2.46	2.23	2.31	31.1	33.2
Significance level <sup>2</sup>									<u> </u>
Variety		**	**		*	n	S	*	**
Treatment		**	**	;	***	**	*	*	**
Variety	х	**	**		ns	n	S	1	ns
Treatment									

<sup>1</sup>sum of ethyl acetate and ethyl lactate; <sup>2</sup>means in columns with unlike superscripts and means within rows bearing unlike capital superscripts differ significantly at  $P \le 0.05$  (Tukey test); <sup>3</sup>Control; <sup>4</sup>L. *plantarum*, 1x10<sup>5</sup> cfu/g forage; <sup>5</sup>L.buchneri (1x10<sup>5</sup> cfu/g forage; 2 (2.5x10<sup>5</sup> cfu/g forage), (5x10<sup>5</sup> cfu/g forage); <sup>6</sup>L. *plantarum* + L. *buchneri* (2x10<sup>5</sup> cfu/g forage); <sup>7</sup>500 g/t sodium benzoate+300 g/t potassium sorbate (applied in 2 l/t aqueous solution)

#### Grass silages

However, the knowledge of the formation of VOC in grass silages and the effects of additives thereon is also still very limited. Weiss and Auerbach (2015) carried out a laboratory ensiling experiment with fourth-cut natural grassland, wilted overnight to 26,8% DM. Forages received the treatment with 21 commercial additives (Table10) which were obtained from the German marketplace and used according to the instructions of the manufacturers.

Grass silages were well fermented as reflected by low pH (Table 10) and no butyric acid was found (data not given). The production of lactic acid was stimulated by some additives of the types Ho, HoHe and HoCh whereas the pure He inoculant as well as two chemicals reduced it. The treatment with homofermentative LAB, either applied alone or in combination with antimycotic chemicals, always resulted in lower acetate levels. The lowest ethanol and ethyl ester contents were detected in silages that had received chemical additives. There was a strong positive linear correlation between these two parameters ( $R^2$ =0.72, P<0.001). The production of 1-propanol was highest in silages treated with the heterofermentative inoculant.

# Legume silages

Investigations concerning effect of wilting and silage additives on silage quality of lucerne, red clover and grass mixtures (Weiss and Kalzendorf, 2016) demonstrate the occurrence of VOC in legume silages. The DM content and silage additives affect the concentrations of alcohols, acids and esters. However, yeast counts were high and increased during wilting period. In accordance to the fact that under anaerobic conditions yeasts are responsible for ethanol formation, the ethanol content in silages without any additives was between 4.8 and 10.9 g kg<sup>-1</sup> DM with a strong negative correlation to DM content ( $R^2$ = 0.81) and positive correlation to ester content ( $R^2$ = 0.65). Therefore elevated levels of alcohols and esters occur in silages with low DM. The total esters ranged between 124 und 197 mg kg<sup>-1</sup> DM in untreated silages and consisted of only ethyl lactate. These ester contents are comparable with contents in grass silage (Weiß and Auerbach, 2013) considering the pH level between 4.0 and 6.3. Silage additives with LAB did mainly not affect the contents of ethanol, the same applies for the contents of esters. The additive salts containing benzoate, nitrite and hexamine strongly reduced the ethanol and ester contents. According to Woolford (1975) these substances are able to inhibited yeasts and possibly heterofermentative LAB which also produce ethanol.

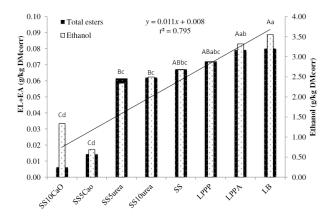
Treat- ment	рН	Lactic acid <sup>1</sup>	Acetic acid <sup>1</sup>	Etha- nol <sup>1</sup>	EE <sup>2,3</sup>	Pro- panol <sup>3</sup>	Ace- tone <sup>3</sup>	Me- thanol <sup>3</sup>	2-Bu- tanol <sup>3</sup>	$AS^4$
Con <sup>5</sup>	4.0	82.0	14.1	10.2	344	236	0	697	205	7.4
Ho <sup>6</sup>	3.9*	85.1	7.9*	8.7	301	0*	119*	789	224	3.3*
Ho <sup>6</sup>	3.9*	96.0*	11.0 <sup>§</sup>	9.0	316	23*	131*	844 <sup>§</sup>	222	7.0
Ho <sup>6</sup>	3.8*	87.2	7.9*	7.5#	258	0*	109*	796	212	2.3*
Ho <sup>6</sup>	3.8*	90.9 <sup>§</sup>	8.5*	8.2 <sup>§</sup>	284	$0^{*}$	108*	845 <sup>§</sup>	128 <sup>§</sup>	$1.8^{*}$
Ho <sup>6</sup>	3.8*	91.2 <sup>§</sup>	7.6*	7.5*	309	$0^{*}$	99*	686	152	$2.7^{*}$
Ho <sup>6</sup>	3.8*	88.6	$8.9^{*}$	7.7#	261	$0^*$	89*	694	160	4.3 <sup>§</sup>
He <sup>7</sup>	$4.1^{*}$	61.1*	$22.8^{*}$	13.4*	353	$1080^{*}$	$100^{*}$	817	195	8.8
HoHe <sup>8</sup>	3.9#	85.2	14.0	10.5	384	44*	126*	828	208	6.3
HoHe <sup>8</sup>	3.9*	79.6	11.3	8.7	342	100 <sup>§</sup>	$76^*$	816	197	7.2
HoHe <sup>8</sup>	3.9*	86.8	11.3	8.8	411	72#	111*	878 <sup>#</sup>	130 <sup>§</sup>	6.4
HoHe <sup>8</sup>	3.9*	96.2 <sup>*</sup>	10.2 <sup>§</sup>	7.8#	329	$0^{*}$	$108^{*}$	853 <sup>§</sup>	214	5.4
HoHe <sup>8</sup>	3.9*	89.0	12.3	8.8	327	78#	<b>9</b> 9*	786	196	6.7
HoCh <sup>9</sup>	3.9*	93.2 <sup>§</sup>	10.3 <sup>§</sup>	9.8	300	$0^{*}$	18	641	155	7.4
HoCh <sup>9</sup>	3.9*	81.7	10.2 <sup>§</sup>	8.9	292	$0^{*}$	24	660	187	8.1
HoCh <sup>9</sup>	3.9*	85.0	9.5#	8.6	272	$0^{*}$	69 <sup>*</sup>	692	179	6.8
HoCh <sup>9</sup>	3.9*	87.0	$7.7^{*}$	$7.5^{*}$	208	$0^{*}$	51*	598	202	7.3
HoCh <sup>9</sup>	3.9*	84.2	$8.6^*$	$7.5^{*}$	265	$0^{*}$	$97^*$	729	241	10.9 <sup>§</sup>
$Ch^{10}$	4.0#	72.4 <sup>§</sup>	16.5	$2.3^{*}$	$80^*$	499*	0	809	161	$15.0^{*}$
$Ch^{11}$	4.0	77.9	15.6	$4.2^{*}$	143*	165*	0	611	153	$15.0^{*}$
$Ch^{12}$	4.0	$61.0^{*}$	11.6	$4.5^{*}$	105*	$0^{*}$	0	583	164	$14.1^{*}$
$Ch^{12}$	3.9*	78.1	11.5	$3.2^{*}$	61*	$0^{*}$	0	630	209	$15.0^{*}$

**Table 10.** Effects of additives on fermentation pattern, volatile organic compounds and aerobic stability of grass silage stored for 72 days (Weiss and Auerbach, 2015)

Means of each additive treatment in columns bearing unlike superscripts differ compared with untreated; \*P < 0.001,  ${}^{#}P < 0.01$ ,  ${}^{§}P < 0.05$ ;  ${}^{1}g/kg$  DM;  ${}^{2}ethyl$  lactate+ethyl acetate;  ${}^{3}mg/kg$  DM;  ${}^{4}aerobic$  stability, days;  ${}^{5}untreated$ ;  ${}^{6}homofermentative$  LAB;  ${}^{7}heterofermentative$  LAB;  ${}^{8}combination$  of homo- and heterofermentative LAB;  ${}^{9}combination$  of homofermentative LAB and antimycotic chemical(s);  ${}^{10}nitrite$ , hexamine, sorbate;  ${}^{11}nitrite$ , benzoate, sorbate;  ${}^{12}buffered$  formic and propionic acid blends.

#### Sugar cane silages

The study of Cardoso et al. (2016), to evaluate the chemical composition, fermentation pattern an microorganism of sugar cane without and with chemical additives and inoculants (Figure 3) confirmed that a correlation between ethanol and ethyl esters is strong. In sugarcane silages with CaO this chemical additive inhibited ethanol and ester formation.



**Figure 3.** Correlation between ethyl acetate and ethyl lactate (EL+EA) and ethanol contents in silage, averages in g/kg of DMcorr. Sugarcane silage without inoculant (SS), SS with *Lactobacillus buchneri* (LB), SS with *Lactobacillus plantarum* and *Pediococcus pentosaceus* (LPPP), SS with *Lactobacillus plantarum* and *Propionibacterium acidipropionici* (LPPA), SS with 5 g/kg CaO (SS5CaO), SS with 10 g/kg CaO (SS10CaO), SS with 5 g/kg urea (SS5urea), and SS with 10 g/kg urea (SS10urea) (Cardoso et al., 2016)

#### White lupin-wheat silages

Laboratory ensiling trials with white lupin-wheat silages (König et al., 2015) demonstrates the occurrence of volatile compounds, also esters, in this special ensiling material. The authors found, that increased proportion of lupin increased concentration of VOC and confirm the safe effect of chemical additives due to their influence on fermentation pattern.

#### Summary

Results from ensiling experiments on the effects of silage additives on ester formation in different ensiling materials clearly indicated that chemical products containing active ingredients with specific antifungal effects can significantly reduce ester concentration. Salts of sorbic, benzoic or propionic acids or mixtures are effective treatment for reducing VOC production.

Buffered formic acid-containing products, which were always applied at 4 l/t, stimulated it due to an increase in ethanol content (Weiss and Auerbach, 2012b; Auerbach and Weiss, 2012).

# VOLATILE ORGANIC COMPOUNDS (VOC) IN MAIZE SILAGES AT GERMAN DAIRY FARMS

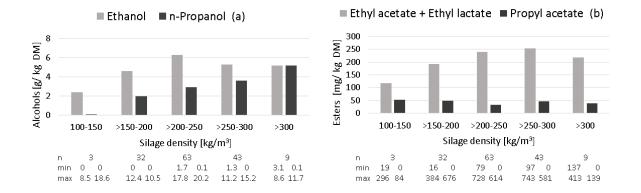
A survey has been carried out to investigate the incidence of VOC in maize silages from German dairy farms and to monitor the concentrations of ethanol, n-propanol and the corresponding esters ethyl acetate, ethyl lactate and propyl acetate, depending on the sampling site in the silo and the compaction of silages (Weiss et al., 2015a).

The survey included a detailed examination of silages stored in bunker silos on 52 dairy farms. Most silages were produced without silage additives (n=43), whereas 9 farms had used biological additives. The highest contents of fermentation acids (acetic, lactic and propionic acids) and alcohols (methanol, ethanol, n-propanol) in maize silages were found in the bottom, highly compacted core and to some extent in middle core samples taken from bunker silos (Table 11), which supports empirical observations by Weiss et al. (2009a). Ethanol was detected at up to 17.8 g/kg DM and the highest n-propanol level was 20.2 g/kg DM (Figure 4a). In agreement with data by Weiss et al. (2009a), ethyl lactate (EL) concentrations in maize silages were higher than the levels of ethyl acetate (EA) and propyl acetate (PA) (Figure 4b). The contents of total esters (up to 925 mg/kg DM) were higher than in silages from laboratory ensiling trials (Weiss et al., 2009a). With increasing compaction, the concentrations of n-propanol and ethanol as well as those of the ethyl esters EA and EL (Figure 2) and aerobic stability (R<sup>2</sup> = 0.920, P < 0.001) increased (data not shown). This may be explained by the usually lower pH in the bottom, more compacted and less air-affected zones in farm silos. Esterification processes were shown to be stimulated by low pH (Weiss and Auerbach, 2013).

Parameter	$BC^4$		$MC^5$		TE <sup>6</sup>		P-Value
DM (%)	34.1	$\pm 0.5$	33.4	$\pm 0.4$	34.0	$\pm 0.5$	0.950
pH	3.85 <sup>a,b</sup>	$\pm 0.18$	3.83 <sup>a</sup>	$\pm 0.02$	3.89 <sup>b</sup>	$\pm 0.03$	0.036
Lactic acid	49.3 <sup>b</sup>	$\pm 2.6$	51.4 <sup>b</sup>	$\pm 1.9$	41.8 <sup>a</sup>	$\pm 2.1$	0.001
Acetic acid	23.0 <sup>b</sup>	$\pm 1.2$	19.5 <sup>a</sup>	$\pm 0.9$	19.6 <sup>a</sup>	$\pm 1.0$	0.009
Prop. acid <sup>1</sup>	$0.8^{b}$	$\pm 0.2$	0.4ª	$\pm 0.1$	$0.6^{a,b}$	$\pm 0.1$	0.028
Methanol	0.3 <sup>b</sup>	$\pm 0.0$	0.2ª	$\pm 0.0$	0.3 <sup>b</sup>	$\pm 0.0$	0.008
Ethanol	6.9 <sup>b</sup>	$\pm 0.5$	5.9 <sup>a,b</sup>	$\pm 0.4$	5.1ª	$\pm 0.4$	0.001
2-Butanol	0.2 <sup>b</sup>	$\pm 0.1$	$0.2^{a,b}$	$\pm 0.1$	0.1ª	$\pm 0.0$	0.015
n-Propanol	4.4 <sup>b</sup>	$\pm 0.7$	2.7ª	$\pm 0.5$	2.1ª	$\pm 0.4$	0.001
Ethyl acetate <sup>2</sup>	51 <sup>a,b</sup>	$\pm 4$	40 <sup>a</sup>	$\pm 3$	59 <sup>b</sup>	± 5	0.007
Ethyl lactate <sup>2</sup>	210 <sup>b</sup>	$\pm 17$	176 <sup>a,b</sup>	±15	150 <sup>a</sup>	$\pm 14$	0.003
Propyl acetate <sup>2</sup>	44	$\pm 17$	30	± 7	46	$\pm 16$	0.626
Total esters <sup>2</sup>	305	$\pm 24$	246	$\pm 18$	255	$\pm 24$	0.080
Ammonia	1.3 <sup>b</sup>	$\pm 0.0$	1.1 <sup>a</sup>	$\pm 0$	1.1 <sup>a</sup>	$\pm 0.0$	<0.001
WSC <sup>3</sup>	8.2ª	$\pm 0.7$	10.5 <sup>b</sup>	$\pm 1.0$	9.9ª	$\pm 0.7$	0.001
AS (d)	7.2	$\pm 4.8$	6.6	$\pm 4.1$	6.3	$\pm 4.2$	0.2613
Yeasts (log cfu g <sup>-1</sup> FM)	4.7 <sup>a</sup>	$\pm 4.6$	6.2 <sup>b</sup>	± 5.9	6.1 <sup>b</sup>	$\pm 5.8$	<0.001
Compaction (kg/m <sup>3</sup> )	256	$\pm 5.6$	226	$\pm 5.8$	217	± 5.9	<0.001

**Table 11.** Fermentation characteristics of maize silages on 52 German dairy farms in different sections of bunker silos (mean  $\pm$  SEM, g/kg DM unless otherwise stated) (Weiss et al., 2015a)

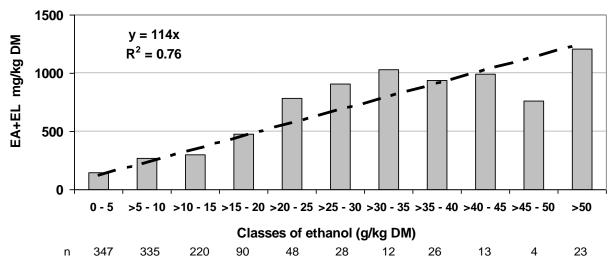
<sup>1</sup> Propionic acid; <sup>2</sup>mg/kg DM, <sup>3</sup>water-soluble carbohydrates; <sup>4</sup>Bottom core; <sup>5</sup> Middle core; <sup>6</sup>Top edge; means in rows with unlike superscripts differ at P < 0.05 (Tukey's test).



**Figure 4.** Average concentrations of ethanol and n-propanol (a) and the esters ethyl acetate + ethyl lactate and propyl acetate (b) as affected by silage density (Weiss et al., 2015a).

## ESTIMATION OF ESTER CONTENT

Based on a total of 1148 data sets from grass silages (Weiss and Auerbach, 2013) as well as from silages from wholecrop maize, whole-crop wheat, sorghum, high- moisture corn (Weiss and Auerbach, 2012), a regression model was used to describe the relationship between total ester and ethanol concentrations, which is valid for all silage types. As shown in figure 5, each incremental increase in ethanol content by 5 g/kg DM resulted in increased total ester concentration by 114 mg/kg DM ( $R^2$ = 0.76). Therefore, the following equation can be applied to calculate ester concentration in silages based on their ethanol content: predicted total ester concentration [mg/kg DM] = ethanol concentration [g/kg DM] x 114/5. The use of this predictive model offers the possibility to avoid laborious and expensive chemical ester analyses.



**Figure 5.** Average total content of esters (ethyl acetate and ethyl lactate) in classes of ethanol in silages from wholecrop maize, whole-crop wheat, sorghum, high- moisture corn and grass (n=1148) (Weiss and Auerbach, 2013)

# CONCLUSION

With regard to the current body of evidence on VOC formation in silages and their potential negative impact on feed intake dairy cows and goat it can be stated that the reduction in ethanol production may lead to lower levels of ethyl esters. This is substantiated by data from ensiling experiments on the effects of different silage additives on ester formation in maize, grass, legume and sorghum silages. Only chemical products containing active ingredients with specific antifungal properties (sodium benzoate, potassium sorbate) consistently and significantly reduced ethyl ester concentrations.

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# Fibre and Protein Quality of Silages and their Effects on Ruminant Performance

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# **INTRODUCTION**

Forage is the main feed for ruminant animals as their microbes in the rumen can utilize cell-wall components, which constitute a significant part of forage dry matter (DM). In forage cell walls, there are the structural carbohydrates cellulose and hemicellulose, which are linked to lignin, which is classified as a polymer of hydroxycinnamylalcohols (Jung and Allen, 1995). The degree of these cross-linkages and the amount of lignification cause variations in microbial degradation of cellulose and hemicellulose as lignin is indigestible (Jung and Allen, 1995). Some of the forage protein is present in the cell walls, of which a small part is linked to the cellulose-lignin matrix, and cannot be utilized by the ruminant animal (Sniffen et al., 1992). Additionally, pectin is part of the cell walls with a larger amount in legumes than in grasses. In nutrition, the cell-wall components are classified as fibre. Fibre is analysed as neutral detergent fibre (NDF), which contains hemicellulose, cellulose, lignin and cell-wall bound protein. However, if sodium sulphite is added to neutral detergent (ND) solution, much of the protein is removed (Mertens, 2002). Neither is pectin found in the NDF as pectin is solubilized in the ND solution (Jung and Allen, 1995). Cellulose, lignin and protein bound to these compounds constitute the acid detergent fibre (ADF), which is the remaining cell wall after treatment with an acid detergent (AD) solution (Van Soest et al., 1991). The lignin can be determined as the residue after treatment with 72% sulphuric acid and defined as acid detergent lignin (ADL; Van Soest et al., 1991).

Forages differ in nutrient composition and perennial forages can be classified in legumes and grasses. Grasses further can be divided into cool-season and warm-season grasses of which the cool-season grasses have lower NDF concentration and higher organic matter (OM) digestibility than the latter ones (Harrison et al., 2003). This paper focuses on cool-season grasses as they are the predominant grasses grown in north Europe, and their comparisons to legumes. At similar maturity stages, legumes contain less sugar and more protein than grasses but the crude protein (CP) concentration of grasses fertilized with high rates of nitrogen can be nearly as high as for legumes (Buxton and O'Kiely, 2003). As grasses and legumes mature, the CP and sugar concentrations decrease while the NDF concentration increases and digestibility of NDF decreases, resulting in a decreased OM digestibility (Kuoppala et al., 2009; Nadeau et al., 2000), which can affect intake and ruminant performance (Nadeau et al., 2015a; Alstrup et al., 2016). Whole crop cereals and maize, which are annual forages, contain moderate-to-high amounts of digestible starch from the grain at later stages of maturity. As whole crop maize and cereals develop, the awn-to-stalk ratio increases, resulting in increasing starch content while the NDF concentration decreases and the NDF of the stalks becomes less digestible, resulting in no or small changes in whole plant OM digestibility and metabolizable energy concentration. These forages have low CP concentrations (Hetta et al., 2012). Nutrient composition of forages plays a role in the ensiling ability of the forage. Legumes, such as lucerne, have low sugar concentrations and high protein concentrations and buffering capacity, resulting in a slow acidification rate to reach a desirable pH to decrease proteolytic activities by enterobacteria and clostridia (Buxton and O'Kiely, 2003). Grasses contain more sugar and have a lower buffering capacity than legumes enhancing the growth of lactic acid bacteria to produce lactic acid and acetic acid, thereby decreasing pH faster to a level that decreases the risks for secondary fermentation (Pahlow et al., 2003). Whole crop cereals and maize have relatively thick stalks, which require to be chopped finely and packed thoroughly in the silo to decrease the risks for air infiltration during storage. Air infiltration enhances yeast growth in the silage, which increases the risks for aerobic instability upon opening of the silo and during feed-out (Weiss et al., 2016). The temperature rise of the silage indicates nutrient losses, which can cause reduced energy intake for the ruminant animal (Muck et al., 2003).

Depending on differences in nutrient composition and fermentation characteristics, various forages complement each other in rations for ruminants, where the main goal is to optimize protein and energy intakes, while providing sufficient amounts of structural fibre for an optimal rumen function to maintain a high feed efficiency of the ruminant animal. Quality of the protein and of the fibre fed is of major importance for a healthy rumen environment resulting in high feed efficiency of the ruminant animal. Therefore, this paper focuses on forage fibre content, mastication, degradation and utilization and on forage protein solubility, degradation and utilisation as affected by forage and animal characteristics.

## STRUCTURAL FIBRE OF FORAGES

The content of NDF is the chemical fraction in forages, which has the largest effect on digestibility (Huhtanen et al., 2006), energy value, intake (Mertens, 2007), chewing activity (Nørgaard et al., 2010), rumen function (Mertens, 1997), the distribution of particle size in rumen content (Schulze et al. 2014b), and faeces characteristics (Jalali et al., 2015). The content of NDF depends on stage of maturity at harvest and type of forage. The legumes, such as lucerne (*Medicago sativa* L.) and red clover (*Trifolium* pratense L.), have in general lower NDF contents than grasses at similar stages of maturity (Table 1). The dietary characteristics of the NDF fraction includes the proportions of ADF, ADL,

indigestible NDF (iNDF), digestible NDF (DNDF) and the rate of degradation of the DNDF fraction ( $k_d_DNDF$ ). The cell walls generally are much more lignified in legumes compared to grasses. The lignification in term of the ADL/NDF ratio generally increases due to advanced maturity at delayed harvest, which leads to decreased digestibility of NDF and DM of the forage. The iNDF content is closely linked to the lignin content and the iNDF value is 2.5-3 times the ADL content in grass and lucerne. However, this relationship is affected by forage type and by stage of maturity at harvest (Huhtanen et al., 2006). The proportion of DNDF in NDF of grasses ranges typically between 70 and 90% of NDF, whereas the proportions of NDF in DM and of DNDF in NDF are much lower in lucerne of similar maturity stage and OM digestibility as illustrated by the values in Table 1. However, the low proportion of digestible NDF in lucerne is partly compensated by a high rate of degradation of DNDF ( $k_d_DNDF$ ) of lucerne, which makes the digestibility of lucerne silage less limiting to increased intake compared to grasses. Red clover has a relatively high proportion of digestible NDF at early harvest compared to lucerne. White clover (*Trifolium repens* L.) has generally much higher content of cell solubles and lower NDF content compared to grasses, lucerne and red clover, which makes white clover highly digestible. For whole-crop maize, there is a strong negative correlation between the concentrations of NDF and starch (r = -0.83) as there is an increased proportion of the starch-rich cob while the proportion of the fibre-rich stalk is decreasing (Nadeau et al., 2010).

**Table 1** Effect of harvest time (early, medium, late) on fibre characteristics and *in vivo* organic matter digestibility (OMD) in different forages.

	Grass silage <sup>1</sup>		Lucern	Lucerne silage <sup>2</sup>		White clover silage <sup>3</sup>		er silage <sup>3</sup>	
	Early	Medium	Late	Early	Late	Early	Late	Early	Late
NDF, % DM	44.9	57.8	63.4	37.9	44.6	19.3	28.7	36.0	45.0
ADF, % DM	26.7	35.7	38.3	30.1	35.1	18.7	27.9	24.7	39.8
ADL, % NDF	4.2	6.7	8.2	18	19	13	18	10	16
iNDF <sup>4</sup> , % NDF	7.7	16	27	43	62	13	22	17	50
DNDF <sup>5</sup> , % NDF	92	84	73	57	48	87	78	83	50
k <sub>d</sub> _DNDF <sup>6</sup> , %/h	6.4	4.7	4.4	$6.4^{*}$	$5.6^{*}$	$6.9^{*}$	$5.9^{*}$	$4.5^{*}$	$4.0^{*}$
OMD, %	80	73	64	$70^*$	59*	$78^*$	75*	$72^{*}$	$66^*$

<sup>1</sup>Jalali et al. 2012a

<sup>2</sup> Kornfelt et al. 2013b, cv. Pondus

<sup>3</sup> Kornfelt et al. 2013a, white clover cv. Klondike and red clover cv. Rajah

<sup>4</sup> Indigestible NDF in situ

<sup>5</sup> Digestible NDF=NDF-iNDF

<sup>6</sup>Rate of degradation of DNDF *in situ* 

\*Kornfelt, 2012

## **Chewing activity**

The daily time spent eating and ruminating, which sums to total chewing time, is closely related to the intake of forage NDF (NDF<sub>f</sub>; Table 2). The time spent eating and ruminating per kg NDF<sub>f</sub> decreases at increasing BW from lamb, sheep, growing cattle to mature dairy cows and beef cows, which are considered to spend 50 minutes eating and 100 minutes ruminating per kg NDF intake according to the Nordic Chewing Index system (Nørgaard et al., 2011). However, the time spent ruminating per kg NDF<sub>f</sub> decreases at increasing intake of NDF<sub>f</sub> /BW (Nørgaard et al., 2010; Schulze et al. 2015), whereas the ratio between eating time and ruminating time increases at increasing intake of NDF<sub>f</sub> and at decreasing iNDF/NDF ratio of the forage (Schulze et al., 2014a). Time spent eating per kg DM intake and per kg NDF<sub>f</sub> intake appears to increase at increasing feeding level up to *ad libitum* intake (Schulze et al. 2014a). In addition, increasing lignification of NDF and increasing iNDF/NDF ratio in forages is considered to increase rumination time per kg NDF<sub>f</sub> according to the Nordic Chewing index system (Nørgaard et al., 2010; Nørgaard et al., 2011), and this effect has been supported by the observation by Schulze et al. (2015). When using a ruminating monitoring system (RuminAct-Milkline, Gariga di Podenzano, Italy) on lactating dairy cows, variation in daily rumination time was to a lesser extent explained by variation in intakes of dietary fractions, such as fibre, starch and sugar, than to the individual variations between cows. Furthermore, rumination time in minutes per kg of DM intake was negatively related to milk fat content (Byskov et al., 2015).

### **Faeces characteristics**

Faeces characteristics in cattle, sheep and goats are strongly affected by the forage intake. Plant species, stage of maturity at harvest, NDF content, lignification of NDF, digestibility and physical form of forages affect faeces characteristics. Faeces have been characterized by the content of DM, the content of particle dry matter in DM (PDM), particle size and distribution of particle size dimensions in the PDM fraction (Table 2). The PDM values of faeces from cattle and sheep generally increase at increasing stage of maturity at harvest, increasing ADL/NDF ratio of forage (Jalali et al., 2015), increasing NDF content and at decreasing apparent digestibility of NDF (Schulze et al., 2014a,b; Schulze et al., 2015; Figure 1).

The dimension size of the faeces particles in the PDM fraction has been characterized by sorting of PDM matter in different sieving fractions, and by density and accumulated distribution functions of particle length and width values

(Jalali et al., 2012a,b). Figure 2 shows the left skewed density distribution of particle length in faeces PDM from small ruminants fed either artificially dried grass hay or grass seed straw (Jalali et al., 2012b). The length and width distributions of particles in faeces from ruminants are characterized by many short and thin particles and a few long and wide particles. The most frequent (mode) width values of faeces particles from ruminating animals fed grasses and legumes are found to range between 0.07 and 0.3 mm, and with increasing mode value due to delayed harvest and increased lignification of NDF (Table 2). Likewise, Schulze et al. (2015) observed a decreasing proportion of small particles < 0.1 mm due to delayed harvest of grass-clover forage conserved as silage or hay. The density distribution for width values of faeces particles from cattle fed forage legumes, such as lucerne and clovers show two peaks (Kornfelt et al., 2013a,b). The second peak value indicates much wider faeces particles compared to faeces particles from cattle or sheep fed grasses, which might be associated with much higher lignification of legumes compared to grasses.

Table 2 Effect of harvest time (ea	y, medium, la	late) of different	forage types or	1 chewing activity and faece	s
characteristics in ruminants.					

	]	Pregnant ewes			Dry cow				
		Ad libitum				80% of	ad libitun	n	
		Grass silage <sup>1</sup>			Lucerne silage <sup>2</sup>		White clover silage <sup>3</sup>		clover age <sup>3</sup>
	Early	Medium	Late	Early	Late	Early	Late	Early	Late
Intake, g NDF/kg BW	11	13	13	11	11	3.1 <sup>x</sup>	4.8 <sup>y</sup>	5.4 <sup>y</sup>	7.7 <sup>z</sup>
Chewing time									
Eating,min/kg NDF	325	359	357	108 <sup>x</sup>	117 <sup>y</sup>	165	139	116	107
Rumination, min/kg NDF	343	323	360	96	106	135	127	125	135
Total, min/kg NDF	668	682	717	201 <sup>x</sup>	224 <sup>y</sup>	300	266	241	243
Faeces characteristics									
Dry matter (DM), %	27 <sup>x</sup>	31 <sup>y</sup>	35 <sup>z</sup>						
Particle DM , % of DM	41 <sup>x</sup>	60 <sup>y</sup>	68 <sup>z</sup>	62	71				
Sieving particle DM									
Mean particle size, mm	0.18 <sup>x</sup>	0.20 <sup>x</sup>	0.23 <sup>y</sup>			0.23 <sup>x</sup>	0.21 <sup>xy</sup>	0.19 <sup>y</sup>	0.26 <sup>z</sup>
LP > 1 mm, %	2.9 <sup>x</sup>	2.4 <sup>xy</sup>	1.8 <sup>y</sup>			8	4	6	4
Image analysis of particles									
Mode particle length <sup>4</sup> , mm	0.37	0.31	0.34	0.35	0.38	0.19 <sup>x</sup>	0.25 <sup>x</sup>	0.27 <sup>x</sup>	0.44 <sup>y</sup>
Mode particle width, mm	0.071	0.064	0.074	0.10	0.10	0.06	0.07	0.06	0.08
Mean particle length, mm	0.82	0.78	0.88	1.16	1.18	0.91 <sup>x</sup>	0.94 <sup>x</sup>	0.81 <sup>y</sup>	1.11 <sup>z</sup>
Mean particle width, mm	0.11 <sup>x</sup>	0.13 <sup>x</sup>	0.16 <sup>y</sup>	0.25 <sup>x</sup>	0.27 <sup>y</sup>	0.21 <sup>x</sup>	0.20 <sup>x</sup>	0.18 <sup>y</sup>	0.25 <sup>z</sup>
95 percentile length <sup>5</sup> , mm	3.8	3.2	3.5	4.3	4.4	4.6	4.3	4.2	4.6
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<sup>1</sup> Jalali et al. 2012a

<sup>2</sup> Kornfelt et al. 2013b

<sup>3</sup>Kornfelt et al. 2013a

<sup>4</sup> Most frequent particle length, see Figure 5

<sup>5</sup> Fractile value, which defines the minium length of the 5% longest particles

<sup>x, y, z</sup>Values within the same row and experiment without common superscript differ (P < 0.05)

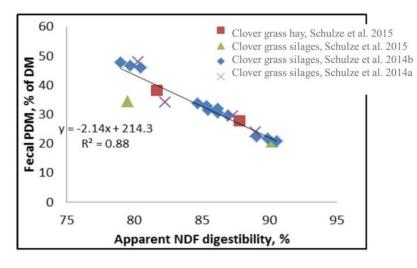
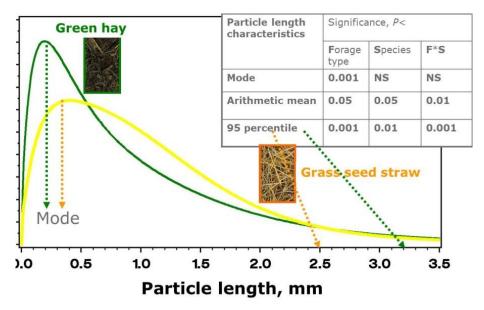


Figure 1 Relationship between the content of faecal particle dry matter (PDM) and the apparent

NDF digestibility in heifers fed highly digestible grass-clover silages or hay (Schulze, 2014).

The large particles (LP) in faeces have been defined as the particles larger than the critical particle size (CPS) of 1.18 mm or as particles longer than the critical particle length (CPL) value of 5 mm in cattle and about 3-4 mm in small ruminants. The CPS and the CPL have been defined as the 95% percentile value for the accumulated distribution of particle matter by sieving and particle length value from image analysis (Table 2). The mean particle size in faeces from ruminant animals increases due to increasing BW, increasing lignification of NDF and increasing intake of forage NDF relative to BW (Jalali et al., 2015).

Rumination is considered as the major process for the physical break down of feed particles, and rumen particles larger than CPS or longer than CPL are selectively retained in the reticulo-rumen system. Schulze et al. (2014a) observed a negative relationship between the mean particle size in faeces and rumination time per g iNDF and between the mean particle size in faeces and rumen degradation of NDF and DNDF. This indicates that a high degradation of DNDF is promoted by an effective particle size reduction during rumination.



**Figure 2** Density distribution of particle length of washed faeces particles from different animal species (S) fed different forage types (F) and the interaction between forage type and animal species (F\*S). F: Green hay and Grass seed straw S: Goats, sheep and llamas (Jalali et al. 2012b).

#### Feed intake

The intake is generally dependent on both animal and feed characteristics, and management (Mertens, 2007). Forage intake generally increases due to increasing digestibility, and decreasing NDF content, whereas the intake of forage decreases due to increased supplementation of concentrates (Mertens, 2007). Randby et al. (2012) observed an intake of 1.5% NDF<sub>f</sub> of BW in lactating dairy cows, whereas Nadeau et al. (20015a) observed and intake of 2% NDF<sub>f</sub> of BW in nursing ewes fed medium cut grass silage. Several models have been established for prediction of intake by dairy cows from concentrate supplementation and forage characteristics, such as digestibility, net energy (NE) content (NE/kg DM), NDF or crude fibre content, DM content and content of fermentation products (Jensen et al., 2015). Jensen et al. (2015) evaluated five recent models on a dataset of 140 treatment means of DM intake values by lactating dairy cows, and observed that models generally over predicted high intake and that the performance of the predictions ranged in RMSPE from 1<sup>1</sup>/<sub>2</sub> to 3 kg DM per day. Nørgaard and Mølbak (2001) observed a decreasing intake of NE at increasing dietary chewing index value (min/NEI) of lactating dairy cows, dry cows and growing cattle. The intercept value, predicted as the chewing index approaching zero, has been interpreted as the metabolic capacity for intake of net energy (NE<sub>0</sub>). Furthermore, the decrease in NE intake at increasing chewing index values has been found to be proportional with  $NE_0^2$ . The  $NE_0$  has been parameterized in lactating dairy cows to be depending on the metabolic body size (BW<sup>0.75</sup>), lactation performance and days in milk (Jensen, 2015; Jensen et al., 2016). In addition, Nielsen (2016) found that the NE<sub>0</sub>/BW<sup>0.75</sup> values did not differ between pregnant Hereford and pregnant large Charolais beef cows, but the ranking order of  $NE_0/BW^{0.75}$  values was lactating dairy cows > nursing ewes > pregnant ewes and pregnant beef cows. As an implication of the new model, Jensen (2015) showed an increasing substitution rate of forage for concentrate due to increased supplementation of concentrate, decreasing  $NE_0$  value and decreasing dietary chewing index values. Nielsen et al. (2015) observed decreasing intakes of metabolizable energy (ME) by pregnant ewes at increasing dietary chewing index values, which were corrected for the lower BW of ewes compared to dairy cows. These new findings of a linear relationship between the dietary chewing index value and the intake of energy appears to be a potential for modelling energy intake of different levels of supplementation and different forage qualities by use of the same intake model across different ruminating species of different body sizes.

#### Effects of forage feed value on intake and performance by ruminants

Delayed harvest of forages results in decreased intake and a need for increased supplementation of concentrate in order to maintain intake. The intake of NE is the principal driver for milk yield and daily gain in cattle, sheep, goats, and wild ruminants. Randby et al. (2012) observed decreasing forage intake, decreasing milk yield, increasing body weight loss and increasing milk acetone content in dairy cows due to decreased feed value of grass silage, which was supplemented with different levels of concentrates. In addition, the intake of NDF<sub>f</sub>/BW was negatively related to the energy balance and positively related with the acetone concentration in milk. Likewise, Helander et al. (2014) and Nadeau et al. (2015a) observed decreasing ME intake in nursing ewes and decreasing performance of their lambs due to decreased feed value of grass silages, which were supplemented with concentrates. Randby et al. (2010) observed decreasing NE intake and daily gain in growing bulls due to delayed harvest of timothy grass silage with or without concentrate supplementation. Dairy cows and growing cattle require a minimum intake of forage and NDF<sub>f</sub> in order to prevent rumen digestive disorders. The intake of NDF<sub>f</sub> is the major source of physically effective fibre (peNDF). Mertens (1997) recommended a minimum content of peNDF in diets for dairy cows of about 20% in order to avoid low rumen pH and low milk fat content. Nørgaard et al. (2011) recommended a minimum dietary chewing index value of 30 minutes per kg DM in order to prevent digestive disorders.

In a Swedish study conducted in 26 dairy herds, which differed in forage types fed to the cows, results showed that combining grass-clover silage with maize silage increased the concentration of undigested NDF in the faeces, contributing to a firmer consistency of the faeces compared to feeding grass-clover silage as the sole forage in the diet (P < 0.05; Mgbeahuruike et al., 2016). Furthermore, increasing forage DM intake decreased faecal DM concentration (r = -0.54, P < 0.01) but tended to result in cleaner cows, whereas increasing DM intake of concentrate increased faecal DM concentration (r = 0.63, P < 0.01), but might result in dirtier cows (Mgbeahuruike et al., 2016).

Based on a large amount of data from maize cultivars grown in different locations in Sweden, Mussadiq et al. (2013) concluded that the starch concentration and fibre digestibility of the whole-crop maize were the most important parameters for predicted milk yield per Mg DM according to MILK 2006 (Shaver et al., 2006). The importance of NDF digestibility of maize silage on milk yield of dairy cows were confirmed by Krämer-Schmid et al. (2016) in a metaanalysis using 96 dietary treatment means from 29 published experiments. A 0.01 unit increase in NDF digestibility of maize silage improved daily milk yield by 82 g (P = 0.04) and daily weight gain by 12 g (P = 0.03). The effect of improved fibre digestibility of maize silage on milk yield has previously been shown by Oba and Allen (1999), who reported 2.6 kg higher 3.5% fat-corrected milk yield (41.0 vs. 38.4, P < 0.0001), when cows were fed the brown midrib 3 mutant (bm3) maize silage with improved NDF digestibility compared to its normal counterpart. Intakes of DM, starch and NDF were 2.1 kg, 0.7 kg and 0.5 kg higher, respectively, for cows fed the bm3 maize silage, thus a higher energy intake. The in vitro NDF digestibility at 30 hours of incubation was 49.1% and 39.4% and the lignin concentration was 17 and 25 g/kg DM for the bm3 maize silage and its normal counterpart, respectively (Oba and Allen, 1999). Increased energy intake also was the driver for increased live-weight gain of bulls of Swedish Holstein and Swedish Red breeds (initial live weight: 390 kg, slaughtered live weight: 638 kg) when maize silage was fed as the sole forage compared with equal proportions of maize silage and grass silage on a DM basis in a total mixed ration (TMR) containing 60-65% forage of diet DM (Zaralis et al., 2014). The daily ME intakes were 140 and 133 MJ (P < 0.01) and the daily live weight gains were 1.78 and 1.67 kg (P < 0.01) for bulls fed 100% maize silage and bulls fed 50% maize silage and 50% grass silage of diet DM (Zaralis et el., 2014). When feeding the same forage treatments in TMR to growing lambs, live weight gain was unaffected (mean live weight gain: 445 g/day; Helander et al., 2015). The authors concluded that daily intake of crude protein and dietary metabolizable protein to ME ratio were closely related to live weight gain of the lambs. Furthermore, including maize silage in the forage component of the diet has a daily concentrate sparing effect in diets to ruminants (Keady, 2014).

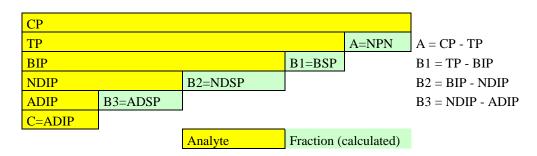
#### **PROTEIN QUALITY OF FORAGES**

Forage is an important protein source for ruminants but a large part of the protein in ensiled forage is already in the form of free amino acids and ammonia and as rumen degradable protein (RDP), which makes the utilization of forage protein in ruminants a challenging topic (Givens and Rulquin, 2004). To capture the free amino acids and ammonia for microbial protein synthesis, instant energy sources, such as sugars are needed, and for the RDP, also digestible fibre is needed as energy source, which tells us that the energy concentration of the forage is at least as important as its CP concentration as a majority of the metabolizable protein (MP) from forage originates from microbial protein (Merchen and Bourquin, 1994). Secondary fermentation of the silage can result in production of biogenic amines, which decrease intake and impair animal health (Pahlow et al., 2003; Krizsan et al., 2007; Saleem et al., 2012)

Protein quality can be described as plant CP fractions according to the Cornell Net Carbohydrate and Protein System (CNCPS; Sniffen et al., 1992) as described below. The fractions differ in solubility and rumen degradation and one fraction is considered indigestible. The fractions give us useful information on the types of energy sources needed for optimal protein utilization by the ruminant animal. It needs to be considered, though, that a portion of the soluble non-ammonia N may escape the rumen degradation and be available for absorption in the small intestine (Choi et al., 2002). There are several plant and management factors that affect the proportions of these CP fractions of which we will give an overview of the most important ones.

#### **Crude protein fractionation**

Licitra et al. (1996) divide the crude protein according to the CNCPS (Sniffen et al., 1992) into five different fractions A, B1, B2, B3, C (Pichard and Van Soest, 1977). Fraction A is the non-protein nitrogen (NPN), which is the nitrogen passing into the filtrate after precipitation with tungstic acid (Figure 3). B1 is the true protein soluble in borate-phosphate buffer at rumen pH and is degraded rapidly in the rumen. B2 is the true protein insoluble in borate-phosphate buffer, but soluble in the ND solution. Fraction B2 means the protein within the plant cell with high molecular weight and has variable degradation. B3 is the protein insoluble in the ND solution but soluble in the AD solution. This protein is normally cell wall-bound, digestible, but slowly degradable of which most occur post-ruminal. The ND solution is used without sodium sulfite, because sulfite cleaves disulfide bridges in cysteine and reduces the protein content in NDF (Licitra et al., 1996). Fraction C is the protein insoluble in the AD solution and is regarded as indigestible (Figure 3). This fraction is also called ADIN (acid-detergent insoluble nitrogen) and means nitrogen associated with lignin, Maillard products or none-enzymatic browning reaction caused by heating and drying (Licitra et al., 1996).



CP (crude protein), TP (true protein), NPN = non-protein nitrogen, BIP (buffer-insoluble protein), BSP (buffer soluble protein), NDIP (ND-insoluble protein), NDSP (ND-soluble protein), ADIP (AD-insoluble protein), ADSP (AD-soluble protein)

Figure 3 Analysis and calculation of crude protein fractions.

Shannak et al. (2000), Kirchhof (2007) and Edmunds et al. (2012) found strong correlations between *in situ* rumen undegraded protein (RUP) content for different feed stuffs and the crude protein fractions. The RUP content of different feedstuffs can be calculated for three rumen passage rates (2% / h, 5% / h, 8% / h) by use of different formulas. The regression equations by Kirchhof (2007) and Kirchhof et al. (2010) can used to calculate RUP for forage (Table 3).

The lower molecular weight proteins are grouped together in fraction B1, which together with fraction A form the parameter 'protein solubility' (A+B1). Normally, fraction C (ADIP) is between 2-8% of CP (Richardt et al. 2011, Nadeau et al., 2012b). Higher values are indicators for heat damaged protein (Weiss et al., 1986).

Table 3 Regression equations for estimating the ruminally undegraded feed protein (RUP) proportion (g/kg crude
protein) assuming passage rates of 2, 5 and 8% / h (RUP2, RUP5, RUP8; mod. Kirchhof et al., 2010)

	RUP2	RUP5	RUP8
Intercept	204.3207	321.923	285.5459
С	1.0753		1.2143
ADF		0.1676	
CP x (A+B1)	-0.0014	-0.0022	
CP x C x C		0.0001	
NDF x B2			0.0005
(A+B1) / NDF			-110.1740
$\mathbb{R}^2$	0.51	0.52	0.56

C = ADIP = acid-detergent insoluble protein, g/kg CP

A = NPN = non-protein nitrogen, g/kg CP

B1 = BSP = buffer-soluble protein, g/kg CP

B2 = NDSP = ND-soluble protein, g/kg CP

CP, ADF and NDF in g/kg DM

#### Effects of forage species and cultivars

Kirchhof et al. (2010) compared fresh forage legumes in the spring growth cycle and found greater proportions of NPN (fraction A) in white clover and kura clover (Trifolium ambiguum M. Bieb.) than in lucerne and birdsfoot trefoil (Lotus corniculatus L.) with red clover being intermediate. Lucerne had the greatest proportion of buffer-soluble true protein (BSP, fraction B1) whereas birdsfoot trefoil had the smallest, and white clover, red clover and kura clover were intermediate. Birdsfoot trefoil had a much greater proportion of ND-soluble protein (NDSP, fraction B2) than the other legumes, which did not differ as much. Red clover contained more of the AD-soluble protein (ADSP, fraction B3) than the other legumes. The AD-insoluble protein (ADIP, fraction C) was somewhat higher in red clover than in the other legumes. These differences in CP fractions resulted in greater RUP for red clover and birdsfoot trefoil than for the other legumes (Kirchhof et al., 2010). In comparison, Nadeau et al. (2016) showed lower NPN and NDSP concentrations but higher ADSP concentration in red clover than in lucerne. Furthermore, Fijalkowska et al. (2015a) showed greater proportion of true protein (TP) but smaller proportion of BSP in red clover than in lucerne. Krawutschke et al. (2011) reported lower concentrations of NPN and BSP but higher concentrations of ADSP and ADIN in red clover than in white clover. The soluble protein fraction can differ in rumen degradability between forage species. Hedqvist and Udén (2006) showed lower in vitro degradation rate and lower effective protein degradation of the soluble protein fraction in fresh red clover than in fresh white clover, birdsfoot trefoil and perennial ryegrass (Lolium perenne), which did not differ. Furthermore, lucerne silage had greater effective protein degradability than silages of red clover and red fescue (Festuca rubra L.), which did not differ (Purwin et al., 2014).

When comparing 27 cultivars of lucerne over two years, Tremblay et al. (2000) concluded that there is variability for protein degradability for cultivars with similar yield potentials. Thus, genetic selection for low rumen degradability and high DM yield is feasible. However, in a more limited experiment, no differences in the CP fractions (A, B1, B2, B3 and C) were found between four lucerne varieties (Nadeau et al., 2016). Similar results were obtained by Krawutschke et al. (2011) when comparing three red clover cultivars. However, when 133 entries of red clover, originating from different countries around the world, were compared in protein characteristics, degradation rate and estimated rumen escape ranged from 0.088 to 0.146/h and from 287 to 409 g CP/kg CP with a normal distribution. Hence, these results are promising in developing lines of red clover with improved protein utilization by ruminants (Broderick et al., 2004).

#### Effects of forage maturity

The *in vitro* CP degradation decreased with advancing maturity of fresh lucerne (bud, 1/10 bloom, full bloom) and smooth bromegrass (*Bromus inermis*; boot, early reproductive, seeded) in spring growth cycle as both NPN and soluble protein decreased (Kohn and Allen, 1995). Likewise, *in situ* degradation of CP decreased from 0.693 to 0.597 at advanced maturity of cooksfoot (*Dactylis glomerata* L.) from heading to flowering (Aufrère et al., 2003). Similar trends were seen in the CP fractions of legumes, where the ADSP (fraction B3) and the ADIN (fraction C) increased by 60% and 80%, respectively, from late vegetative to mid flowering stage of maturity. This resulted in increased RUP from 192 to 257 g/kg CP (Kirchhof et al., 2010). Also, Grabber (2009) reported increased ADSP with advancing maturity of ensiled lucerne and red clover.

#### Effects of nitrogen fertilization

Tremblay et al. (2005) did an extensive study on the effects of nitrogen fertilization rate on silage quality of timothy (*Phleum pratense* L.). The experiment involved four rates of N fertilization (0, 60, 120 and 180 kg N/ha) prior to the start of the growth in spring at two locations in Canada over two years. The main results were decreased concentration of water soluble carbohydrates (WSC), increased buffering capacity (BC) and nitrate concentrations, primarily in the early stages of development. Hence, the ensiling ability of timothy was diminished when high rates of N were applied. Silage pH, NPN, soluble-N and NH<sub>3</sub>-N concentrations increased with increasing N-fertilization rates, especially at the early development stages. Thus, silage quality was reduced at increasing N-fertilizer application rates and the effect was more evident at the early stages of maturity of timothy (Tremblay et al., 2005). Similarly, Keady and O'Kiely (1996) showed increased BC, pH and NH<sub>3</sub>-N concentrations of grass silage with increasing N application rates. Nitrogen fertilization increases the CP of plants with a greater increase in NPN than in protein-N (Fijalkowska et al., 2015b).

#### Effects of wilting and ensiling

Proteolysis during wilting seems to be affected by species that differ in NPN. Among the legumes, lucerne and white clover usually have higher levels of proteolysis during wilting than red clover (Owens et al., 1999; Krawutschke et al., 2011), which could be related to the presence of polyphenol oxidase, which produces phenolic compounds that inhibit proteolysis in red clover (Jones et al., 1995). Birdsfoot trefoil, which has low levels of tannins, had intermediate levels of proteolysis relative to lucerne and red clover (Papadopoulos and McKersie, 1983). In agreement with previous mentioned studies, Fijalkowska et al. (2015a) reported extensive proteolysis of lucerne during wilting and ensiling but very limited proteolysis in red clover silage. However, the ADIN was substantially higher in the silage than in the fresh forage of red clover, which will decrease the utilization of the protein by ruminants. Recently, Nadeau et al. (2016) reported that BSP (B1 fraction) decreased from 169 to 74 g/kg CP while the ADSP (B3 fraction) increased from 26 to

72 g/kg CP during wilting of lucerne (90%) /white clover (10%) forage to 40% DM in sunny weather for 6 hours. There was no effect on the NPN concentration during wilting (Nadeau et al., 2016). During wilting of white clover and red clover to 40% DM in another study, the TP decreased while the NPN increased and the proteolysis continued during ensiling (Krawutschke et al., 2011).

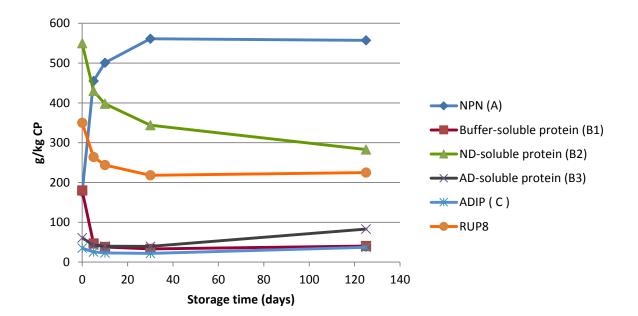
Wilting of early harvested grass-dominated forage (77% grass, 18% clover, 5% lucerne) from 15% to 35% DM for 23 hours in good weather conditions decreased BSP while the NPN, NDSP and ADSP increased resulting in an increased RUP at 8% passage rate per hour (Table 4; Nadeau et al., 2012b). When the wilted grass-dominated forage was ensiled for 125 days, there was a further decrease in BSP from 180 to 33 g/kg CP (Table 4). In addition, the NDSP decreased while the NPN further increased from 175 to 593 g/kg CP, which resulted in a decreased RUP (Table 4; Nadeau et al., 2012b). Changes in CP fractions over the course of ensiling until 125 days are shown in figure 4. Most of the proteolysis occurred during the first 10 days of ensiling and thereafter the rate of proteolysis decreased and stabilized after 30 days. Instead, there was a conversion from NDSP (B2) to ADSP (B3) after 30 days of ensiling (Nadeau et al., 2012b).

In an experiment where both extent and rate of wilting on CP fractions of grass silage were evaluated, it was reported that NPN decreased quadratically with increasing DM from 20 to 65% (Edmunds et al., 2014). Rapid wilting also decreased NPN, implying decreased proteolysis during wilting due to shorter wilting time. Furthermore, fast wilting resulted in more NDSP than slow wilting at all DM concentrations. Fast wilting and increasing DM concentrations of the grass silage (Edmunds et al., 2014). Likewise, McEniry et al. (2007) showed decreased NH<sub>3</sub>-N in grass silage with more rapid wilting. Ensiling alters the amino acid profile of the protein (Edmunds et al., 2014; Purwin et al., 2015), but rumen exposure basically reverts the amino acid profile to the profile found in the forage before ensiling (Edmunds et al., 2014).

**Table 4** Crude protein (CP), true protein (TP), CP fractions and rumen undegraded protein of forage as affected by wilting and ensiling (125 d of storage; adapted from Nadeau et al., 2012b).

	Unwilted forage	Wilted forage	Untreated silage	SEM	P - value
CP, g/kg DM	150 <sup>a,b</sup>	143 <sup>b</sup>	152 <sup>a</sup>	2.1	< 0.05
TP, g/kg DM	132 <sup>a</sup>	118 <sup>b</sup>	62 <sup>c</sup>	1.8	< 0.001
g/kg CP1					
NPN (A)	115 <sup>c</sup>	175 <sup>b</sup>	593 <sup>a</sup>	6.2	< 0.001
BSP (B1)	352 <sup>a</sup>	180 <sup>b</sup>	33°	6.9	< 0.001
NDSP (B2)	475 <sup>b</sup>	550 <sup>a</sup>	259°	8.9	< 0.001
ADSP (B3)	17 <sup>b</sup>	61 <sup>a</sup>	79 <sup>a</sup>	5.9	< 0.001
ADIP (C)	40	35	35	4.2	NS
RUP8	292 <sup>b</sup>	350 <sup>a</sup>	210 <sup>c</sup>	7.4	< 0.001

<sup>1</sup>NPN = non-protein nitrogen, BSP = buffer soluble protein, NDSP = neutral detergent soluble protein, ADSP = acid detergent soluble protein, ADIP = acid detergent insoluble protein, RUP8 = rumen undegraded protein at a ruminal passage rate of 8%/h. <sup>a,b,c</sup>Means with different superscripts within a row differ significantly at P < 0.05. NS = none significance.



**Figure 4** Changes in CP fractions and rumen undegraded protein at 8% passage rate per hour (RUP8) during ensiling of grass silage until 125 days of ensiling. Values are means over untreated and additive-treated silages, n=9 (Nadeau et al., 2012b).

#### Effects of silage additives

Both chemical and biological additives can reduce proteolysis during ensiling (Nadeau et al., 2000; Slottner and Bertilsson, 2006; Nadeau et al., 2012a,b; Nadeau and Auerbach, 2014). The effectiveness of inoculants on proteolysis is dependent on the WSC concentration of the forage. Inoculants will restrict proteolysis of forages with moderate to high WSC concentrations but will be limited in their actions at low forage WSC concentrations and under those conditions, formic acid is more effective (Davies et al., 1998). This was recently confirmed by Nadeau et al. (2016), who reported no effect of an inoculant containing homofermentative lactic acid bacteria (Kofasil Lac, Addcon Europe GmbH) on the proteolysis of lucerne silage, whereas the use of a formic acid based additive (GrasAAT SP, Addcon Nordic AS) decreased the NPN of the silage from 612 to 554 g/kg CP. Furthermore, Purwin et al. (2014) reported increased effective protein degradability in formic acid treated lucerne silage but similar effect was not found in silages of red clover and red fescue. When highly digestible grass with a WSC content of 215 g/kg DM was ensiled, both a bacterial inoculant and a salt-based additive decreased NPN from 597 to 537 g/kg CP and increased RUP at 8% passage rate per hour from 210 to 233 g/kg CP (Nadeau et al., 2012b). The NH<sub>3</sub>-N concentration was 73, 54 and 69 g/kg total N of the untreated, inoculant treated (Kofasil Life, Addcon Europe, GmbH) and chemically treated (Kofasil Ultra K, Addcon Europe GmbH) silages at 125 days of ensiling (Nadeau et al., 2012a). Others also have found decreased NPN by use of a bacterial inoculant to grass silage (Kramer et al., 2012).

#### Effects on ruminant performance

When red clover silage has been compared to lucerne silage in production trials with dairy cows, red clover has resulted in higher feed efficiency in kg milk/kg DM intake, higher nitrogen efficiency in milk N/N intake and lower milk urea content (Broderick et al., 2001). The authors related the results to the lower NPN concentration of the red clover silage and the higher apparent digestibility of dietary DM, OM and fibre resulting in higher net energy for lactation of the red clover silage diets compared to the lucerne silage diets. The improved apparent nitrogen efficiency of red clover diets compared to lucerne diets also was shown in later trials, although the response in milk yield was lacking, which partly was explained by energy partitioning into fat storage rather than milk fat secretion (Broderick et al., 2007). There also was an elevated ADIN concentration in the red clover silage that could have impaired the N utilization by the cows (Broderick et al., 2007). When red clover silage partially replaced grass silage in diets, the microbial protein flow from the duodenum per unit of DM intake was decreased. However, lower rumen degradable protein could have increased the flow of undegraded feed protein to the duodenum, resulting in similar total protein flow (Merry et al., 2006; Moorby et al., 2009). Increasing proportions of red clover in the diets increased DM intake and milk yield and the proportion of C18 poly-unsaturated fatty acids but decreased concentrations of fat and protein in the milk (Moorby et al., 2009). Birdsfoot trefoil, with higher levels of condensed tannins, has been shown to increase milk yield, milk protein yield, decrease milk urea content and improve milk nitrogen use efficiency compared to lucerne silage in diets to dairy cows (Hymes-Fecht et al., 2013).

As mentioned previously, wilting improves RUP of grass silage (Nadeau et al., 2012b) and Kebreab et al. (2000) reported increased content and yield of milk protein and decreased proportion of N excretion in urine of total manure excretion of dairy cows when fed grass silage, which was wilted for 24 hours to a DM of ca 30% compared to direct cut grass silage at ca 20% DM. The same response on milk protein and N excretion in urine was achieved when a medium application rate of 75 kg of N/ha in the spring before the first harvest was used instead of a high application rate of 150 kg of N/ha. Also, early harvested grass fertilized at a medium N application rate resulted in higher milk protein yield and a lower proportion of total manure N excretion being excreted in the urine than early and late harvested grass fertilized with high N application rate. In summary, early harvest of grass, which is fertilized with a medium rate of N and wilted to ca 30% DM is preferred to dairy cows (Kebreab et al., 2000). Increased RUP will not necessarily result in improved dairy cow performance if its amino acid profile will not meet the requirements of the first limiting amino acids. Edmunds et al. (2013) concluded that rumen degradation changes the amino acid composition of forage and that the amino acid composition of RUP is more similar between forages than to their original composition. This information can help decreasing the number of samples that need to be analysed to gain more knowledge on the effect of rumen exposure on the amino acid composition of forages (Edmunds et al., 2013).

In agreement with Nadeau et al. (2016), Broderick et al. (2007) reported decreased proteolysis and, thereby, lower contents of NPN, ammonia N and free AA N in lucerne silage treated with ammonium tetraformate (GrasAAT, Norsk Hydro ASA) compared to untreated lucerne silage. When fed to dairy cows, the daily DM intake increased by 1.0 kg and the 3.5% fat-corrected milk increased by 2.1 kg. Content and yield of milk true protein increased and nitrogen efficiency in milk N per unit of N intake increased by 1.3 units (Broderick et al., 2007). This production response was, though, not repeated in a second trial.

In an experiment by Nadeau et al. (2014) dairy cows were fed a diet containing 52% grass silage of total DM intake and 170 g CP/kg DM. The silages were treated with the bacterial inoculant Kofasil Life (*Lactobacillus plantarum* DSM 3676, 3677; Addcon Europe GmbH), with the chemical additive Kofasil Ultra K (sodium nitrite, hexamine, sodium benzoate, potassium sorbate; Addcon Europe GmbH) or left untreated. The additives decreased NH<sub>3</sub>-N (5.8 vs. 7.3% of total N, P < 0.001) increased BSP (6.1 vs. 2.4% of CP, P < 0.01) and tended to increase the more slowly degradable ADSP (7.4 vs. 6.1% of CP, P < 0.10) compared to untreated silage. The chemical additive decreased contents of urea in milk without affecting the daily nitrogen intake and milk yield, indicating improved protein utilization (Table 5). The excretions of purine derivatives in urine were higher from cows fed silage treated with the chemical additive, indicating a tendency for increased microbial protein flow to the duodenum. Furthermore, the chemical additive decreased the somatic cell count in milk (Table 5). The cows increased in live weight by 5 kg during the 20-day period, which corresponds to a daily milk yield of ca 1.8 kg calculated on an energy basis (GfE, 2001). Use of silage additives can decrease proteolysis during ensiling resulting in potentially improved protein utilization and udder health of dairy cows (Nadeau et al., 2014).

**Table 5** Intake, live weight, milk yield, milk and urine components of dairy cows fed diet containing grass silage treated with or without additives<sup>1</sup> (Nadeau et al., 2014).

		Bacterial	Chemical		
	Control	inoculant	additive	SEM	P - value
Dry-matter intake (% of liveweight)	3.62	3.46	3.54	0.109	NS
Crude protein intake (kg/day)	3.92	3.84	3.84	0.112	NS
Live weight (kg) <sup>2</sup>	645 <sup>b</sup>	650 <sup>a</sup>	650 <sup>a</sup>	8.6	< 0.05
Energy-corrected milk (kg/day)	40.0	39.9	39.4	0.74	NS
Milk urea (mg/l)	240 <sup>b</sup>	248 <sup>a</sup>	230°	4.2	< 0.0001
Milk somatic cell count (no./ml)	92 046 <sup>a</sup>	58 787 <sup>a,b</sup>	51 766 <sup>b</sup>	16 351	< 0.05
Urea in urine (g/day)	383	409	395	13.7	NS
Allantoin in urine (g/day)	91	97	109	6.1	< 0.10
Purine derivatives in urine (g/day)	95	108	115	10.6	< 0.10

<sup>1</sup>control; without additive, bacterial inoculant Kofasil Life, chemical additive Kofasil Ultra K (Addcon Europe GmbH) <sup>2</sup>SEM is calculated from the variance of the random factor cow within treatment and the variance of the error term in the model of which the factor cow (=variation between cows within treatment) is much greater than the variance of the error term in the model for live weight. When the treatments are compared, the variance of the error term is used. NS = none significance

In a later experiment, grass silage was treated with the inoculant Kofasil Duo (*Lactobacillus plantarum/Lactobacillus buchneri*, 200,000 cfu/g) or with the chemical additive Kofasil Ultra K (Addcon Europe GmbH), which were compared with untreated silage (Nadeau et al., 2015b). The silage contained 15% CP, 47% NDF, 3.3% WSC, 8.2% lactic acid, 2.1% acetic acid and 0.25% NH<sub>3</sub>-N of DM with minor differences between treatments. The RUP of the silage at 5% passage rate per hour was 20% of CP for the control and 22% of CP for the inoculant and the salt-based additive. Diets were isonitrogenous (15% of DM) and isoenergetic (11.1 MJ ME/kg DM) varying in RUP (4.9% (high) and 2.9% (low) of DM). Dietary forage proportion of the TMR was 58% of DM. High RUP diet had higher milk yield than low RUP diet (29.4 *vs.* 27.9 kg; P < 0.05). The DM intake was not affected by RUP and silage treatment. Yields of milk and ECM were higher for the diets containing additive treated silages than for the control diet at low RUP (28.9 *vs.* 26.0 kg milk, P < 0.01; 30.6 *vs.* 27.1 kg ECM, P < 0.001) whereas there was no effect of additive treatment in the high RUP diet. Milk fat and protein percentages did not differ between silage treatments. Feed efficiency was higher for the diets containing the adidtive-treated silages than for the control diet at the low RUP (1.6 *vs.* 1.3 kg of ECM/kg DM intake, P < 0.001) but not at the high RUP. The increased milk yield and feed efficiency when fed a diet with low RUP can partly be explained by increased RUP of the additive-treated silages (Nadeau et al., 2015b).

# CONCLUSIONS

Forage characteristics, as affected by intrinsic plant factors in combination with management factors during harvest, storage and feed out, have major impact on the concentrations and fractionations of fibre and protein in forages, resulting in differences in solubility and digestibility of protein and digestibility of fibre and DM of forages. Variations in nutrient contents of forages determine the amounts of concentrates needed for maintaining growth rate and milk yield of ruminant animals. Forage fibre content affects DM intake and time spent chewing for mastication of forage fibre and protein in the faces. Improved fibre digestibility of forages can increase milk production and live weight gain of ruminants. The energy released during fermentation of carbohydrates is used for the microbial protein synthesis from ammonia, free amino acids and peptides from NPN and from degraded true protein in the rumen. The microbial protein forms together with the rumen undegraded feed protein metabolizable protein that is used for growth and milk yield by ruminant animals.

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# A review on additives for grain silages

**BACK** 

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# Introduction

Ensiling is an efficient strategy for grain storing and processing. Improvements on the nutritive value and the lower costs compared with other processing methods has stimulated the use of high moisture grain silages (HMGS). The typical lower field/harvesting losses accompanied of early harvesting are considered side advantages, which may increase farming efficiency. Insect and rodent damages typically observed in dry grains are also reduced by adopting HMGS. Additionally, HMGS allows the use of homegrown, traceable (source-verified) feedstuffs instead of purchased concentrates. However, it will constraint the cash crop at farm level because the wet storage.

To exploit the benefits of HMGS, a proper management is mandatory to minimize fermentative losses and prevent the aerobic deterioration. A number of studies have accessed of the effects of silage additives on HMGS, however, to our knowledge, a systematic analysis of these data has not been conducted. Moreover, little is known about the consequences of silage additives on the performance of animals fed HMGS.

This review focused on high moisture corn (HMCS) and winter cereal grain (HMWCS; barley, wheat and triticale) silages. The objective of this meta-analysis was to address the effect of additives (chemical and microbial) on the conservation and nutritive value of HMCS and HMWCS. We also addressed the effects of treating HMGS with additives on animal performance.

#### Data set and statistical analysis

Two data sets based on a literature review were compiled from scientific papers that reported treatment means. Silages made from whole or processed grains were considered and the minimal storage period adopted was 30 d. To analyze the effect of applying additives on winter cereals silages, the data set included 3 referred journal articles (Adesogan et al., 2003; Mathison et al., 1989; Pieper et al., 2011) and 3 abstracts (Davies et al., 2009; Seppala et al., 2015; Stacey et al., 2009). In this case, the small number of publications found is due to the lower use of grain silages when compared to the production of whole plant silages of winter cereals.

The data set used to study high moisture corn silages was composed by 24 referred journal articles (Bíro et al., 2006, 2009; Canibe et al., 2014; Da Silva, T. et al., 2015; Dawson et al., 1998; Doležal and Zeman, 2005; Dutton and Otterby, 1971; Ferrareto et al., 2015; Flores-Galarza et al., 1985; Gálik et al., 2007, 2008; Ítavo et al., 2006, 2009; Jobim et al., 2008; Kung et al., 2004, 2007; Loučka, 2010; Morais et al., 2012; Prigge et al., 1976; Pys et al., 2009; Reis et al., 2008; Taylor and Kung, 2002; Wardynski et al., 1993); one technical note (Basso et al. 2012) and 10 abstracts published in international scientific meetings on forage conservation (Auerbach et al., 2015; Coudure et al., 2012; da Silva, N. et al, 2015; Davies et al., 2009; Doležal et al., 2014; Gallo et al., 2015; Mlynar et al., 2006; Pys and Kowalski, 2014; Pys et al., 2010; Revello-Chion et al., 2012).

Data sets of corn and winter cereals were analyzed separately. A minimal of four treatment means from at least two articles was the prerequisite for keep the dependent variable into the data set. Data were analyzed using the mixed procedure of SAS (Littell et al., 1996). The model included a fixed effect of treatment (control or additive) and random effect of experiment, due to the variations across experimental protocols that would contribute to study effects in these comparisons (St-Pierre, 2001).

Because the current knowledge indicates divergent responses for types of silage additives, they were sorted in different classes: "Homolactic" (homolactic bacteria), "Hetero" (heterofermentative bacteria), "Combo" (Homolactic plus heterofermentative bacteria) and "Chemical" (chemical additives).

## Additives to HMCS

# Chemical additives

Chemical additives reported in the reviewed articles included in the final data set are shown in Table 1, with the respective application rates as reported by the authors. While some compounds were used alone (pure compounds or solutions), most treatments were based on mixtures of chemical substances.

Table 1	Description of	chemical	additives us	sed in the	meta-analysis
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Additive	Application rate
Ammonia	1.1% to 2.3%
Ammonium isobutyrate	2%
Diammonium phosphate	4,6%
Formic acid	3L/t to $4L/t$
Sulfur dioxide	1.3% to 1.7%
Urea	0.4 to 2%
Urea solution	50L/t
Acetic acid, isobutyric acid	8L/t
Ammonium formate, propionate, ethyl benzoate and benzoate	4 L/t
Ammonium propionate, sodium propionate, acetic acid, benzoic acid and sorbic acid	0.1% to 0.2%
Formic acid, ammonium formate, propionic acid, benzoic acid	4L/t
Formic acid, ammonium formate, propionic acid, benzoic acid and ethylbenzoate	6L/t
Formic acid (42.5%), formic ammonia (30.3%) and propionic acid (10%)	6 L/t
Formic acid (55%), propionic acid (20%), ammonium formate (4.3%) and potassium sorbate (2.5%)	4 L/t
Formic acid (55%), propionic acid (5%) and ammonium formate (24%)	4 L/t to 4.5 L/t
Formic acid (55%), propionic acid (5%) and ammonium formate (24%) plus ammonium	
Propionate	4.5 L/t
Propionic acid (80%) and acetic acid (20%)	1.5%
Propionic acid and formic acid	3.5 kg/t
Propionic acid-based additive: ammonium and sodium propionate, ethoxyquin, BHA, and BHT	0.1 to 0.2%
Propionic acid (50%) and formic acid (50%)	3 L/t
Propionic acid (90%), ammonium propionate (4%) and 1,2-propandiol (4%)	3 L/t
Propionic acid, ammonium propionate, sodium benzoate, potassium sorbate	1.5 to 3L/t
Propionic, acetic, benzoic and sorbic acids, sodium and ammonium hydroxide, methylparaben and	
propylparaben (Liquid mold inhibitor, 82% acid content)	0.1%
Propionic acid, formic acid, benzoic acid and calcium formate	3.4 kg/t
Propionic acid (37%), sodium benzoate (14%) and sodium propionate (11%)	5 L/t
Sodium benzoate (22.9%) and sodium propionate (8.3%)	3 L/t to 6 L/t
Sodium benzoate (5 to 50%), potassium sorbate (5 to 35%) and sodium nitrite <5%	2 L/t to 6 L/t
Sodium benzoate, sodium azide and calcium formate	3.5 kg/t
Potassium sorbate, sodium benzoate, ammonium propionate	1 L/t to 2L/t

High moisture grain silages with or without chemical additives displayed a large variation in chemical composition, DM loss, microbial counts and aerobic stability, indicating that the data set was broad, representative and covered a large part of the practical wide range of HMCS (Table 2). As expected, HMCS treated with chemical additives revealed a significant fermentation inhibition as indicated by the higher content of WSC and lower content of fermentation end-products, especially lactic acid.

Chemical additives were also effective in preventing DM losses, which explains the higher DM content of silages containing chemicals. Changes in variables such ash, CP, N-NH<sub>3</sub> and pH reflect the formulations of chemicals added to the silages. The presence of minerals in the chemicals altered the ash content of silages and the presence of nitrogenous compounds affected CP and N-NH<sub>3</sub> concentration, in addition to the inhibitory effects on the microorganisms, preventing the pH drop.

A noticeably response achieved with chemical additives was the higher aerobic stability of silages, since spoiling microorganisms, such as yeasts were markedly decreased. Higher stability associated with lower nutrient oxidation upon air exposure is a reasonable justification to recommend chemical additives for HMCS.

	Data set					Treatme	CEM		
Item	$n^1$	Mean	SD	Minimum	Maximum	Control	Chemical	SEM	Р
DM, g/kg	62	661.8	38.2	598.0	748.0	657.8	665.8	6.88	0.02
Ash, g/kg DM	26	15.2	1.49	13.3	19.1	14.6	15.8	0.38	0.01
NDF, g/kg DM	24	99.7	6.79	79.2	107.0	99.5	99.9	2.00	0.59
ADF, g/kg DM	24	39.7	12.3	22.8	57.4	39.8	39.7	3.64	0.85
Hemicellulose, g/kg DM	24	59.9	11.9	42.4	76.2	59.7	60.2	3.50	0.35
Starch, g/kg DM	40	712.9	51.8	593.0	796.7	711.5	714.3	11.7	0.44
Ether extract, g/kg DM	14	43.3	8.33	34.8	66.2	45.5	41.1	3.15	0.36
CP, g/kg DM	36	95.8	29.9	57.3	202.0	88.9	102.8	6.95	0.04
N-NH <sub>3</sub> , g/kg DM	40	0.54	1.07	0.00	5.00	0.33	0.76	0.24	0.21
Soluble protein, g/kg CP	6	520.1	21.7	476.7	531.8	531.8	508.3	11.3	0.28
WSC, g/kg DM	20	9.84	3.97	1.00	15.2	8.34	11.3	1.19	< 0.01
pН	74	4.41	0.72	3.70	8.30	4.31	4.51	0.12	0.08
Lactic acid, g/kg DM	78	12.4	7.09	0.20	26.5	14.0	10.8	1.11	$<\!0.01$
Acetic acid, g/kg DM	78	4.89	2.84	0.00	16.0	5.20	4.57	0.46	0.15
Propionic acid, g/kg DM	64	1.02	2.52	0.00	18.3	0.16	1.89	0.42	< 0.01
Butyric acid, g/kg DM	24	0.23	0.26	0.00	0.70	0.27	0.18	0.07	0.23
Ethanol, g/kg DM	58	6.91	9.05	0.00	44.0	9.50	4.32	1.62	< 0.01
Lactic:Acetic ratio	78	3.79	3.33	0.00	11.3	3.82	3.77	0.58	0.88
LAB, log cfu/g	6	2.25	0.23	2.00	2.45	2.45	2.04	0.03	0.01
Yeasts, log cfu/g	20	3.35	1.19	0.57	4.69	4.06	2.65	0.31	< 0.01
DM losses <sup>2</sup> , g/kg	10	14.2	11.9	5.8	41.0	16.7	11.8	5.50	0.11
Aerobic stability, h	52	125	112	21	500	59	190	18	< 0.01

 Table 2
 Data set of high moisture corn silages treated without or with chemical additives and their effects on silage quality

<sup>1</sup> Number of means, <sup>2</sup> Fermentative losses.

#### **Microbial additives**

Nowadays, there is enough knowledge indicating divergent responses for homolactic and heterofermentative microbial inoculants (Kung et al., 2003). Thus, homofermentative, heterofermentative and combinations of homolactic and heterofermentative bacteria were evaluated separately. The microbial species used as silage inoculants are described in Table 3.

Homofermentative bacteria are recognized for their efficiency in producing lactic acid, which is a strong acid (pKa = 3.86) capable to quickly drop the pH decreasing fermentative losses. On the other hand, heterofermentative bacteria are skilled in ferment sugars (pentoses and hexoses) into other products besides lactic acid, for instance acetic and propionic acids. These weak acids are good antifungal agents able to promote aerobic stability in silages (Moon, 1983).

 Table 3
 Microorganisms used as silage inoculants in the current meta-analysis

Bacteria	Inoculation rate (cfu/g as fed)
Lactobacillus buchneri	$5 \times 10^4$ to $5 \times 10^6$
Lactobacillus fermentum	1×10 <sup>5</sup>
Lactobacillus plantarum	$5 \times 10^4$ to $1 \times 10^7$
Leuconostoc mesenteroides	$1 \times 10^{5}$
Propionibacterium acidipropionici	1×10 <sup>7</sup>
Propionibacterium freudenreichii	$1 \times 10^{7}$
L. buchneri and L. plantarum	$2.5 \times 10^5$ to $6 \times 10^5$
L. buchneri and P. pentosaceus	$7.5 \times 10^5$ to $9 \times 10^5$
L. plantarum and $\hat{P}$ . acidipropionici	$1.5 \times 10^5$ to $3 \times 10^5$
L. plantarum and P. freudenreichii	$1 \times 10^5$ to $1 \times 10^7$
L. rhamnosus and E. faecium	$1 \times 10^5$ to $5 \times 10^5$
P. pentosaceus and P. freudenreichii	$1.2 \times 10^5$ to $2.4 \times 10^5$
L. buchneri, L. plantarum and E. faecium	5×10 <sup>6</sup>
L. plantarum, E. faecium, and P. acidilactici	$1.5 \times 10^5$ to $2 \times 10^6$
L. plantarum, P. pentosaceus and P. acidipropionici	$1.5 \times 10^{5}$
L. plantarum, L. bulgaricus and L. acidophilus	$1 \times 10^{5}$
L. plantarum, L. casei, E. faecium and P. pentosaceus	$5 \times 10^{4}$
L. buchneri, L. plantarum, E. faecium, L. casei, and P. pentosaceus	$1.5 \times 10^{5}$
L. buchneri, L. plantarum, L. brevis, L. rhamnosus and P. pentosaceus	$2.5 \times 10^{5}$

## Homolactic bacteria

Chemical composition, DM loss, microbial counts and aerobic stability of HMGS with or without homolactic inoculants are shown in Table 4. Nutrient composition of HMCS were quite similar. Silages treated with the homolactic inoculants showed higher protein content and reduced ammonia content mainly due to the inhibition of proteolysis.

Although the database did not provide quantification of LAB, there was a trend towards greater use of soluble carbohydrates in the inoculated silages. As consequence of the typical metabolism of added bacteria, the lactic acid content was higher in silages inoculated with homolactic bacteria, and this difference promotes significant changes in pH. The DM losses has been numerically lower in inoculated silages, however, both control and treated silages had shown low fermentative losses. Low concentrations of other organic acids indicated that fermentation profile was generally shortly interrupted.

Table 4	Data set of high moisture corn silages treated without or with homolactic inoculants and their effects on silage
quality	

Data set					Treat	ment effect	CEM	Р
$n^1$	Mean	SD	Minimum	Maximum	Control	Homolactic	SEM	Ρ
28	669.3	54.9	595.0	768.0	665.9	672.5	14.9	0.13
6	34.5	3.37	29.3	38.4	32.3	36.7	1.54	0.18
20	85.7	11.1	66.0	97.6	84.5	86.9	3.57	0.03
18	0.26	0.17	0.10	0.70	0.20	0.19	0.04	0.06
4	296.3	119.1	225.0	473.0	225.0	367.5	74.6	0.41
8	26.5	10.8	12.3	37.4	29.1	23.9	5.62	0.12
24	4.29	0.48	3.88	5.65	4.42	4.16	0.14	0.02
16	24.5	17.4	8.80	69.3	21.4	27.6	6.25	0.10
16	9.13	9.14	1.10	28.7	9.85	8.41	3.33	0.40
10	0.77	0.61	0.00	1.50	1.00	0.54	0.27	0.27
12	7.85	9.66	2.70	28.5	8.23	7.47	4.13	0.22
16	3.96	2.42	1.64	9.74	3.02	4.89	0.81	0.12
6	4.77	0.73	3.92	5.67	4.76	4.78	0.47	0.95
10	17.4	24.2	4.60	68.0	18.3	16.5	11.5	0.42
4	118	27	96	156	138	98	13	0.27
	$28 \\ 6 \\ 20 \\ 18 \\ 4 \\ 8 \\ 24 \\ 16 \\ 16 \\ 10 \\ 12 \\ 16 \\ 6 \\ 10 \\ 4$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$

<sup>1</sup> Number of means, <sup>2</sup> Fermentative losses.

Unsurprisingly, homolactic inoculants were less effective in controlling aerobic deterioration, since lactic acid has a typical weak antifungal property (Moon, 1983). The influx of air into the silage mass has negative effects on silage quality, especially in HMGS due to its high content of nutrients, low moisture, and because it ferments more slowly and less extensively compared to typical forage crop silages (Taylor and Kung, 2002). Nutrient losses and excessive production of heat by microbial spoliation result in lower feed quality and may result in poor animal performance (Hoffman and Ocker, 1997; Salvo et al., 2015). This makes the use of exclusively homolactic microorganisms inappropriate for HMGS.

#### Heterofermentative bacteria

The characteristics of HMCS treated or not with heterofermentative inoculants are presented in Table 5. Overall quality of HMCS was typical for well-preserved silages, although DM and WSC contents, which are key factors for silage fermentation, showed a large range.

Indeed, the production of antifungal compounds (e.g., acetic and propionic acids) by heterofermentative bacteria was an effective way for decreasing yeast and fungi population (Honing and Woolford, 1980) and largely improved the aerobic stability of HMGS. Silages inoculated with heterofermentative strains had lower WSC and higher content of total acids, indicating higher fermentative activity. *Lactobacillus buchneri*, a typical heterofermentative bacteria, has a predominant metabolic pathway leading to accumulation of acetic acid, whereas lactic acid concentration and pH, in general, remains similar to control silages. Furthermore, heterofermentative strains increases propionic acid as well, which might be produced either by addition of *Propionibacterium* spp or by the degradation of 1,2- propanediol (Krooneman et al., 2002) resulted from *L buchneri* metabolism.

It is important to highlight the inoculation rates of microbial inoculants. In the study reported by Taylor and Kung (2002), the inoculation of HMGS stored for 92 d with a low dose of *L. buchneri*  $(1 \times 10^5 \text{ cfu/g})$  did not enhance the aerobic stability. In contrast, application rates  $\geq 5 \times 10^5$  cfu/g improved the aerobic stability more than six-fold compared with untreated HMGS stored for the same period. However, in silages stored for 166 d, *L. buchneri* improved the aerobic stability even at  $1 \times 10^5$  cfu/g. Additionally to the inoculation rate, extending the length of storage is a potential practice to improve the aerobic stability and nutritive value of HMGS (Taylor and Kung, 2002; Hoffman et al., 2011; Der Bedrosian et al., 2012).

			Dat	a set	Treatmen	nt effect	SEM		
Item	$n^1$	Mean	SD	Minimum	Maximum	Control	Hetero	SEM	Р
DM, g/kg	74	698.9	37.4	629.5	738.0	700.7	697.1	6.19	0.01
Ash, g/kg DM	10	14.0	0.92	12.7	15.6	14.0	13.9	0.44	0.73
NDF, g/kg DM	26	78.2	17.3	55.3	106.5	78.3	78.1	4.89	0.85
ADF, g/kg DM	26	26.9	13.7	10.4	56.8	26.9	26.9	3.87	0.92
Hemicellulose, g/kg DM	26	51.3	10.7	38.6	74.3	51.5	51.1	3.02	0.78
Starch, g/kg DM	12	740.2	38.0	687.0	794.0	739.8	740.6	16.3	0.85
CP, g/kg DM	32	94.9	11.6	72.3	109.6	95.3	94.5	2.95	0.48
N-NH <sub>3</sub> , g/kg DM	64	0.24	0.21	0.03	0.81	0.23	0.26	0.04	0.02
WSC, g/kg DM	38	1.89	2.51	0.10	10.8	2.38	1.41	0.57	< 0.01
рН	66	4.22	0.43	3.73	5.65	4.24	4.20	0.07	0.39
Lactic acid, g/kg DM	66	13.5	10.6	1.40	39.0	13.8	13.2	1.87	0.17
Acetic acid, g/kg DM	66	5.75	4.21	0.40	27.1	4.06	7.44	0.68	< 0.01
Propionic acid, g/kg DM	36	0.37	0.67	0.00	3.50	0.10	0.65	0.14	0.01
Butyric acid, g/kg DM	8	0.16	0.24	0.00	0.60	0.23	0.10	0.12	0.19
Ethanol, g/kg DM	46	5.87	4.92	1.20	18.0	6.07	5.67	1.04	0.25
Lactic:Acetic ratio	66	3.11	2.95	0.48	10.9	3.71	2.51	0.51	< 0.01
LAB, log cfu/g	6	7.88	0.54	7.11	8.46	7.63	8.13	0.30	0.34
Yeasts, log cfu/g	46	4.24	1.34	1.34	6.70	4.83	3.65	0.24	< 0.01
Molds, log cfu/g	30	3.65	1.98	1.10	7.29	3.95	3.36	0.51	< 0.01
DM losses <sup>2</sup> , g/kg	16	30.1	10.4	7.50	41.0	27.2	33.0	3.65	0.07
Aerobic stability, h	72	129.0	114	20.0	450	70	188	16	< 0.01

 Table 5
 Data set of high moisture corn silages treated without or with heterofermentative inoculants and their effects on silage quality

<sup>1</sup> Number of means, <sup>2</sup> Fermentative losses.

# Combination of homo- and hetero-fermentative bacteria

Chemical composition and fermentative characteristics of silages treated with homo and heterofermentative combined inoculants are presented in Table 6. Silage protein content was greater whereas ammonia concentration tended (P=0.11) to be lower in treated silages, which would indicate a lower proteolysis in those silages

**Table 6**Data set of high moisture corn silages treated without or with combinations of homo- and hetero-fermentativebacteria and their effects on silage quality

	Data set				Treatme	SEM	Р		
Item	$\mathbf{n}^1$	Mean	SD	Minimum	Maximum	Control	Combo	- SEM	Ρ
DM, g/kg	52	685.3	47.8	596.3	739.0	686.2	684.3	9.46	0.17
Ash, g/kg DM	28	14.0	0.708	12.7	15.5	13.8	14.1	0.19	0.12
NDF, g/kg DM	8	100.1	5.3	91.1	107.0	101.6	98.5	2.72	0.02
ADF, g/kg DM	8	40.9	13.9	27.3	56.8	41.2	40.6	7.51	0.18
Hemicellulose, g/kg DM	8	59.2	14.8	39.7	74.3	60.4	57.9	7.95	0.10
Starch, g/kg DM	30	718.7	31.7	679.0	794.0	719.6	717.8	8.32	0.50
Ether extract, g/kg DM	8	38.2	3.83	33.9	44.2	38.2	38.2	2.07	1.00
CP, g/kg DM	36	82.4	8.98	72.3	97.2	81.8	83.1	2.14	0.05
N-NH <sub>3</sub> , g/kg DM	44	0.20	0.21	0.00	0.80	0.23	0.18	0.04	0.11
WSC, g/kg DM	10	4.05	3.94	0.10	10.8	4.86	3.24	1.83	0.04
pH	52	4.21	0.41	3.73	5.65	4.29	4.13	0.08	0.03
Lactic acid, g/kg DM	44	15.3	6.81	3.90	25.1	15.3	15.4	1.47	0.90
Acetic acid, g/kg DM	44	5.77	3.14	1.50	14.2	5.08	6.47	0.66	0.03
Propionic acid, g/kg DM	18	0.37	0.47	0.00	1.38	0.16	0.59	0.14	0.04
Butyric acid, g/kg DM	14	0.19	0.19	0.00	0.62	0.23	0.16	0.07	0.47
Ethanol, g/kg DM	42	3.52	3.02	0.90	15.6	4.06	2.98	0.66	< 0.01
Lactic: Acetic ratio	42	3.18	2.10	0.14	8.63	3.11	3.25	0.46	0.78
Yeasts, log cfu/g	10	3.61	1.13	2.00	5.67	4.37	2.84	0.37	0.03
Molds, log cfu/g	18	1.83	0.58	1.09	2.90	1.82	1.83	0.20	0.94
DM losses <sup>2</sup> , g/kg	24	27.8	9.96	4.00	42.2	27.3	28.3	2.94	0.56
Aerobic stability, h	28	216	117	35	427	194	237	31	0.22
1 Number of moons 2 Former					.= /	-> .	-01	51	

<sup>1</sup> Number of means, <sup>2</sup> Fermentative losses.

Silages treated with combo inoculants showed lower NDF content, which can be attributed to a higher hemicellulose disappearance (P = 0.10). A recent research on the production of ferulic acid esterase (FAE) by *L. buchneri*, reported that FAE was active only during the onset (3 days) of fermentation (Addah et al., 2015). It is likely that a high pH (> 5.6) is needed for FAE activity, as observed at the beginning of fermentative process. It should be also noted that in the current meta-analysis, the inoculation with heterofermentative bacteria did not change the fibrous components of HMGS, most probably because none of the strains tested provided FAE activity.

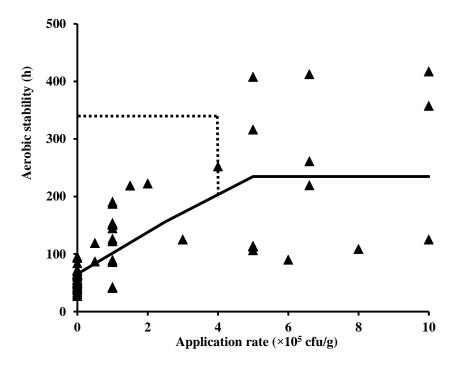
The fermentation profile observed in silages treated with combo inoculants blended features from both homo- and hetero-fermentative bacteria. Inoculated silages had a greater consumption of soluble carbohydrates. In despite of the lower pH value attributed to the action of homolactic bacteria, treated silages had similar concentrations of lactic acid and higher levels of acetic and propionic acids than control silages. In turn, the presence of weak acids with antifungal power reduced yeast counts and ethanol concentrations.

Silages included in this data set generally were stored for longer than 60 days (85% of evaluated averages). Therefore, it is likely that assimilation of lactic acid by *L. buchneri* strains occurred throughout the fermentation process, giving rise to organic acids derived from the heterofermentative pathways. Puzzling, fermentative losses and aerobic stability were not altered.

#### Optimal dose of additives for improving aerobic stability to HMCS

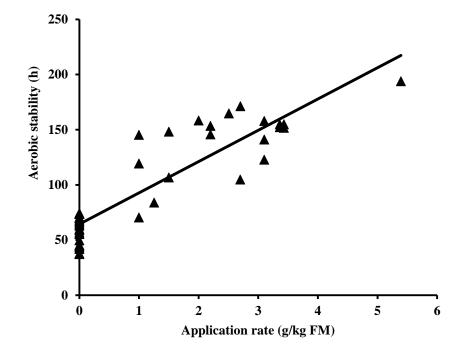
In the current data set, aerobic stability was the most important response improved by additive utilization. Heterofermentative bacteria and chemical additives successfully enhanced aerobic stability of HMCS. For recommending an optimal application rate, a broken-line regression model was fitted to the data set.

For heterolactic bacteria, treatment effectiveness was achieved when bacteria was applied up to  $4.67 \times 10^5$  cfu/g (Figure 1). Similar results were found by Kung and Ranjit (2001) with barley silage, which reported no improvements in aerobic stability when *L. buchneri* was applied at rates lower than  $5 \times 10^5$  cfu/g.



**Figure 1** Aerobic stability of HMCS according to inoculation rate of heterofermentative bacteria. If  $Dose \le 4.67 \times 10^5$  ufc/g, aerobic stability = 64.4 + 28.3 × Dose (g/kg); otherwise, aerobic stability = 235 h. *P* < 0.01, R<sup>2</sup> = 0.50, RMSE = 47.88.

For chemical additives, the aerobic stability increased linearly within the studied range of application rates (Figure 2). Inhibition of spoiling microorganisms (e.g., yeasts and molds) requires a minimum acid concentration in silage aqueous fraction. Organic acids concentrations between 12.5 and 30 g/kg of water may be required to control spoiling microorganisms in feedstuffs with high DM content (Collins, 1995).



**Figure 2** Aerobic stability of HMGS according to chemical additive dosage. Aerobic stability =  $64.4 + 28.3 \times \text{Dose}$  (g/kg). P < 0.01,  $R^2 = 0.81$ , RMSE = 20.14.

For instance, a HMGS with 70% DM would require a dose between 3.8 and 9.0 g/kg of organics acids to control yeasts and molds. In the present data set most of the positive responses were reached with application rates higher than 3.0 g/kg, however, 3.0 to 4.0 g/kg are the most frequent range of application rate. Probably, cost:benefit ratio issue and negative effect on animal responses might be plausible justifications for these lower application rates. The lack of data for higher dosages focusing on aerobic stability in HMGS also contribute for this trend. Extrapolations should be avoided for silages with high moisture content (i.e., whole plant silages) because they typically contain higher levels of fermentation end-products.

#### Additives to HMWCS

Compared to corn, winter cereals have in their composition a substantial content of soluble carbohydrates. This increased availability of fermentable nutrients may impact on the performance of additives during fermentation and feed-out phases. Despite of the scarcity of data for wheat, barley and triticale silages, the consequences of heterofermentative bacteria and chemical additives in winter cereal grain silages will be presented hereafter.

#### Heterofermentative bacteria

The chemical composition, pH and fermentation end products of HMWCS treated with heterofermentative bacteria inoculants are shown in Table 7. The lower DM content of treated silages may be associated with the heterofermentative pattern, evidenced by the increase in acetic acid production. Despite losses have not been measured, the metabolic pathway of acetic acid production leads to carbon losses, which may explain the lower DM content of silages.

Even with an increase in acetic acid levels, ethanol production was not controlled in treated silages. The final content of lactic acid was similar, but the pH of the inoculated silages was lower, highlighting that winter cereals have enough substrate for an effcient acidification of the mass. Anaerobic assimilation of lactic acid performed by *L. buchneri* may explain the similar content of this acid in the silages (Oude-Elferink et al., 2001).

 Table 7
 Data set of high moisture winter cereal grain silages treated without or with heterofermentative inoculants and their effects on silage quality

			Dat	a set		Treatment	effect	SEM	Р
Item	$\mathbf{n}^1$	Mean	SD	Minimum	Maximum	Control	Hetero	SEM	Г
DM, g/kg	18	584.2	66.5	499.0	744.0	618.6	549.8	19.4	0.02
Starch, g/kg DM	18	623.2	43.7	537.0	680.8	631.4	615.0	14.7	0.22
CP, g/kg DM	18	101.2	4.8	93.8	110.0	100.1	102.4	1.59	0.08
WSC, g/kg DM	18	46.6	27.9	15.0	100.0	53.6	39.5	9.26	0.17
pH	18	4.37	0.49	4.00	5.90	4.57	4.17	0.15	0.06
Lactic acid, g/kg DM	18	13.4	5.86	1.80	19.7	12.7	14.1	2.00	0.50
Acetic acid, g/kg DM	18	9.10	10.2	1.00	33.6	3.66	14.5	2.93	0.02
Ethanol, g/kg DM	18	10.4	4.48	5.29	19.4	11.3	9.55	1.51	0.27
Lactic: Acetic ratio	18	2.21	1.48	0.35	6.00	3.06	1.36	0.41	0.02

<sup>1</sup> Number of means.

#### Chemical additives

The changes imposed on HMWCS by adding chemical additives are shown in Table 8. The starch disappearance in silages treated with additives may have been favored by chemical solubilization of the protein matrix. With the release of the granules primarily waxy, there was attack on the starch molecules. However, the agents promoting such breakage are not easily identifiable, since the chemical compositions are diversified and there were no amylolytic enzymes present in the products.

Crude protein,  $N-NH_3$  and pH were directly influenced by applying chemical additives and their composition. Nitrogenous compounds added to silages certainly contributed to increase CP and  $N-NH_3$  concentrations. In addition, the chemical additives increased the buffering capacity and impaired the pH drop. Moreover, the lower concentration of lactic acid in the treated silages suggests that the fermentation process was suppressed. The chemicals were efficient in controlling fungi and yeast growth, reducing the formation of ethanol. The antifungal activity has also proven been effective at the feedout phase, increasing the aerobic stability of the grains.

 Table 8
 Data set of high moisture winter cereal grain silages treated without or with chemical additives and their effects on silage quality

			Dat	a set		Treatmen	nt effect	CEM	ת
Item	$n^1$	Mean	SD	Minimum	Maximum	Control	Chemical	SEM	Р
DM, g/kg	52	641.6	67.4	527.0	756.0	639.6	643.6	13.3	0.35
ADF, g/kg DM	10	63.8	11.9	46.3	82.0	62.3	65.3	5.59	0.61
Starch, g/kg DM	40	608.7	34.1	532.0	679.8	614.3	603.1	7.62	0.07
CP, g/kg DM	48	118.8	29.8	94.6	221.0	111.7	125.9	5.96	0.02
N-NH <sub>3</sub> , g/kg DM	12	0.325	0.23	0.20	0.90	0.28	0.37	0.09	0.04
WSC, g/kg DM	42	45.5	25.3	16.8	100.0	43.9	47.1	5.57	0.57
рН	52	5.16	1.44	3.80	9.20	4.73	5.59	0.27	0.01
Lactic acid, g/kg DM	52	11.0	10.5	0.88	40.0	12.6	9.41	2.05	0.07
Acetic acid, g/kg DM	52	3.55	3.70	0.10	21.7	3.16	3.95	0.73	0.35
Ethanol, g/kg DM	50	8.70	5.91	0.02	20.8	12.0	5.45	0.99	< 0.01
Lactic: Acetic ratio	52	4.59	4.15	0.37	21.6	4.50	4.68	0.82	0.86
Yeasts, log cfu/g	10	4.88	1.95	1.50	6.80	6.44	3.32	0.50	0.01
Molds, log cfu/g	10	4.40	2.34	1.70	6.90	5.86	2.94	0.84	0.03
DM losses <sup>2</sup> , g/kg	4	31.5	15.2	16.0	45.0	33.0	30.0	13.0	0.37
Aerobic stability, h	6	223	78	87	301	165	281	29	0.10

<sup>1</sup> Number of means, <sup>2</sup> Fermentative losses.

#### Performance of dairy and beef cattle fed grain silages with additives

In order to characterize the productive responses of ruminants fed silages treated with additives, two experiments with dairy cows were developed by the Forage Quality and Conservation Team of the Luiz de Queiroz College of Agriculture (University of São Paulo, Piracicaba, SP, Brazil). Other two experiments with beef cattle were carried out at São Paulo State University (Colina, SP, Brazil). All trials have not yet been published in full, but represent a broad approach to the use of additives in sorghum and corn grain silages.

#### Performance of dairy cows fed rehydrated sorghum grain silage treated with L. buchneri or sodium benzoate

The aim of this study was to evaluate the performance of dairy cows fed rehydrated sorghum grain silage treated with sodium benzoate or *L. buchneri*. Sorghum grain (Biomatrix, BM 737) was ground and rehydrated to reach 350 g/kg of moisture. The treatments were dry ground sorghum (DGS), rehydrated sorghum grain silage inoculated with *L. buchneri* at  $5 \times 10^5$  cfu/g FM (LB), rehydrated sorghum grain silage treated with sodium benzoate at 2.0 g kg-1 FM (Benz) or rehydrated sorghum grain silage without additives (Control). The silages were stored for 60 days prior opening. Cows were housed in free stall barns and fed twice daily. Twenty Holstein cows ( $31.5 \pm 5.8$  kg milk day-1 and  $92 \pm 75$  days in milk) were blocked based on milk yield and assigned to one of the four treatments in replicated 4x4 Latin square, with 21-day periods, being 14 days of adaptation and seven days for sample collection. Experimental diets had 380 g/kg corn silage, 90 g/kg Tifton haylage, 100 g/kg whole cottonseed, 175 g/kg soybean meal, 25 g/kg minerals and 230 g/kg sorghum grain silages or dry ground sorghum. Data were analyzed with the proc mixed of SAS and treatments compared by orthogonal contrasts: DGS *vs.* Ensiled; Control *vs.* (LB + Benz) and LB *vs.* Benz.

As show in Table 9, the DM intake was lower for silages compared with dry ground sorghum, however, there was no difference among silages treated with additives. These results suggest that less energy was available in DGS diets, which can be proven by the lower NFC and starch digestibility in those grains. Milk yield was not affected by treatments. As consequence, feed efficiency was 6.8% higher for diets containing ensiled compared with dry sorghum. Milk composition only differed in nitrogen urea content between sorghum grain silages and dry ground sorghum, suggesting greater efficiency of N coupling into the rumen, probably due to the more digestible starch source as grain silages were fed.

It was concluded from this study that the processing of sorghum grain was the predominant factor affecting the performance of dairy cows. Although the doses of sodium benzoate and *L. buchneri* adopted in the current study consistently improve the aerobic stability of HMGS, no effects were observed in animal performance.

		Treat	ment				Р	
					_	$DGS \times$	Control ×	$LB \times$
Item	Control	Benz	LB	DGS	SEM	Ens.	(LB + Benz)	Benz
DMI, kg/d	202.0	203.3	201.0	213.2	4.7	< 0.01	0.75	0.40
NFC digestibility, %	93.5	93.3	92.7	89.1	0.75	< 0.01	0.56	0.55
Starch digestibility, %	87.2	86.8	86.9	79.1	0.88	< 0.01	0.73	0.86
Milk, kg/d	32.47	31.57	32.10	31.78	11.2	0.57	0.21	0.37
3.5% FCM, kg/d	31.84	31.30	31.27	32.04	12.3	0.33	0.37	0.96
Fat, g/kg	33.9	34.6	33.5	35.5	1.2	0.07	0.85	0.25
Fat, kg	1.09	1.08	1.07	1.12	0.05	0.13	0.60	0.65
Protein, g/kg	31.1	31.3	31.0	30.8	0.5	0.25	0.98	0.42
Protein, kg	1.00	0.97	0.98	0.96	0.02	0.18	0.21	0.58
MUN, mg/dL	16.6	16.8	16.4	17.7	0.4	< 0.01	0.93	0.28
Milk/DMI	1.61	1.56	1.60	1.49	0.05	< 0.01	0.31	0.14
Milk NE <sub>L</sub> /DMI, Mcal/kg	1.07	1.05	1.06	1.02	0.02	0.02	0.47	0.50

 Table 9
 Performance and feed efficiency of dairy cows fed dry ground sorghum or rehydrated sorghum grain silages with or without additives

#### Performance of dairy cows fed rehydrated corn grain silage treated with sodium benzoate

This research aimed to evaluate the effects of sodium benzoate in rehydrated corn grain silages, as well as evaluate the replacement of dried corn (DGC) for rehydrated corn grain silages on the performance of dairy cows. Ground corn grains from a commercial source of flint hybrid with initial DM of 915 g/kg FM was rehydrated to reach 650 g DM/kg FM, using only water to control silages (Control) or water plus sodium benzoate to a final dose of 2g/kg as fed (Benz). Silages were stored in 200-L plastic barrels for 170 d. Diets contained (DM basis) 380 g/kg corn silage, 100 g/kg Coast-cross hay, 207 g/kg soybean meal, 115 g/kg dried citrus pulp, 25 g/kg premix mineral + vitamins, and 173 g/kg corn grains (DGR, Control or Benz).

Eighteen Holstein cows ( $\pm$  30 kg milk /d and  $\pm$  259 days in milk) housed in a tie-stall barn were assigned to a replicated 3 × 3 Latin square design, with 21-d periods. Dry matter intake (DMI), milk yield and composition were recorded from d-15 to d-21 in each experimental period. Analysis of variance was performed using the Mixed procedure of SAS and treatment means compared by orthogonal contrasts adjusted by Tukey Kramer, to test DGC × (Control + Benz) and Control × Benz.

Dry matter intake was similar across treatments, whereas starch and CNF digestibility was higher for fermented grains (Table 10). The higher energy of silages when compared to dry grain tended to increase the production of 3.5 % fat-corrected milk. Therefore, animals fed rehydrated grain silages showed a trend for increased feed efficiency. Dairy cows fed silages had lower excretion of milk urea nitrogen, suggesting a better use of nitrogen for microbial protein synthesis. Other components of milk were not affected by the diets, however, protein content tended to be higher with fermented grains. Treating rehydrated corn grain silage with sodium benzoate did not affect animal performance.

		Treatmen	nt		Р	
-					DGC ×	Control ×
Item	Control	Benz	DGC	SEM	(Control + Benz)	Benz
DMI, kg/d	21.1	20.9	21.4	0.79	0.41	0.80
NFC digestibility, %	91.4	91.6	88.1	0.01	< 0.01	0.87
Starch digestibility, %	93.8	94.9	92.9	0.08	0.05	0.18
Milk, kg/d	28.7	29.0	27.4	1.74	0.14	0.75
3.5% FCM, kg/d	29.4	29.6	28.0	1.88	0.10	0.82
Fat, g/kg	36.1	36.1	38.0	2.50	0.36	1.00
Fat, kg	1.05	1.05	1.00	0.08	0.19	0.92
Protein, g/kg	36.2	35.5	35.5	0.80	0.62	0.42
Protein, kg	1.05	1.03	0.95	0.06	0.06	0.72
MUN, mg/dL	11.9	11.9	13.0	0.50	0.01	0.91
Milk/DMI	1.35	1.36	1.31	0.06	0.13	0.86
3.5% FCM/DMI	1.39	1.38	1.31	0.06	0.10	0.84
Milk NE <sub>L</sub> /DMI, Mcal/kg	0.97	0.96	0.91	0.04	0.07	0.71

 Table 10
 Performance of dairy cows fed dry ground corn or rehydrated corn grain silages with or without sodium benzoate

# Performance of finishing bullocks fed high moisture or rehydrated corn grain silages treated with L. buchneri

One-hundred and eighty Nellore bullocks  $(310 \pm 17 \text{ kg BW})$  were blocked by BW body weight and housed in 30 pens  $(4.15 \times 15 \text{ m})$  with 6 animals/pen (6 pens/treatment). The animals were confined for 119 d, being 35 d for adaptation and 84 d for comparison of treatments. The experimental diets were iso-nitrogen (crude protein = 141 g/kg DM) and contained sugarcane bagasse (125 g/kg DM), dried citrus pulp (170 g/kg DM), peanut meal (40 g/kg DM), mineral mix (30 g/kg DM), urea (10 g/kg DM), and one of the following corn sources (625 g/kg DM): 1 ) dry ground corn (DGC), 2) high moisture corn silage (HMCS), 3) HMCS treated with *L. buchneri* at  $1 \times 10^5$  cfu/g (HMCS-LB), 4) rehydrated corn grain silage (RCGS), and 5) RCGS treated with *L. buchneri* at  $1 \times 10^5$  cfu/g (RCGS-LB). Corn grain silages had approximately 650 g/kg of DM. Animals were fed twice daily (7:30 and 13:30 h) in equal amounts, adjusted to provide 103% of the previous intake.

**Table 11** Performance and carcass traits of feedlot Nellore fed dry ground corn (DGC), high moisture corn silage (HMCS), HMCS treated with *L. buchneri* at  $1 \times 10^5$  cfu/g (HMCS-LB), rehydrated corn grain silage (RCGS) and RCGS treated with *L. buchneri* at  $1 \times 10^5$  cfu/g (RCGS-LB)

Item	DGC	HMCS	HMCS-LB	RCGS	RCGS-LB	SEM	Р
DMI, kg/d	9.81ª	7.88 <sup>b</sup>	7.89 <sup>b</sup>	7.34°	7.32 <sup>c</sup>	0.24	< 0.01
Initial BW, kg	311	310	310	311	310	7.26	0.24
Final BW, kg	514 <sup>a</sup>	505 <sup>ab</sup>	510 <sup>a</sup>	505 <sup>ab</sup>	493 <sup>b</sup>	7.17	0.08
Average daily gain, kg	1.73 <sup>a</sup>	$1.66^{ab}$	1.69 <sup>a</sup>	1.64 <sup>ab</sup>	1.55 <sup>b</sup>	0.05	0.09
Feed efficiency	0.18 <sup>c</sup>	0.21 <sup>b</sup>	0.21 <sup>ab</sup>	0.22ª	0.21 <sup>ab</sup>	0.01	< 0.01
Hot carcass weight, kg	290 <sup>a</sup>	$282^{ab}$	286 <sup>a</sup>	$282^{ab}$	275 <sup>b</sup>	6.49	0.06
Dressing, %	56.3	55.9	56.2	56.0	55.9	0.31	0.84
Ribeye area, cm <sup>2</sup>	76.0 <sup>b</sup>	75.5 <sup>b</sup>	$78.0^{\mathrm{a}}$	79.6ª	75.3 <sup>b</sup>	2.60	< 0.01
Ribeye area, cm <sup>2</sup> /100 kg BW	15.0 <sup>b</sup>	15.2 <sup>b</sup>	15.5 <sup>b</sup>	16.2ª	15.5 <sup>b</sup>	0.34	0.02
Marbling score	2.75	2.93	2.85	2.51	2.80	0.12	0.12
Fat thickness, mm	5.55	6.20	5.81	5.90	5.59	0.21	0.23
Fat thickness, mm/100 kg BW	1.09 <sup>b</sup>	1.24 <sup>a</sup>	1.16 <sup>b</sup>	1.20 <sup>ab</sup>	1.15 <sup>b</sup>	0.04	0.08

<sup>a,b</sup>Means within a row with different superscripts differ (P < 0.05)

Ensiled grains resulted in lower DMI, whereas weight gains were similar or slightly lower than that of animals fed dry corn based diets (Table 11). Therefore, cattle fed grain silages had higher feed efficiency. Moreover, carcass traits such as ribeye area and backfat thickness have been improved or were similar to dry corn fed animals. Within each type

of silage, inoculation with *L. buchneri* resulted in similar production responses without systemic changes in carcass traits. Backfat thickness standardized to BW (mm/100 kg BW) was lower for treatments with inoculant, however, the degree of marbling was not affected by treatments.

#### **Final remarks**

Control of fermentative losses is not a concern in properly made HMGS. Therefore, use of additives is justified if aerobic stability is improved. Additives based on chemical or heterofermentative bacteria proven to be effective in prevent aerobic deterioration in a same magnitude. Overall, treatment effectiveness was achieved when chemical additives were applied to a rate of at least 3 g/kg as fed and heterolactic bacteria up to  $5 \times 10^5$  cfu/g FM.

Experiments with cattle fed corn and sorghum grains showed that *L. buchneri* and sodium benzoate did not change animal performance. This reinforces that the promotion of aerobic stability is the main feature being sought with application of additives in HMGS.

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# SESSION 1 – PRODUCTION OF FORAGE CROPS

# Oral presentations

<u>Recent developments in evaluation of grassland swards in the context of forage conservation</u> Hopkins, A.

<u>The quality and nutritive value of meadow hay as a horse feed from western region of Slovakia</u> Gálik, B., Bíro, D., Rolinec, M., Juráček, M., Šimko, M., Herkel', R., Hanušovský, O.

<u>Phenotype studies to select "*Lfy*" architecture maize silage hybrids</u> Pinter, J., Tothne Zsubori, Zs., Nagy, Z., Pok, I., Spitko, T., Berzy, T., Szoke, Cs., Glenn, F. B., Marton, Cs. L.

Influence of dry weather on maize production in 2015 Loučka, R., Homolka, P., Jančík, F., Kubelková, P., Tyrolová, Y., Výborná, A.

White and narrow-leafed lupine as an alternative source of quality forage Jančík, F., Kubelková, P., Koukolová, M., Homolka, P.

# Poster presentations

<u>Changes in dry matter production of abandoned grassland throughout its revitalization</u> Hanzes, Ľ., Britaňák, N., Ilavská, I.

<u>Changes in energy value of permanent grassland in area of Nitra city</u> Rolinec M., Bíro D., Šimko M., Juráček M., Gálik B.

<u>Productive ability and nutritive value of alfalfa (Medicago sativa l.) and its simple mixtures when grown in a mountainous region</u> Ilavská, I., Jančová, M., Hanzes, Ľ., Britaňák, N., Pollák, Š.

<u>Effect of silage hybrid on maize forage quality</u> Bíro D., Juráček M., Šimko M., Gálik B., Rolinec M., Herkel' R., Hanušovský O., Piterka P., Píšová A., Hatala L.

<u>Chemical quality changes during ripening of silage maize hybrids</u> Tóthné Zsubori, Zs., Pintér, J., Spitkó, T., Szőke, Cs., Nagy, Z., Berzy, T., Marton, L. Cs.

Effect of maize cutting height on its nutritive value Férard A., Uijttewaal A., Meslier E., Kardacz P.

<u>The effect of harvest date and fermentation process on the levels of carotenoids and tocopherols in virginia fanpetals (Sida hermaphrodita) herbage and silage</u> Antoszkiewicz, Z., Fijałkowska, M., Purwin, C., Mazur, M.

Ruminal degradability of dry matter and crude protein from virginia fanpetals (Sida hermaphrodita rusby l.) herbage and silage Purwin, C., Fijałkowska, M., Nogalski, Z., Lipiński, K., Michalski, J. P.

Forage production from genetically modified crops Chrenková, M., Pomikalová, S., Polačiková, M., Chrastinová, Ľ., Formelová, Z., Rajský, M., Mlynár, R.

# Recent Developments in Evaluation of Grassland Swards in the Context of Forage Conservation

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#### Introduction

Planning of the supply of feed resources on farms and requires accurate information on the amounts and quality of forage at the point of harvesting for silage or hay. Although techniques for assessing forage mass (by sampling, drying and weighing) have been available to researchers for decades these are seldom applied in farm situations. Similarly, assessments of forage quality are more likely to be made (if at all) on harvested and ensiled forage, later on the conserved product rather than before harvesting. There is a need to ensure that conserved forage meets the maximum possible proportion of the feed requirements of housed livestock, and also that inputs, such as N fertilizer, are utilized to maximum efficiency. Readily available information for farmers and extension services is necessary on both the quantity per hectare and feed value of forage for optimum planning of harvesting schedules. The paper considers the role and potential applicability of advances in methodologies and technologies including: non-destructive sampling, Normalised Difference Vegetative Indices, hyperspectral imaging, predictive models, spatial data sourcing using GIS, and remote sensing including use of drones and satellite data.

# Advancement in techniques for non-destructive sampling

Traditional 'destructive sampling' methods, as used on research farms for herbage production assessment involve the cutting and weighing of accurately sampled herbage from a precise area. If replicated adequately the same approach can be applied at a farm scale but is likely to be inappropriate because of labour requirements. Indirect non-destructive assessments include sward surface height (SSH) and compressed herbage mass (HM). The use of rising plate meters (Castle, 1976) and sward-height measurement sticks dates to the 1970s (Davies, 1993). Further advances and commercial production of electronic rising plate meters (RPM) and automatic recording sticks (http://jenquip.co.nz) have led to these becoming tools for measuring SSH or HM in swards by farmers and extension workers, as well as in research. Their use requires calibration and may be inappropriate on some heterogenous swards, as well as being time-consuming on large farms. Devices such as the C-DAX Pasturemeter (PM) (http://www.pasturemeter.co.nz) which can be attached to aquad-bike, can further reduce the workload. A recent report on the verification of the estimation accuracy of the C-DAX PM compared with a plate meter (RPM), calibrated against measured mown and weighed areas of the sward (Schori, 2015). This technique gave similar quality estimations to the RPM, with potential for adoption for other plant communities subject to further calibrations.

#### Evaluation of herbage nutritive value

A major challenge for forage-based livestock production is to ensure sufficient dry mass (DM) and quality (energy and protein) either throughout the entire year or to ensure a seasonal surplus for use as conserved feed (silage/ hay) during feed-deficit periods. Among recent developments and trends are the following. The in-vitro gas production (IVGP) technique is a non-invasive method, requiring only small amounts of sample, for estimating degradation rates of forages and other feedstuffs (Dijkstra *et al.*, 2005) and has been used to predict total digestible nutrients (TDN) (Rymer *et al.*, 2005; Aguiar *et al.*, 2011). For example, from the results of a 4-year study with warm-season perennial grasses, Aguiar *et al.* (2011) obtained empirical relationships between forage chemical composition and IVGP fermentation parameters, and also developed this for calculating TDN. Based on these developments it was proposed that this technique has potential to assist producers improve animal productivity and to support grazing management decision-making for using warm-season forages.

Although the evaluation of herbage nutritive value has mainly focussed on the main aspects of feed value linked to digestible organic matter and crude protein content, emphasis has also moved to the role of secondary compounds, such as tannins, and on the balance of fatty acids. In the case of tannins, a number of forage species are attracting interest for their potential to be regarded as beneficial rather than anti-nutrional (see review of Piluzza *et al.*, 2013). Attention focusses on the potential of tannin-containing species to mitigate methane emissions and to improve nutrient-use efficiency and reduced N emissions. The increasing interest in the fatty acid composition of pasture species is linked to their nutrional effect on meat or dairy produce, particularly through increasing the *n*-3 PUFA content. Green forage is high in 18:3*n*-3 and can increase delivery of *n*-3 PUFA through the ruminat into milk and meat (Morgan *et al.*, 2012, and references therein).

#### Use of models and indices

Models have been widely used in agriculture generally and notably so in grassland (Thornley and France, 2006). In addition to the Hurley Pasture Model (Thornley, 1998) and the patch-scale models of Schwinning and Parsons (1999), more recent developments that have attracted considerable interest in terms of pasture evaluation. First, the DairyMod and EcoMod (Johnson *et al.*, 2008) are biophysical pasture-simulation models that were developed for Australia and New Zealand, but have been applied to simulations of greenhouse gas emissions, interannual growth of pasture swards (Chapman *et al.*, 2009) and herbage mass accumulation of tall fescue in Argentina (Berger *et al.*, 2014). Second, the GrazeIn model, which has been developed to predict intake of dairy cows at pasture, including under rotationally stocked and continuously stocked swards (Faverdin *et al.*, 2011; Delagarde *et al.*, 2011a; b) and is still the subject of further work on improving its accuracy and usability.

Economically based evaluation system for forages in dairy systems have been established by McEvoy *et al.* (2011, 2014). The use of the grass economic index allows cultivars to be assessed in terms of their economic value. In essence, the system ranks cultivars for key traits using cultivar performance values linked to economic values.

## Integration new technologies in forage evaluation

## **Remote sensing**

The use of GIS and application of remote sensing has been a major advance particularly in mapping of large areas in terms of their suitability for particular forage crops, taking account of layers of data including climate, soils, slopes, environmental problems (Rossini et al. 2012). For example, Hannaway *et al.* (2005) reviewed this application in the context of evaluating the potential of a range of forage species in two large countries, China and Australia. GIS also allows for studies of interactions between grazing animals and pastures at a range of scales. For example, Rutter (2007) described how GPS, used for spatial recording of grazing animals, can be used with GIS to evaluate pastures and grazing behaviour at a range of scales from the patch to the landscape level.

The low cost and availability of UAV (Drones) which can be used to photograph at small scales, as well as carry small instruments, has also opened up new opportunities for recording information on experimental areas and also on large farmland area where ground access may be time consuming. Satellite technologies from major companies like Airbus 'One Tasking' now offer the agricultural industry cost-effective access to an Airbus Defence and Space satellite 'all from the comfort of their own desk or when out in the field' to high-resolution and wide-swath satellite sensors. (http://www.intelligence-airbusds.com/one-tasking/)

#### Hyperspectral imaging and chlorophyll and N prediction and vegetation indices

Hyperspectral imaging has also been used to assess the health and feed value of crops in the field; e.g. Ferwerda (2005) used this method to predict chlorophyll and nitrogen content using indices based on red and infrared bands (there is a positive correlation between leaf chlorophyll and N content). Assessment of N (or CP) content of forage is important for both animal production and reducing N losses. Therefore, non-destructive chlorophyll-measuring devices such as the chlorophyll content meter (CCM), have received acceptance as measurement tools to determine leaf chlorophyll content and aid in nitrogen management in agricultural crops (Hughes et al., 2016). These authors report on evaluation the Fieldscout CM 1000 NDVI and Yara N-Tester as easy-to-use and cost-effective tools for predicting foliar chlorophylls (a, b and total) and CP concentrations in herbage of different grass species, and found that optical chlorophyll measurements and CP concentrations were highly correlated with both types of instrument (Hughes et al., 2016).

Vegetation indices are widely used as model inputs and for non-destructive estimation of biomass and photosynthesis but only recently have relationships between normalized difference vegetation index (NDVI), leaf area index (LAI), brown and green above-ground biomass and photosynthesis potential (PP) been investigated (Metzger et al., 2016). Biomass and LAI were found to be correlated poorly, with high species-specific variability, but intensive meadows had a higher ratio of LAI to biomass than extensive grasslands. Using NDVI instead of LAI could reduce uncertainty in photosynthesis models (Metzger et al., 2016).

#### Conclusions

Given the challenges of food security and uncertainties in response to climate change, the use of forage is likely to assume increased importance. This will require increasing attention to detail not only in terms of quality of conservation but in ensuring that harvested forage is utilised at its optimum stages of growth and quality, and consistent with efficient utilisation of nutrients. The examples reviewed here indicate that many new technologies are becoming available to extension services, supply companies, researchers and farmers.

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# The Quality and Nutritive Value of Meadow Hay as a Horse Feed from Western Region of Slovakia

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#### Abstract

The aim of the study was to analyze the nutritive value of meadow hay collected from different farms of western region of the Slovak Republic. A total 40 samples of meadow hay were analyzed in Laboratory of Quality and Nutritive Value of Feeds (Department of Animal Nutrition, Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra). Content of dry matter in analyzed hay samples ranged between 820.40 and 949.60 g/kg. In analyzed samples, average crude protein content 81.90 g/kg of dry matter was analyzed. In ADF content, the highest value of 44.45% and NDF of 71.76% were detected. However, the lowest ADF content was analyzed at the level 29.40% and NDF 49.93%. Content of ash in analyzed samples were between 48.70 and 113.10 g/kg of dry matter (75.12 g/kg of dry matter in average). Content of organic matter ranged from 886.90 to 951.30 g/kg of dry matter (average value 924.88 g/kg of dry matter). Energy value (as digestible energy content) was in analyzed hay samples from 6.70 to 8.65 MJ/kg, in average 7.66 MJ/kg.

#### Introduction

Hay is dry forage with typical higher dry matter content and higher crude fibre content (depending of forage species and maturity stage during the harvest time). There are many changes in herbage during the vegetation, from nutritive point of view, mainly in dry matter, crude protein and crude fibre and energy contents (Tallowin and Jefferson, 1998; Gálik et al., 2011). However, hay is a very important feed in ruminants and horses nutrition (Gálik, 2012).

#### **Material and Methods**

A total 40 samples of meadow hay were analyzed in Laboratory of Quality and Nutritive Value of Feeds (Department of Animal Nutrition, Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra). Analyzed samples were collected from different farms in western region of the Slovak Republic. All samples were from hay conserved in natural way at the boot vegetation stage. For chemical analysis, standard laboratory methods and principles were used (AOAC, 2000). After the sensory evaluation, laboratory samples were prepared (predrying at the 60°C, grinding for 1 mm particles). Content of dry matter was analyzed with drying  $(103\pm2^{\circ}C)$ , content of crude protein according Kjedahl method, content of crude fat according Soxhlett-Henkel principle, content of crude fibre according Hennenberg-Stohmann, fractions after the hydrolysis (in acidic solution for ADF or in neutral solution for NDF). Content of ash was determined after the burning (temperature  $530\pm20^{\circ}C$ ). Content of digestible energy for horses was calculated with formula of Zeyner, Kienzle (2002):

#### DE (MJ/kg) = -3.6 + 0.211 x %CP + 0.421 x %F + 0.015 x %CF + 0.189 x %NFE

To calculate basic statistic characteristics (mean, standard deviation) the SAS statistical package was used (SAS Inc., New York City, U.S.A.).

### **Results and Discussion**

Content of nutrients and digestible energy for horses are shown in Table 1. In analyzed hay samples, we detected content of dry matter between 820.40 and 949.60 g/kg. The average crude protein content in analysed hays was 81.90 g/kg of dry matter (in samples from 50.80 till 125.70 g/kg of dry matter). Schellberg et al. (1998) reported that crude protein in meadow hay is affect by many agroclimatic factors, and these authors found in meadow hay crude protein content between 116 and 120 g/kg of dry matter. Turgut et al. (2008) reported that for meadow hay harvested in early-bloom or late-bloom stage is typical crude protein content 8% or 6% respectively. The highest crude fat content was analyzed at the level 24.80 g/kg of dry matter.

	DM	СР	F	CF	ADF	NDF	Α	ОМ	DE
	g/kg		g/kg of DM		% of DM		g/kg of DM		MJ/kg
Mean	902.35	81.90	16.0	330.95	36.56	59.06	75.12	924.88	7.66
S.D.	31.87	20.52	4.67	29.95	3.72	4.80	16.78	16.78	0.60
Min.	820.40	50.80	8.30	274.80	29.40	49.93	48.70	886.90	6.70
Max.	949.60	125.70	24.80	388.50	44.45	71.76	113.10	951.30	8.65

 Table 1
 Nutritive value of analysed meadow hay

DM: dry matter, CP: crude protein, F: fat, CF: crude fibre, ADF: acid detergent fibre, NDF: neutral detergent fibre, A: ash, OM: organic matter, DE: digestible energy for horses, S.D.: standard deviation.

**BACK** 

Content of crude fibre in hay is the marker of the quality. However, from nutritional point of view, hay is very important source of crude fibre for ruminants and horses (Gálik et al., 2011; Bíro et al., 2014). The highest crude fibre content in analysed hay samples was 388.50 g/kg of dry matter, and the lowest 274.80 g/kg of dry matter. Petrikovič et al. (2000) published the average crude fibre content in meadow hay between 283 and 354 g/kg of dry matter. However, higher crude fibre content than 30 % in hay is indicator of late harvest, mostly after the blooming stage (Gálik et al., 2011; Biro et al., 2014). Also Chesson et al. (1995) published, that cutting of grasses when they aren't an advanced state of phenological maturity means, that there will be a high proportion of lignin and structural carbohydrates in the harvested dry matter. This point of view confirmed our results, in hay samples with higher crude fibre content than 36 %, content of ADF higher than 40 % was found. In analysed hay samples, the average NDF content ranged from 49.93 to 71.76 %. Content of organic matter in analysed hay samples ranged between 886.90 and 951.30 g/kg of dry matter (the average value 924.88 g/kg of dry matter). Meadow hay is an important feed in horses feed rations (Gálik et al., 2011; Biro et al., 2014). It is very important source of crude protein, crude fibre and minerals. Meadow hay with high quality can have energy value higher than 9 MJ/kg of digestible energy in dry matter. In analysed hay samples, digestible energy content between 6.70 and 8.65 MJ/kg of dry matter. Content of digestible energy, as well as the content of crude protein and crude fibre is affect mainly by maturity stage of grassland during the harvest (Biro et al., 2014).

## Conslusions

The aim of the study was to analyse the nutritive value of 40 samples meadow hay collected from different farms from western region of the Slovak Republic. Different nutritive values of samples were found out. For hay samples with high quality were typical higher crude protein content, lower crude fibre and ADF content and higher value of digestible energy.

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# Phenotype Studies to Select "Lfy" Architecture Maize Silage Hybrids

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BACK

Silage maize is one of the most important forage for ruminant feeding in the whole World. The hybrids which were used before were generally characterized by high amount of green mass with large proportion of dry matter. Most of these hybrids were bred for dual purposes. In the practice unfortunately - there is still present an another theory ; hybrids which are good for grain they are good for silage, too. Later on the breeders tried to find another usable traits in order to improve the quality e.g. with using BMR materials – more or less success. Nowdays, when the big dairy farms - with intensive plant production background - want to produce more and more Kgs of milk from a unit area, they try to request from the breeding companies and institutions to develop new type of maize hybrids which were bred specifically for production top yielder and high quality silage with the best digestibility.

The dominant Lfy1 mutant geen was discovered in the USA in the early 1980 years. It transforms the architecture of the maize plant by significantly increasing the number of leaves above the ear .It is – of course together with the ear - the most valuable part of the plant. The advantage of this green will be manifesting in higher photosynthetic area, changes the whole canopy structure of the plant, so the hybrid, which contain it will produce much bigger amount of good quality green mass. The greater ratio of fresh and stay-green leaves in the total plant dry matter (DM) have significantly beneficial effect on the silage quality, firs of all in the digestibility. Modifying the morphology of a plants –trough changes the ratio of different plant parts only – itself doesn't influence the biomass quality. The firs results, when we used a Lfy inbred line as partner in our new experimental silage maize combinations, we have been found more useful changes in the whole canopy ; beside the enhanced LA, the ear position (setting) became lower, so the stability of plants are stronger and not least, the texture of the kernels became more soft than the conventionals'.

Fourteen years ago, in 2002 the first leafy silage maize hybrid of Martonvásár Institute was registered in Hungary under the name of Kamasil. In this category this was the first in Europe. Since then, seven additional Martonvásár silage hybrids have been registered and added to the European Comission's Plant Variety List. These result are coming from the collaboration between of Agricultural Institute of the Agricultural Research Center of the Hungarian Academy of Sciences, Martonvásár, Hungary and Glenn Seed Ltd, Canada.

The maturity FAO maturity of these hybrids are ranging 240 to 700 and names as follows : Limasil, Dunasil, Massil, Megasil, Lactosil, and Classil). Several of these hybrids have been popular with dairy farmers because of high yield, high fiber digestibility, good starch content and high rumen available starch related to the softer, more floury kernel texture. Compared to conventional silage maize hybrids, these leafies have significantly more leaves above the ear (10 to 12 or even more), so the proportion of them in the total plant dry matter is verifiable higher.

In our 3 years-long phenotyping experiment we have found that the Lfy hybrids comparison to the non-leafy genotype averages 1. *in plant height 2. in leaf number above the ear and 3. leaf area above the ear(just as in total)* are significantly better. One of our best Lfy hybrid (Massil) on the basis of this 3 years experiment produced 18.89 (total), 10,59 leaves above the ear, compared to the Stds' average where the total leaf number were14,38 pcs, and 6,58 above the ear. The advantage of the Lfy hybrid in total leaf area was more than 30 % and above the ear was 50 %.

Over the past decade the question of the renewable energy became more important in Europe. Silage maize can be a possible source of "green" energy. Especially the Lfy hybrids, owing to their phenotypic characters, produce large dry matter yield. At Martonvásár, research has started to confirm whether the leafy phenotype and plant densities that are most suitable for best silage for dairy cattle are also the canopy architecture and planting populations that would yield the best biogas yields. In our earlier studies, a strong positive correlation was found between the biogas production and the starch content, so the phenotype studies of these hybrids from other aspects (such as optimal plant density and harvest time) have been continued. It was found that in case of these Lfy genotypes, in order to achieve the highest biomass yield, we have to find the optimum plant density of each genotype to give them the opportunity to "open their sun collector". Both in the conventional and Lfy hybrids the high plant density significantly reduced the leaf area above the ear, especially in case of Lfys, where the leaf area reduction was 0,49 to 0,39 qm/plant compared to 0,28 to 0,23 qm/plant in the conventional silage.

On the basis of our experiment we can make the following conclusions :

- 1. Breeders are looking for new quality parental inbreds to develop not only high yielder, but high value hybrid to produce "top quality" forage;
- 2. It is not easy to develop an "ideotype" hybrid which meets the needs of both silage growers and users;
- 3. According to the silage grower experience, using the leafy hybrids they are able to reach higher digestible nutrition;
- 4. *Lfy* hybrids : faster rate of leaf area development;
- 5. More leaves (and more younger leaves) above the ear;
- 6. The total canopy structure of them have about an extra 40 % efficient leaf area, this significantly greater photosynthetic leaf area provides the ear with more carbohydrates, which increase the potential dry matter accumulation;
- 7. Higher starch and sugar content higher nutrition value;
- 8. Lower lignin content and soft kernel structure more efficient digestibility;
- 9. It is important to study the relationship between the canopy structure, light distributing and the photosynthesis;
- 10. Lfy hybrids should be used for production of renewable energy : biogas.

Further studies are requested on the field of plant phenotyping of the *Lfy* hybrids to achieve better quality of silage maize hybrids and using of them as green energy.

Keywords: Lfy, maize silage, phenotyping, canopy structure, biogas

# Influence of Dry Weather on Maize Production in 2015

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#### Introduction

Dry weather in the vegetation period can have a significant impact not only on the yield of maize but also on its quality. During fermentation, biochemical processes take place in maize silages, accompanied by increasing silage temperatures caused by insufficiently displaced air. With increasing proportion of dry matter, the risk of higher content of residual oxygen in the ensilaged matter grows. The biochemical processes accompanying the increasing temperatures lead to losses of energy and of nutritional value of the silages (Whitlock et al., 2000). Further losses may occur due to insufficient aerobic stability of the silage after opening the silo, which can lead to a decreased consumption of the silage (Filya and Sucu, 2010). The year 2015 was very dry and warm. The extremely low precipitation in the summer period, combined with high temperatures, had a very negative impact primarily on maize. The dry matter in the harvested maize was higher than is usual for ensilaging, as the plants dried very quickly in the field, although the grain often was in the milk phase or only at the beginning of the milky-waxy ripeness. Therefore maize was harvested with high content of fibre in the dry matter and with lower content of starch, often also with higher content of dry matter.

The study was aimed at ascertaining the effect of use of selected biological and chemical ensilaging additives on the quality of the silages of maize affected by drought during the vegetation period.

#### **Materials and Methods**

The maize was ensilaged in special bags. The bags were filled with 10 kg chops each. The control bags were without preservative; the other bags had 3 biological and 3 chemical preservatives applied. The Thermochron sensor (Maxim Integrated, Dublin) for continuous measurement of temperature was inserted in each bag. The sensor was adjusted for hourly measurement, with an accuracy of  $0.065^{\circ}$ C. The bags were then put into the bunker silo, covered with chops that were tamped down and subsequently covered with two layers of film, weighted down with tyres all over the surface, to create an anaerobic environment.

After 4 months of fermentation, the bags were removed from the bunker silo and taken to the laboratory to be analysed with standard AOAC methods (1995). The anaerobic stability was determined according to Ranjit and Kung (2000). The statistical values were processed with the help of the ANOVA single-factor analysis of variance, Tukey's HSD test in the Statistica 10 program (StatSoft, Inc. 2011, Tulsa, OK, USA), at significance level  $\alpha = 0.05$ .

#### **Results and Discussion**

The maize was ensilaged with dry matter at a level of 46.7 %, neutrally detergent fibre (NDF) at a level of 57.5 % DM and a lower content of starch at a level of 29.5 % DM.

Index/Month	IV	V	VI	VII	VIII	IX
Precipitation at Uhřiněves [mm]	31	49	59	36	67	33
Long-term normal (1961-1990)	47	74	84	79	78	52
% of normal	66	66	70	46	86	63

Table 1Precipitation in year 2015

# Table 2 Nutritional values of maize silages

Table 2 Nutritiona		sinages				
Additives/Index	Dry matter	Protein	Sugar	ADF	NDF	Starch
	%	% DM	% DM	% DM	% DM	% DM
Control	43.5 <sup>ab</sup>	9.36 <sup>ab</sup>	$0.76^{ab}$	25.7	53.3	28.5 <sup>a</sup>
Biological:						
Lalsil Fresh	43.6 <sup>ab</sup>	9.30 <sup>ab</sup>	$0.77^{ab}$	25.6	50.8	30.6 <sup>ab</sup>
Ecosyl	46.4 <sup>b</sup>	9.24 <sup>ab</sup>	1.43 <sup>bc</sup>	26.6	48.7	32.4 <sup>ab</sup>
Formasil	42.1 <sup>a</sup>	9.21ª	0.43 <sup>a</sup>	27.7	55.8	27.5ª
Chemical:						
Albisil	45.3 <sup>b</sup>	9.24 <sup>ab</sup>	$0.76^{ab}$	28.0	55.1	27.7 <sup>a</sup>
Alicin	43.9 <sup>ab</sup>	9.22 <sup>a</sup>	0.69 <sup>ab</sup>	26.7	52.2	30.8 <sup>ab</sup>
Safesil	43.7 <sup>ab</sup>	9.31 <sup>ab</sup>	1.00 <sup>abc</sup>	25.4	52.6	28.1ª

DM = dry matter, ADF = acid detergent fibre, NDF = neutral detergent fibre, Different letter superscripts within a column indicate statistical differences,  $\alpha = 0.05$ 

The differences between the maximal and minimal temperatures between silages with different preservatives were not higher than 2°C all over the time of storage of the silage in the bunker silo. The application of the biological and chemical additives on the ensilaged maize chops did not have any effect on the content of dry matter or on the nutritional values in the dry matter, as compared to the control samples without preservatives.

Additives/Index	pH	AWE mg KOH/100 g	LA % FM	VFA % FM	LA/VFA	N-NH3 mg N/100 g
Control	3.89 <sup>c</sup>	1234 <sup>a</sup>	2.10	0.93 <sup>ab</sup>	2.25	23.8 <sup>ab</sup>
Biological:						
Lalsil Fresh	3.84 <sup>b</sup>	1331 <sup>ab</sup>	1.82	$0.95^{ab}$	1.93	25.2 <sup>ab</sup>
Ecosyl	3.84 <sup>b</sup>	1347 <sup>abc</sup>	2.09	0.91 <sup>ab</sup>	2.31	23.5 <sup>ab</sup>
Formasil Maize	3.82 <sup>ab</sup>	1459°	2.50	0.92 <sup>ab</sup>	2.76	23.8 <sup>ab</sup>
Chemical:						
Albisil	3.82 <sup>ab</sup>	1459°	2.40	1.04 <sup>b</sup>	2.30	23.3ª
Alicin	3.81 <sup>a</sup>	1392 <sup>bc</sup>	2.41	$0.88^{a}$	2.78	26.6 <sup>b</sup>
Safesil	3.82 <sup>ab</sup>	1391 <sup>bc</sup>	2.02	0.99 <sup>ab</sup>	2.03	24.8 <sup>ab</sup>

Table 3 Quality of	fermentation o	of maize silages
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AWE = acidity of water extract, LA = lactic acid, VFA = voluntary fatty acids, FA = fresh matter, Different letter superscripts within a column indicate statistical differences,  $\alpha = 0.05$ 

All ensilaging (both biological and chemical) additives contributed to reduce the pH, as compared to the control silage without preservatives. A significantly higher acidity of the water extract, as compared to the control silage, was measured in all silages preserved by chemical agents and by the agent containing *L. buchneri* and *P. pentosaceus*. While the aerobic stability of the silage without preservatives was 107 hours on average, the aerobic stability of the silage with the combined hetero and homo fermentative ensilaging agent was 142 hours, and that of the silage with the chemical agent containing natrium benzoate, potassium sorbate and sodium nitrite was more than 170 hours.

A similar experiment made on maize with 37.8 % dry matter but with lower NDF (45.1 % DM) was evaluated by Filya (2003) for control silage without preservatives, with *L. buchneri*, with *L. plantarum* and with their combinations. His results show that the agent with *L. buchneri*, as compared to the control silage, significantly reduced the content of lactic acid and increased the content of acetic acid, while the agent with *L. plantarum* had an opposite effect, significantly reducing the lactic acid and increasing the acetic acid.

#### Conclusion

The biological ensilaging agent containing *L. buchneri* and *P. pentosaceus*, or possibly the chemical agent containing natrium benzoate, potassium sorbate and sodium nitrite, can be recommended to ensilage the maize affected by dry weather during vegetation.

## **Dedication:** MZERO0716

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# White and Narrow-leafed Lupine as an Alternative Source of Quality Forage

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# Introduction

White lupine (*Lupinus albus*) and narrow-leafed or blue lupine (*Lupinus angustifolius*) are main species of lupines used in agriculture. The main focus of lupine growing is seeds production, which have high protein content (295-482 g/kg dry matter) and can successfully replace soybean meal in dairy cows feeding (Niwinska and Andrzejewski, 2011). Borreani et al. (2009) described lupins as annual crops that are well suited for short crop rotations when harvested as ensiled forage. Same authors concluded that lupin can be successfully ensiled with the application of a LAB inoculant and therewithal detected significantly higher crude protein content for lupin forage in comparison with field pea and faba bean. Fraser et al. (2005a,b) demonstrated that white lupine and narrow-leafed lupine could be successfully harvested as whole crop, but differences among species and cultivars were found. Qualitative parameters of lupine crops could be influenced not only by species and cultivars but mainly by habitat conditions. However, very little is known about yields, chemical composition and digestibility of these crops when grown under Czech Republic conditions.

The aim of the experiment was determination of yield, chemical composition and digestibility of white lupine and narrow-leafed lupine harvested as whole-crop.

#### **Materials and Methods**

White lupine (*Lupinus albus* cv. Amiga) and narrow-leafed lupine (*Lupinus angustifolius* cv. Primadona) were grown in 2015 on brown soil at Praha-Uhříněves, Czech Republic. Three samples from each lupine were harvested as whole crop during growing season. First cut was done when two-thirds of flowers were faded; pods and seeds were small and green. During the second cut, plants were completely faded; pods were light green and seeds full and green. Third harvest was realized when plants had fallen leaves; pods desiccated and seeds were in wax ripeness. Each harvest of both lupines was done in three replicates. Height of plants and yield were measured during harvests. All lupine crop samples were dried at 50°C for 48 hours, ground to pass through a 1-mm screen and analysed for dry matter (DM), crude protein (CP), ash, ether extract, neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL) and crude protein fractions (Fraction A – soluble part; B1 – fast degradable part; B2 medium degradable part; B3 – slowly degradable part; C – indigestible part of CP). Digestibility of organic matter was analysed using *in vitro* pepsin cellulase solubility method.

The data were analysed using GLM procedure of SAS (SAS Institute Inc., Cary, NC, USA, 2002), with crops, maturity and their interaction as fixed effects. The Scheffe test (P<0.05) was used to interpret any significant differences among mean values.

#### **Results and Discussion**

The effects of maturity and lupine species on plant height, yields, chemical composition and organic matter digestibility are presented in Table 1. White lupine had higher plants during whole harvest period. Generally, white lupine reached lupines (P<0.01). Fraser et al. (2005a) detected increasing of NDF content from 314 to 456 g/kg DM (P<0.01) for two varieties of white lupine.

Differences of crude protein fractions between lupine species were not detected except medium degradable part (B2) where higher B2 content was determined for narrow-leafed lupine (P<0.01) in comparison with white lupine (69 vs. 57 g/ higher yields of DM and CP (P<0.05) in comparison with narrow-leafed lupine (9.2 vs. 6.9 and 1.5 vs. 1.2 t/ha, respectively), but in first and third harvest differences were not significant. During maturing of both lupines yields grew and at the last harvest dropped again, which is in line with the results of Fraser et al. (2005a). Dry matter content grew during maturing similarly at both lupines (P<0.05), however higher dry matter content was detected for narrow-leafed lupine (P<0.05) in comparison with white lupine (184 vs. 166 g/kg, respectively). Content of CP was also higher for narrow-leafed lupine (183 vs. 170 g/kg DM) during all harvests and ranged from 226 to 143 g/kg DM. Crude protein contents of white lupine ranged from 215 to 136 g/kg DM. Similar CP contents for both lupine species were detected also by Fraser et al. (2005 a,b).

The NDF, ADF and ADL contents had higher (P<0.01) white lupine in comparison with narrow-leafed lupine (428 vs. 376, 361 vs. 300 and 42 vs. 31 g/kg DM, respectively). Fibre fractions contents were increased during the maturing of both kg DM, respectively).

Digestibility of organic matter detected in vitro pepsin cellulase solubility method was higher (P<0.01) for narrowleafed lupine (70% of OM) in comparison with white lupine (67% of OM). Statistically different was OM digestibility of forages from third harvest compared with first two harvests (P<0.01). This corresponds with results of DM digestibility white lupine silages made from different maturity forages presented by Fraser et al. (2005a).

Maturity		1	2	2		3			Р	
Crop	NL	WL	NL	WL	NL	WL	sem	С	Μ	C*M
Height (cm)	60 <sup>f</sup>	100 <sup>c</sup>	66 <sup>d</sup>	128 <sup>a</sup>	63 <sup>e</sup>	120 <sup>b</sup>	3.106	**	**	**
Yield (t/ha)										
Dry matter	3.80 <sup>d</sup>	4.54 <sup>d</sup>	7.55°	12.7 <sup>a</sup>	9.24 <sup>bc</sup>	10.3 <sup>ab</sup>	0.734	**	**	**
Crude protein	0.86 <sup>d</sup>	0.97 <sup>cd</sup>	1.34 <sup>b</sup>	2.00 <sup>a</sup>	1.31 <sup>bc</sup>	1.41 <sup>b</sup>	0.012	**	**	**
Chemical composition (g/l	kg of dry n	natter)								
Dry matter	99.2 <sup>d</sup>	97.3 <sup>d</sup>	147 <sup>cd</sup>	152 <sup>c</sup>	307 <sup>a</sup>	249 <sup>b</sup>	26.98	*	**	*
Ash	139 <sup>a</sup>	101 <sup>b</sup>	99.2 <sup>bc</sup>	59.0 <sup>d</sup>	85.0 <sup>c</sup>	60.9 <sup>d</sup>	2.27	**	**	*
Crude protein	226	215	179	158	143	136	9.31	*	**	Ν
Ether extract	13.0	12.5	8.7	7.0	18.1	21.1	0.48	Ν	**	Ν
Fibre fractions (g/kg of dr	y matter)									
NDF	320	382	361	395	446	507	26.46	**	**	Ν
ADF	284	342	279	336	338	405	14.89	**	**	Ν
ADL	31.2	37.8	25.0	38.1	37.8	49.5	1.20	**	**	Ν
Crude protein fractions (g/	kg of dry i	matter)								
А	84.4	70.2	86.5	84.8	39.3	48.7	5.40	Ν	**	Ν
B1	24.9	23.6	14.4	16.1	19.2	28.8	2.56	Ν	*	Ν
B2	69.6ª	78.6ª	63.3 <sup>ab</sup>	45.3 <sup>b</sup>	73.0 <sup>a</sup>	47.4 <sup>b</sup>	4.40	**	**	**
B3	22.7	22.9	8.3	3.3	2.9	3.1	1.31	Ν	**	Ν
С	24.7	19.7	6.0	8.4	8.1	8.4	1.08	Ν	**	Ν
Digestibility (%)										
Organic matter	71.8	69.3	71.1	67.7	67.8	65.2	0.622	**	**	Ν

 Table 1
 Yields, forage chemical composition and digestibility of white and narrow-leafed lupines harvested in three different maturities

\*\* P < 0.01; \* P < 0.05; N - P > 0.05; C - crop; M - maturity; NL - narrow-leafed lupine; WL - white lupine; Maturity 1 - two-thirds of flowers were faded; pods and seeds were small and green; 2 - plants were completely faded; pods were light green and seeds full and green; 3 - plants had fallen leaves; pods desiccated and seeds were in wax ripeness. Fraction A - soluble part; B1 - fast degradable part; B2 medium degradable part; B3 - slowly degradable part; C - indigestible part of CP.

#### Conclusion

Narrow-leafed lupine had higher crude protein content and organic matter digestibility. But white lupine had higher fibre contents and also higher yields of dry matter and crude protein. The data show that both lupines are suited for whole crop use but high forage quality could be obtained from younger growth of both tested lupine species.

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# Changes in Dry Matter Production of Abandoned Grassland Throughout its Revitalization

**BACK** 

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# Introduction

Colonization of grassland by the plant species which are typically present in an ongoing process of succession brings not only changes in above-ground phytomass and net primary production, but has effects on sunshine and the light reaching the surface, on microclimatic conditions and on availability of nutrients and water (DIERCHKE, 2006; VAN AUKEN, 2009). Revitalization of unused grassland leading to the recovery of species composition may increase positive and long-time effects of exploitation in relation to the production function (BULLOCK et al., 2007).

#### **Materials and Methods**

The research objective was to assess the production changes in abandoned grassland when revitalized by different management procedures. Over three years (2013-2015), a research trial was performed applying a range of management techniques to the partly revitalized abandoned grassland at *Liptovská Teplička* site (altitude 900 m). There were 8 trial treatments, namely: Treatment (T) 1- control (non-utilised grassland); T2 - one cut a year; T3 - two cuts a year (PK applied); T4 - two cuts a year; T5 - mulching once a year; T6 - two cuts a year (PK + N45 applied); T7/ two cuts a year (PK + N90 applied); T8/ three cuts a year (PK + N90 applied). In the spring, phosphorus (P) and potassium (K) fertilizers were applied at the rates of 30 kg P ha<sup>-1</sup> year<sup>-1</sup> and 60 kg K ha<sup>-1</sup> year<sup>-1</sup>. Nitrogen (N) fertilizer rate (90 kg) was split into two dressings of 45 kg ha<sup>-1</sup>, applied in the spring and after the 1<sup>st</sup> cut, respectively. This fertilizer application pattern was identical for the two-cut and three-cut treatments. At the one-cut treatment, cutting was made at the onset of earing and then at full earing of dominant grasses. At the two-cut treatments, the 2<sup>nd</sup> cut followed ca 60-65 days after the 1<sup>st</sup> cut. At the three-cut treatments, the cutting intervals for the 2<sup>nd</sup> and 3<sup>rd</sup> cuts were ca 40-45 days. The cutting dates were determined in compliance with the immediate condition of grassland, considering the phenological stage and regrowth of sward. At each of the cuts, dry matter (DM) of above-ground phytomass was determined as tha<sup>-1</sup>.

## **Results and Discussion**

In 2013, the highest total DM production of above-ground phytomass was recorded at the two-cut treatments with the fertilizer application (Table 1). The DM yields were: 6.43 t ha<sup>-1</sup> at T3 (PK); 6.12 t ha<sup>-1</sup> at T6 (N<sub>45</sub>) and 5.92 t ha<sup>-1</sup> at T7 (N<sub>90</sub>). The lowest DM production (4.24 t ha<sup>-1</sup>) was recorded at T7 (mulching). The assessment of the  $2^{nd}$  cuts showed increasing DM production with the level of intensification (fertilizer application). The yields of DM were: 2.23 t ha<sup>-1</sup> at the three-cut T8 (PK+N90); 1.39 t ha<sup>-1</sup> at two-cut T7 (PK+N90) and 1.28 ha<sup>-1</sup> at T6 ((PK+N45).

In 2014, the highest DM yields were recorded at the two-cut treatments T3 and T7, namely 6.28 ha<sup>-1</sup> at the treatment with PK applied (Table 2). The fertilizer effect was found also at the three-cut treatment (when compared to the year 2013), because the DM yield increased to 5.59 ha<sup>-1</sup>, what was the third highest yield recorded in 2014. The lowest DM yield was found at the control (3.88 t ha<sup>-1</sup>, which was just a small difference when compared to the 4.13 t ha<sup>-1</sup> DM at T2 (one cut a year). The year 2015 was a specific one, due to the weather conditions (Table 4). By comparison with the previous two years, the temperature increased, but rainfall decreased over the growing season (529.5 mm in 2013; 558.3 mm in 2014 and 301.9 mm in 2014). These factors influenced the decrease in DM production at all the treatments. Averaged over the years, the differences were significant (ANOVA  $F_{2.45}=13.38$ ; P<0.0001). The statistical differences between the years 2015 and 2013 as well as between 2015 and 2014 were very significant. In 2015, the highest annual DM yields were recorded at the treatments with N fertilizer application, as given in Table 3 (4.36 t ha<sup>-1</sup> at T8; 4.20 t ha<sup>-1</sup> at T7 and 3.82 t ha<sup>-1</sup> at T6). The multiple-cut treatments were most productive at the 1<sup>st</sup> cuts (2.85 t ha<sup>-1</sup> at T6 and 2.70 t ha<sup>-1</sup> at T7 (Table 3). However, the highest DM yields of the treatments with N fertilizer were found at the 2<sup>nd</sup> cuts, namely 1.50 t ha<sup>-1</sup> at T7 and 1.49 t ha<sup>-1</sup> at T8. Increasing the rates of mineral fertilizers, N especially, results in marked effects on productivity of grassland. The effects were reported by a range of authors (LICHNER et al., 1977; HOLÚBEK, 1987 and others). The factor of trial treatment (T1-T8) had also statistically significant effects on DM production (ANOVA F<sub>7.40</sub>=2.97; P=0.0132).

#### Conclusions

There were differences in DM production relating to the treatments applied during the revitalization process at abandoned grassland. Over the first two years, the highest DM production was recorded at the two-cut treatments with fertilizer application (T3 and T6 in 2013; T3 and T7 in 2014), the highest DM yields were found at the PK fertilizer application treatments. In the last year, the DM production was affected by the weather conditions, mainly by the rainfall deficiency over the growing season. In the mean of years, there were significant differences in DM yields, mainly between the years 2015 and 2013, and between 2015 and 2014. The highest DM production of this grassland type was recorded at the treatments with N fertilizer only in 2015 (the 3<sup>rd</sup> year of trial). This was the year when the positive effects of N fertilizer were shown not only in the increased production, but also in sustainable stability of yields throughout the dry years.

Table I Diy matter	yields in 2015 (t lid )			
Treatments	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cut	Σ
1	5.37	-	-	5.37
2	4.77	-	-	4.77
3	5.18	1.25	-	6.43
4	4.09	1.05	-	5.13
5	4.24	-	-	4.24
6	4.84	1.278	-	6.12
7	4.52	1.39	-	5.92
8	1.99	2.23	0.73	4.95

**Table 1**Dry matter yields in 2013 (t ha<sup>-1</sup>)

# **Table 2**Dry matter yields in 2014 (t ha<sup>-1</sup>)

Treatments	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cut	Σ
1	3.88	-	-	3.88
2	4.13	-	-	4.13
3	3.19	3.09	-	6.28
4	2.35	3.17	-	5.52
5	4.42	-	-	4.42
6	3.58	1.53	-	5.11
7	4.08	2.08	-	6.16
8	2.92	1.58	1.08	5.59

# **Table 3** Dry matter yields in 2015 (t $ha^{-1}$ )

Treatments	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cut	Σ
1	3.03	-	-	3.03
2	2.70	-	-	2.70
3	2.22	1.24	-	3.46
4	1.75	0.89	-	2.65
5	3.43	-	-	3.43
6	2.85	0.97	-	3.82
7	2.70	1.50	-	4.20
8	2.17	1.49	0.70	4.36

Table 4 Mean daily temperature (t<sub>d</sub> °C), rainfall (R mm) in total and over the growing season (VO) in 2013 – 2015

Year	t <sub>d</sub> /	Months									vo	Vaar			
	R	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sep.	Oct.	Nov.	Dec.	VU	Year
2013	t <sub>d</sub>	-5.0	-3.7	-2.4	7.5	11.9	15.4	18.0	17.8	9.5	8.3	2.3	-0.8	13.4	6.6
	R	96.0	106.4	89.4	36.0	143.0	154.7	51.2	58.1	86.5	52.6	120.7	36.3	529.5	1030.8
2014	t <sub>d</sub>	-1.6	0.5	4.11	7.72	10.74	14.9	16.3	14.2	12.4	7.6	3.7	-1.5	12.7	7.5
	R	52.6	51.2	61.1	67.8	164.1	80.8	125.5	74.1	46.0	19.0	10.1	10.5	558.3	762.8
2015	t <sub>d</sub>	-2.5	-2.93	1.72	6.1	11.2	15.1	18.2	19.3	18.2	5.8	3.4	0.5	13.7	7.4
	R	16.2	5.8	4.3	6.5	83.7	22.0	35.6	21.5	132.6	42.5	57.2	8.8	301.9	436.7

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# Changes in Energy Value of Permanent Grassland in Area of Nitra City

**BACK** 

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## Introduction

The basic function of grass ecosystem is the production of fytomass, which provide for human and animal nutrition, energy renewal, landscape creation and conservation of biodiversity on the earth (Novák, 2008; Hrúzová et al., 2015). Grasslands we use in two different ways, first is grazing and second is mowing (Jančovič et al., 2015). Grazing is natural and the cheapest form of animal nutrition. Grazing has advantages, bud has as well disadvantages (Čunderlíková et al., 2002, Rajčáková et al., 2002). Nutritional value of grassland is affected by the proportion of nutrients and by vegetative stage. Some nutritional value have as well the weed, however the presence of weed in grassland is evaluated as a negative (Gálik et al., 2011). One from the factors determining the quality of grassland is energy (Bíro et al., 2014). The knowledge about energy concentration of feed is important factor for objectification of calculation of feed ration (Juráček et al., 2013). The aim of this study was to analyze the energy concentration of permanent grassland in four different areas around Nitra city.

### **Materials and Methods**

Samples for determination of dry matter and energy concentration of permanent grassland were collected around Nitra city. Four areas were selected (east, west, north and south area of Nitra city). Permanent grasslands in our study were not utilized, it means they were without grazing or mowing. Average sample for each area and each month consist from 10 to 15 subsamples (subsamples were mixed together). Samples were taken during first week of every month from April to October. Concentration of dry matter and energy was analysed at Department of Animal Nutrition (SUA in Nitra) in Laboratory of Quality and Nutritive Value of Feeds. Dry matter (DM) content of the samples was determined by oven drying at 103°C to the constant weight. Ash content was obtained in a muffle oven at 550°C. Organic matter (OM) was calculated, OM=dry matter - ash. Coefficients of digestibility of organic matter (DOM) was calculated according formula: DOM= (OM \* coefficients of digestibility of organic matter (DOM) was calculated according formula: DOM= (OM \* coefficients of digestibility of organic matter (DOM) was calculated according formula: ME= 0.01517 \* DOM (Bíro et al., 2012). Net energy lactation (NEL) was calculated according formula: NEL= ME \* [0.463 + (0.24 \* q]; where "q" was calculated according formula: q= ME/GE (Bíro et al., 2012). The calculations and graphs were processed in program Excel (Microsoft Office 2013).

#### **Results and Discussion**

Skládanka et al. (2014) published that, the sum of meteoric water is for permanent grassland very important and during vegetation period should by 400 to 500 mm. Air temperature affect the photosynthesis and together with meteoric water is the main factor affecting production of fytomass. Climatic conditions affects species structure and production of permanent grassland (Skládanka et al., 2014). The climatic conditions during whole sampling period are presented in Table 1.

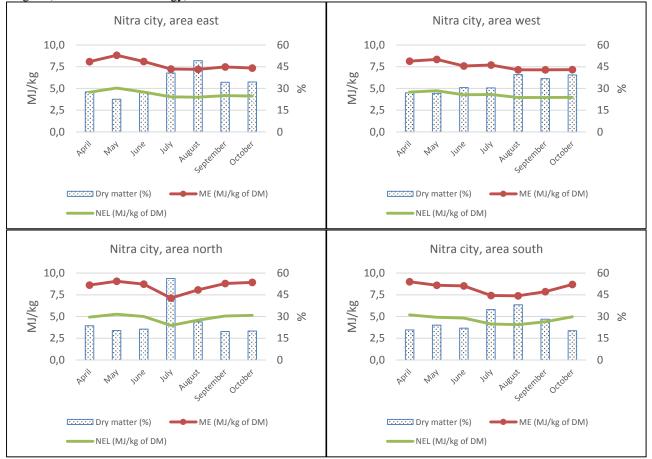
	April	May	June	July	August	September	October
Meteoric water /mm/	18	73	42	1	62	67	28
Temperature /°C/	11.9	15.9	20.4	22.9	22.6	14.6	11.7

 Table 1
 Climatic conditions in Nitra city during sampling months (www.shmu.sk)

The effect of aging of grasses is decrease in concentration of energy (Skládanka et al., 2014), which corresponded with our results (Figure 1). The highest concentration of energy was in months April (ME from 8.1 to 9.0 MJ/kg DM; NEL form 4.6 to 5.2 MJ/kg DM) and May (ME form 8.3 to 9.0 MJ/kg DM; NEL form 4.7 to 5.3 MJ/kg DM). This correlated with the lowest concentration of dry matter in the permanent grassland in months April (from 21 to 28% of DM) and May (from 20 to 27% of DM). The lowest concentration of energy was determined in months July (ME from 7.1 to 7.7 MJ/kg DM; NEL from 4.0 to 4.3 MJ/kg DM) and August (ME from 7.1 to 8.1 MJ/kg DM; and NEL from 4.0 to 4.6 MJ/kg DM). This correlated with the highest concentration of dry matter in the permanent grassland in months July (from 30 to 56%) and August (from 26 to 49%) (Figure 1). Skládanka et al. (2014) published concentration of ME of grass for mid phase of growth on 10.61 MJ/kg DM and for old grass stand on 8.36 MJ/kg DM. Concentration of NEL in all our samples is lower in comparison to concentration of NEL for meadows (5.5 to 5.6 MJ/kg DM) published by Gálik et al. (2011). Skládanka et al. (2014) published NEL concentration for permanent grassland from 6.38 at the beginning of growth and then with decrease to 4.73 MJ/kg DM connected with the old grass stand.

## Conclusions

The energy concentration of permanent grasslands around Nitra city was in general lower than showed the information in literature. The energy concentration is depended on the concentration of dry matter, organic matter and its digestibility. By the increase of dry matter come to decrease in energy content and vice versa. The highest concentration of energy in permanent grassland was in months April and May. July was the month with lowest amount of meteoric water, which negative affects the concentration of energy in permanent grassland in months July and August (lowest content of energy).



**Figure 1** Development of dry matter and energy concentration of permanent grassland in areas of Nitra city during grazing and mowing season. DM – dry matter; ME – metabolizable energy; NEL – net energy lactation

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# Productive Ability and Nutritive Value of Alfalfa (*Medicago Sativa* L.) and its Simple Mixtures When Grown in a Mountainous Region

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BACK

### Introduction

Sown grassland provides high yields of quality forage. Many authors (Frame, 2005, Hopkins and Wilkins, 2006) think that red clover (*Trifolium pratense* L.) is crucial for a short-term sown sward, albeit alfalfa (*Medicago sativa* L.) is considered the leading species of *Fabaceae*. Production of dry matter (DM) and nutrients outperforms other perennial forage crops, especially in warm and dry areas. Hrabě et al. (2004) reported high content of calcium (Ca), phosphorus (P), potassium (K) and other macro-elements and vitamins, especially  $\beta$ -carotene. In mixtures, grasses support wilting, accelerate preservation at ensiling, increase DM content and contain more water-soluble carbohydrates (WSC) than clovers (Pozdíšek and Kohoutek, 2003). The intergeneric hybrids – x *Festulolium* rank among important components of grass/clover mixtures. The hybrids are easy to produce and they combine the nutritive value of *Lolium perenne* with the persistence and drought resistance of *Festuca arrundinacea* Schreb. and with the frost resistance of *Festuca pratensis* Huds. (Kopecký et al., 2006, Kościelniak et al., 2006).

#### **Materials and Methods**

A research into efficiency in sward, DM yields and nutritive value comprised some cultivars of *Festulolium*, *Medicago sativa* L. and their simple mixtures when grown in a mountain region of Slovakia. The research site characteristics: longitude 20° 06'; latitude 48° 55'; altitude 960 m; mean annual rainfall 950 mm; mean rainfall over the growing season 525 mm; mean daily temperature  $3.5^{\circ}$ C; mean daily temperature over the growing season  $9.5^{\circ}$ C; geological substratum: carbonates; soil type: rendzina; soil texture: loamy. The research treatments (T) were: T1: x *Festulolium* cv. Hostyn; T2: *Festulolium* cv. GR14; T3: *M. sativa* cv. Tereza; T4: *M. sativa* / grass mixture (cv. Tereza +cv. Hostyn); T5: *M. sativa* / grass mixture (cv. Tereza +cv. GR14). The swards were utilised by 3 cuts a year.

#### **Results and Discussion**

The yield formation of sown grassland relates to the site characteristics, management techniques and the status of sward. The sward condition and weather were reflected in the amount of DM production (Ilavská and Jančová, 2016). In the 1<sup>st</sup> harvest year, the DM yield was high, ranging between 9.4 and 10.5 t ha<sup>-1</sup> (Table 1) but the differences between the treatments were not significant. The highest proportion of total yield was found at the 1<sup>st</sup> cut and it was decreasing towards the 2<sup>nd</sup> and 3<sup>rd</sup> cuts over the growing season (except for T3 treatment). In the 2<sup>nd</sup> harvest year, as a result of notable rainfall deficiency, the yield rapidly decreased by 42, 39, 37, 28 and 36 % at the treatments T1-T5, respectively. The significant difference was recorded only between the treatments T2 and T4. Also in this year, the yields were decreasing after the 1st cut towards the 2nd and 3rd cuts, except for T1 treatment, but the yields were balanced. The ensiling capacity of herbage is given by water-soluble carbohydrates to crude protein ratio (WSC: CP). Table 2 shows the ensiling coefficients; the higher the coefficient, the better suitability of crop to be ensiled. The highest coefficients were recorded at the treatments with intergeneric grass hybrids (IGH). The lowest coefficients were found at M. sativa in both research years. By comparison between the cuts, the lowest coefficients were recorded at the 3<sup>rd</sup> cuts, but the highest ones were found at herbage from the 1<sup>st</sup> cuts. The nutritive value of silage made from IGH corresponded with the nutrient content and with the quality of preserved herbage. The content of silage parameters was higher at the grass treatments in the  $2^{nd}$  harvest year (Table 3). The significantly higher (P<0.05) values of PDIN (protein digested in the small intestine when nitrogen is limiting), PDIE (protein digested in the small intestine when energy is limiting) and PMP<sub>PDI</sub> (productive milk potential) were found at IGH cv. Hostyn. The PDIN was significantly higher in silages from the 3<sup>rd</sup> cuts, while NEL (net energy for lactation), NEV (net energy for fattening) and ME (metabolizable energy) were higher in the silages from the  $1^{st}$  cuts. This trend was identical in the *M. sativa* silages – higher PDIN and PDI in those from the 3<sup>rd</sup> cuts, while NEL, NEV and ME were higher in the silages from the 1<sup>st</sup> cuts. The PDIN, PDIE and PMP<sub>PDI</sub> in the mixture silage were significantly higher at the 3<sup>rd</sup> cut. The parameters of nutritive value and potential production efficiency of silage were higher in the 2<sup>nd</sup> harvest year. The degradable CP parameters (PDIN, PDIE) were significantly higher at *M. sativa* silage than in the other treatments in both trial years. The potential production efficiency ( $PMP_{NEL}$ ) was significantly higher at the IGH treatments. At all the cuts, PMP<sub>PDI</sub> values were higher at the IGH treatments than those at the grass monoculture treatments.

#### Conclusions

Growing the intergeneric grass hybrids (x *Festulolium*), *M. sativa* and their simple mixtures showed a suitability to be grown within a system of perennial forage crops at mountain regions. The mixtures of *M. sativa* were even more efficient and produced more compact swards than its monocultures. In the  $2^{nd}$  harvest year, the mixtures were also better than the IGH swards supported by nitrogen fertilizer application.

r	- T	natter (DN	1) producti							
Treatm				014					015	
		DM prop	ortion at the	e cuts (%)		DM	DM prop	ortion at the	cuts (%)	DM
					P	production				production
		1 et .	and	Ord		(t ha <sup>-1</sup> )	1 et .	and	ard	$(t ha^{-1})$
		1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cu	t	10 7 17	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cut	
T 1		44	38	18		10.565	44	45	11	6.124
T 2		47	39	14		9.425	49	43	8	5.744
Т 3		32	35	33		9.457	46	29	25	5.975
T 4		43	29	28		9.627	42	38	20	6.947
Т 5		41	31	28		10.376	42	34	24	6.610
LSD <sub>0.</sub>						2.043				1.161
		ficients of	ensiling				1			
Treat	ment			2014					15	
		1 <sup>st</sup> cu		2 <sup>nd</sup> cut		3 <sup>rd</sup> cut	1 <sup>st</sup> cut		cut	3 <sup>rd</sup> cut
Т		1.00		0.82		0.66	1.11		79	0.68
Т		0.63		0.30		0.49	0.68		58	0.55
Т		0.39		0.21		0.31	0.55	0.		0.21
T		0.48		0.57		0.43	0.89	0.	64	0.50
Т	5	0.77		0.44		0.51	1.04	0	42	0.21
Table 3	3 Nutri	tive value	of silage				•	•	I	
Years	Cuts	Treatme		N P	DIE	NEL	NEV	ME	PMP <sub>NEI</sub>	PMP <sub>PDI</sub>
			ş	g kg <sup>-</sup> 1 DN	1		MJ kg <sup>-1</sup> DM	[		g FCM
	1 <sup>st</sup>		68.5		5.24	5.64	5.50	9.54	1.80	1.37
	$2^{nd}$	T 1	61.5		5.23	5.56	5.42	9.42	1.78	1.23
	3 <sup>rd</sup>		75.2		7.36	5.43	5.30	9.20	1.74	1.50
	1 <sup>st</sup>		66.9	9 6	5.43	5.57	5.44	9.42	1.78	1.34
	$2^{nd}$	Т2	64.7	4 6	5.91	5.58	5.44	9.44	1.78	1.29
	3 <sup>rd</sup>		64.9	04 6	5.50	5.41	5.29	9.15	1.73	1.30
	1 <sup>st</sup>		100.	14 6	7.60	4.81	4.46	8.73	1.65	1.71
2014	$2^{nd}$	Т3	97.7	4 6	7.63	4.75	4.40	8.28	1.52	1.95
	3 <sup>rd</sup>		101.	97 6	7.54	4.55	4.23	7.93	1.45	2.04
	1 <sup>st</sup>		68.1	3 6	3.03	5.16	4.90	8.86	1.65	1.36
	$2^{nd}$	T 4	73.1	6 6	3.41	4.97	4.71	8.56	1.59	1.46
	3 <sup>rd</sup>		96.1	6 6	9.20	4.94	4.68	8.52	1.58	1.92
	$1^{st}$		71.9		1.69	5.09	4.83	8.76	1.63	1.44
	$2^{nd}$	Т 5	75.9		4.38	5.01	4.74	8.62	1.60	1.52
	3 <sup>rd</sup>		104.		1.24	4.95	4.67	8.55	1.58	2.09
	1 <sup>st</sup>		69.9		7.73	5.63	5.49	9.53	1.80	1.40
	$2^{nd}$	T 1	71.7		5.42	5.62	5.47	9.53	1.80	1.44
	3 <sup>rd</sup>		75.7		7.93	5.65	5.50	9.58	1.81	1.51
	1 <sup>st</sup>		74.4		7.02	5.60	5.45	9.50	1.79	1.49
	$2^{nd}$	T 2	67.4		5.27	5.62	5.47	9.53	1.80	1.35
	3 <sup>rd</sup>		68.2		4.35	5.60	5.45	9.49	1.79	1.36
	1 <sup>st</sup>		82.5		4.40	4.89	4.55	8.50	1.56	1.65
2015	$2^{nd}$	Т 3	96.3		7.22	4.86	4.51	8.48	1.55	1.93
	3 <sup>rd</sup>		112.		1.16	4.85	4.99	8.45	1.55	2.25
	1 <sup>st</sup>		82.4		5.39	5.13	4.85	8.85	1.64	1.65
	$2^{nd}$	T 4	95.1		0.34	5.12	4.84	8.84	1.64	1.90
	3 <sup>rd</sup>		92.9		9.33	5.10	4.83	8.79	1.63	1.86
	1 <sup>st</sup>		83.7		5.11	5.09	4.81	8.78	1.63	1.68
	$2^{nd}$	Т 5	86.9		7.56	5.13	4.85	8.83	1.64	1.74
	3 <sup>rd</sup>		92.9	03 6	9.79	5.17	4.89	8.90	1.65	1.86

# Table 1 Dry matter (DM) production (t ha<sup>-1</sup>)

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The list of references is available at the authors and/or at the conference secretary.

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## Effect of Silage Hybrid on Maize Forage Quality

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## Introduction

Maize is among easy ensiled roughage and high-quality maize silage is a good source of structural but also nonstructural carbohydrates (Vršková and Bencová, 2011). The selection of appropriate hybrids is critical for the production of quality maize silages (Polák and Jančová, 2006; Bíro et al., 2014; Plachý et al., 2014). Silage hybrid affects maturity at harvest, chemical composition, digestibility of nutrients, nutritive value, composition of epiphytic microflora, dry matter yield, energy and nutrients yield, production efficiency per 1 kg of silage and the overall efficiency of livestock production (Látal et al., 2010; Doležal et al., 2012; Skládanka et al., 2014). Intensive cultivation of maize hybrids broadens the spectrum of grown maize by new hybrids that are of higher yielding capacity, better state of health and bring nutritionally more valuable feeds than the older ones (Rajčáková et al., 2013).

The aim of this study was to analyze the parameters of nutritive value of medium-late hybrids of maize with the same FAO number and type of grain harvested in the same maturity stage.

#### **Materials and Methods**

Monitored hybrids were grown in the same agro-ecological conditions of the University farm Ltd. Kolíňany on a farm in Oponice and harvested in the same growth phase of *milky-wax ripening*. Harvested crop corn hybrids were mechanically adjusted to the 10 mm length of cut. Maize hybrid FAO 440a is a medium late hybrid with versatile use for grain or silage, including biogas in corn and beet production area. The hybrid is characterized by good stay green effect. Grain type is a dent. Maize hybrid FAO 440b is a medium late hybrid with universal application (silage/grain) in corn and beet production area. Grain type is a dent. Hybrids were sown on 11th April 2014. The hybrid FAO 440a was sown on 23 hectare areas and hybrid FAO 440b on 30 hectare area. The maize hybrids (FAO 440 a, b) are based on the final density - 75,000 plants per hectare, and grown in 180 mm row spacing. Before and during the growing period, nitrogen fertilizers, urea at a rate of 130 kg/ha and ammonium nitrate at a rate of 220 kg/ha were applied to both hybrids . Corn hybrids were harvested and processed by mobile Harvester Claas Jaguar (with technology - corn cracker) with theoretical length of cut: 10 mm. We have taken samples for laboratory analysis to determine the parameters of the nutrient content (n = 3) after adjusting of matter. Content of nutrients was analysed at Department of Animal Nutrition (SUA in Nitra) in Laboratory of Quality and Nutritive Value of Feeds. Standard analytical methods (Regulation no. 2136/2004-100) were used for analyzing organic (crude protein, crude fiber, fat, total sugars) and inorganic nutrients (ash). Dry matter (DM) was determined by drying at 103°C. Content of nitrogen fee extract (NFE) was calculated: NFE = dry matter – (crude protein + crude fiber + fat + ash). Starch was analyzed by polarimetric method. Organic matter (OM) was calculated, OM = dry matter – ash. Acid detergent fiber (ADF), acid detergent lignin (ADL) and neutral detergent fiber (NDF) were determined according to methods of Van Soest et al. (1991). Results were statistically analysed in statistic program SAS Enterprise Guide 5.1. (SAS Institute, Inc).

#### **Results and Discussion**

Statistically significant (P<0.05) higher dry matter content was in hybrid FAO 440a, despite the fact that tested hybrids had the same FAO number, type of grain, were sown at the same time, grown under the same agro-ecological conditions and harvested in the same growth phase milky-wax grain maturity. Maize is typically deficient in the crude protein content (Gálik et al., 2011). Crude protein content and their fractions depends mainly on the maturity of the crop at harvest, climatic conditions and fertilizer application (Jendrišáková, 2010). Statistically significant higher crude protein content (P<0.05) was in hybrid FAO 440a compared to hybrid FAO 440b. According to Juráček et al. (2012), with the increasing number of FAO, the crude protein content increases. The content of crude protein in maize decreased with advancing maturity (Mahanna et al. (2014). Statistically significant (P<0.05) lower content of ash contained hybrid FAO 440b, compared to hybrid FAO 440a (43.6 vs. 46.4 g/kg of dry matter). Similar content of ash was found by Juráček et al. (2012), who determined ash content 44.0 g/kg of dry matter in hybrid FAO 450. The content of starch, which is an important source of energy (Gálik et al., 2011), was 372.2 g/kg of DM (FAO 440a) and 408.1 g/kg of DM (440b). Differences in starch content were statistically significant (P<0.05). Ryšavá et al. (2001) reported lower value of starch in the hybrid FAO 420 (328.0 g/kg of DM) when the dry matter content of fresh matter was 33 %. Total sugar content had values 57.4 g and 71.3 g/kg of DM (FAO 440a and 440b), the differences were statistically significant (P<0.05). Mlyneková and Čerešňáková (2005) reported in their work that there was less total sugar content in matter of hybrid FAO 440 (52.0 g/kg of DM). The content of acid detergent (ADF) and the neutral detergent (NDF) fraction of fiber was statistically significant (P <0.05) lower in the matter of the hybrid FAO 440b. Mlyneková and Čerešňáková (2005) found higher content of ADF and NDF (287.2 and 559.0 g/kg of DM) in maize hybrid FAO 440 (dry matter 37.5 %) as we determined in the tested hybrids.

Iusie	1 1100	intre vare	e of mail	e nom an	fierent ny	onas						
n=3		DM	СР	Fat	ADF	NDF	ADL	Ash	NFE	Starch	TS	OM
FAO	$\overline{\mathbf{X}}$	380.3 <sup>a</sup>	77.2 <sup>a</sup>	32.9	170.4 <sup>a</sup>	335.3 <sup>a</sup>	18.8	46.4 <sup>a</sup>	696.8	372.2 <sup>a</sup>	57.4 <sup>a</sup>	953.6ª
440a	S.D.	0.05	0.50	0.45	0.70	1.85	1.20	0.15	2.80	0.75	1.10	0.15
FAO	$\overline{\mathbf{X}}$	335.8 <sup>a</sup>	71.6 <sup>a</sup>	33.4	167.0 <sup>a</sup>	328.5 <sup>a</sup>	16.7	43.6 <sup>a</sup>	701.2	408.1 <sup>a</sup>	71.3 <sup>a</sup>	956.4 <sup>a</sup>
440b	S.D.	0.20	1.50	0.35	0.05	2.30	1.00	0.65	0.40	0.55	1.20	0.65
		~ 1										

 Table 1
 Nutritive value of maize from different hybrids

DM: dry matter, CP: crude protein, ADF: acid detergent fiber, NDF: neutral detergent fiber, ADL: acid detergent lignin, NFE: nitrogen free extract, TS: total sugars, OM: organic matter, <sup>a</sup> the values with identical superscripts in a column are significantly different at P < 0.05

#### Conclusions

Our results confirm differences in the nutritional value of maize hybrids, despite the fact that tested hybrids had the same FAO number, type of grain, were sown at the same time, grown under the same agro-ecological conditions and harvested in the same growth phase milky-wax grain maturity. In analyzed medium-late hybrids, we found statistically significant (P <0.05) higher content of dry matter in hybrid FAO 440a (380.3 g/kg) and lower dry matter content in hybrid FAO 440b (335.8 g/kg). In matter of hybrid FAO 440b, we determined statistically significant (P <0.05) higher levels of organic matter, total sugars, and starch, as compared to the hybrid FAO 440a. Statistically significant (P <0.05) lower content of ADF and NDF was found in the hybrid FAO 440b, as compared to the hybrid FAO 440a.

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# Chemical Quality Changes During Ripening of Silage Maize Hybrids

**BACK** 

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## Abstract

The chemical composition of silage maize hybrids was tested by NIR spectroscopy at different stages of maturity in three consecutive years (2013-2015). Dry matter content increased faster in the ears than in the stalks. Protein content reached its maximum on the 30th day after flowering. Starch content was consistent in the stalks while intensively increased in the ears. Lignin content of the stalks increased but of the ears decreased during ripening. Digestible organic matter content was the highest between the 30th and 40th days after flowering, followed by a slow decrease. The effect of the year was significant for chemical quality traits.

## Introduction

The extremities in weather conditions have unfavourable effects on the yield of forage crops such as silage maize, and also on the metabolism of the plants, the accumulation of nutrients into the kernels and the process of maturity. This can be traceable by detection of the changes in the chemical composition of the forages during ripening. If the high temperature is accompanied by the lack of precipitation after flowering - in the early stage of maturity-, plants dry down faster, kernels are forced to mature earlier, the grain filling process is blocked, which results in yield loss. In case of lower temperature and more rainfall the maize plant stays green longer, and reaches the ideal dry matter content for harvest later. Sometimes the influence of the genotype on the yield is greater than the effect of year, due to the maturity type or the good adaptation to environmental stresses, but the effect of the changes in climatic conditions cannot be neglected.

#### **Materials and Methods**

The changes of the chemical composition of the plant parts of silage maize hybrids during the ripening process were studied in our experiments. Four commercial silage maize hybrids – widely grown in Hungary – were tested in three consecutive years (2013-2014-2015) in Martonvásár, under irrigated conditions. Samples were taken at female flowering time and every 10 days after on, five times until harvest. Plants were chopped and divided into two fractions: ears with husk and cob, and stalk with leaves. The chemical composition of each fraction was analyzed by NIR spectroscopy and evaluated by INGOT software. At harvest, the green and dry matter yields of the plants were also measured. The digestible dry matter yield (DDMY) was calculated based on the yield and digestibility data.

#### **Results and Discussion**

The climate of the three examined years was very different. In 2013 there was a severe drought during July and August with 41 hot days (daily average temperature above 25C, maximums above 35C). The next year was cooler with twice of the average annual rainfall. Summer of 2015 was warm with moderate drought. It was expected that this kind of variation of the years would take effect on the chemical composition and the yield.

Based on the analysis of the results of the three years it has been found that the dry matter content of the ear increased more intensively than that of the stalk, and it did not depend on the year. The dry matter content of the whole plant at harvest was lower in 2014 - when the summer was cool and wet – than in the other two years. The protein content changed similarly in the ear and the stalk and reached its maximum on the  $30^{th}$  day from flowering. The starch content of the stalk was relatively stable, while there was an intensive growth observed in the ear, due to the accumulation of more and more nutrients into the kernels. The starch content of the whole plant at harvest was lowest in 2015. Lignin content, which has the greatest – and negative – effect on the digestibility, was constantly growing in the stalk, and decreasing in the ear during ripening every year. The lignin content of the stalk at harvest was greater in 2015 than in the previous years. The digestible organic matter content (IVDOM) of the ear was higher than that of the stalk, and this difference was expressed more and more with maturity. IVDOM reached its maximum between the  $30^{th}$  and  $40^{th}$  days from flowering, followed by a slight decrease. The IVDOM content of the ear was the greatest in 2013, while that of the stalk was greatest in 2014. This may have been caused by the fact that stalks remained green during August in 2014, while dried down early in the other two years. The digestible dry matter yield of the hybrids was the lowest in 2015. In the last two years, between the  $10^{th}$  and  $30^{th}$  days from flowering the DDMY doubled, but there was only a 40% increase of it in 2015.

#### Conclusion

The effect of the year was significant for chemical quality traits and maturity. In hot and dry years plants dried down and matured earlier. They produced less dry matter yield but with better quality than in the wet year. In drought years, it worth to harvest earlier in order to reach optimal digestible dry matter yield.

## Effect of Maize Cutting Height on its Nutritive Value

**BACK** 

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## Introduction

Increasing maize cutting height permits to harvest forage with a higher dry matter (DM) digestibility (Wu and Roth, 2003). Thus, the harvested forage could be used to feed high yielding cattle with high nutrient requirements. Moreover, introducing maize silage with a high digestibility could allow improving farms feed autonomy by increasing grass forage proportion in the diets. Our study aimed to measure yield, chemical composition and nutritive value of different maize cutting heights to provide technical recommendations for breeders.

## **Material and Methods**

Five maize hybrids were grown in two locations in France (La Chapelle Saint Sauveur – 44370 and Saint Hilaire en Woëvre – 55160). Samples were harvested at 360 g DM/kg (classical cutting height) from two experimental plots per hybrid. Maize plants were cut down at ground level (=0cm) and sliced into 5 stover sections: 0-15 cm; 15-35 cm; 35-55 cm; 55-75 cm and above 75 cm (section containing the cob). Mean plant height was 224 cm +/-20 cm and cob insertion height was 99 +/-9 cm. Nutritive values of the 20 sections (up to 15 cm) were obtained by chemical composition analysis and rumen degradability trial lead at La Jaillière experimental farm (FR-44). DM degradability was measured *in sacco* for 6 incubation times with 6 replicates. Effective degradability (ED) of DM was calculated with the step by step method assuming a rumen outflow of 6%.h<sup>-1</sup>. OMD was estimated from OMD *in sacco* (Férard *et al.*, 2014). The characteristics of forages harvested at 15 cm or 55 cm cutting height were calculated from data of each fraction. Nutritive values were calculated using Systali equations included in Systool Web 1.1 (Chapoutot *et al.*, 2015) with *in sacco* parameters. Consequences of raising maize cutting height on ration formulation (INRATION 3.22 software) and farm main surface area have been observed by replacing the classical '15 cm maize silage' by a '55 cm maize silage' in a dairy cow diet (fat corrected milk yield: 35 kg/day) base on maize silage, grass silage, wheat grain and soya meal.

#### **Results and Discussion**

#### **Chemical composition**

Between 0 and 75 cm above ground level, DM yield was very stable with 36+/- 6 kg DM/cm/ha. This result is a bit lower than the international reference set at 45 kg/cm/ha (Wu and Roth, 2003). NDF contents of sections under 75 cm are two times higher than sections 'above 75 cm'. Regarding CP content, sections under 75 cm are two times poorer than the rest of the plant above 75 cm (table 1). For a given harvest date, raising cutting height from 15 cm to 55 cm increases starch content by 100 g/kg and decreases NDF content by 70 g/kg. Raising cutting height from 15 cm to 55 cm concentrates the harvested forage in ears which permits to increase DM content by 20 or 30 g/kg to reach more quickly the optimal stage. This method could also be a mean to obtain a better standardization of the harvested forage composition over the different fields or over years of harvest.

		Plant s	-	ant cutting		
	15-35 cm	35-55 cm	55-75 cm	+75 cm	15 cm	55 cm
DM content (g/kg)	193	201	212	400	354	384
DM yield (tDM/ha)	0.7	0.7	0.6	13.6	15.6	14.2
Starch content (g/kg)	-	-	-	367	320	351
NDF content (g/kg)	674	672	647	365	404	377
ED DM	0.391	0.400	0.429	0.575	0.554	0.569
OMD	0.462	0.497	0.533	0.752	0.720	0.742
PDIN Systali (g/kgDM)	19	20	26	44	41	44
PDIE Systali (g/kgDM)	48	50	53	54	53	54
UFL Systali (/kgDM)	0.56	0.61	0.68	1.06	0.96	1.00

Table 1	Chemical composition,	digestibilities and	I nutritive values	of the different	plant sections

## Degradability and nutritive value

DM effective degradabilities were very stable for sections under 75 cm and much lower than the section above 75 cm containing the cob. The predicted organic matter digestibility increased from an average of 0.497 for stover sections 15-75 cm to 0.752 for sections above 75 cm. The increase in protein truly digestible in the small intestine (PDIN and PDIE) for the 5 hybrids were estimated on average at 1 and 3 g/kgDM higher for maize cut at 55 cm than for '15 cm maize'. At the same time, the OMD increased by 2.2 which is very close to what Wu and Roth (2003) found (+ 2.0). This better digestibility improves digestible energy (UFL) by 0.045 UFL/kgDM and increases forage ingestibility.

#### Diets and main forage area

The possibility to pull up the cutting height from 15 to 55 cm allows an increase of a few points of the forage:concentrate ratio without affecting NDF diet level nor milk quality (Dominguez and Satter, 2003). Based on a frequent dairy cow diet, replacing 15 cm maize silage by 55 cm maize silage lead to increase the forage:concentrate ratio from 69:31 to 74:26 and 76:24 respectively for diets aiming to maximize grass and maize intake (table 2). Due to a lower crop yield with a high cut, maize forage area needs to be multiplied by more than 1.1. The main forage area (MFA) tends to be higher when a '55 cm maize silage' is used in the ration formulation. To maintain the milk production per hectare of MFA, the DM yield ratio wheat:maize silage should not exceed 0.45 and 0.31 respectively for the diets formulation maximizing maize or grass intake.

	D	iets >>>	Control	maximize maize intake	maximize grass intake
	Ray grass silage (Lolium multiflorum)		4	4	4.8
	15 cm maize silage		11.5		
Diet	55 cm maize silage			12.9	11.5
formulation	Wheat grain		2	0.8	1.3
(kgDM/d/cow)	Soya meal		4.8	4.5	4.45
	Mineral		0.1	0.1	0.1
	Total DMI		22.4	22.3	22.2
	Diet starch content (g/kgDM)		25.6	25.9	25.1
Diet	Total diet NDF content (g/kgDM)		31.5	31.1	31.4
characteristics	Forage NDF content (g/kgDM)		27.2	27.8	27.8
	Diet forage:concentrate ratio		69%	76%	74%
	Ray grass - 11 tDM/ha (ha for 1000 ration	s)	0.36	0.36	0.44
	15 cm Maize - 15.6 tDM/ha (ha for 1000 th	rations)	0.74		
Farm land use	55 cm Maize - 14.2 tDM/ha (ha for 1000 th	rations)		0.91	0.81
	Wheat – 7.7 tDM/ha (ha for 1000 rations)		0.26	0.10	0.17
	Main forage area (ha for 1000 rations)		1.36	1.38	1.42
	Surface ratio wheat:maize forage		0.35	0.12	0.21

Table 2 Ration formulations introducing '55 cm maize silage' and their farmland use consequences

## Conclusions

Raising maize whole plant cutting height allows getting a better nutritive value for the harvested forage but penalize a bit the DM yield per hectare. The adjustment of the height cut could be an effective mean to modify the forage chemical composition to reach more easily the quality required depending on the cattle needs. The use of high cut maize silage is efficient to level up the proportion of forage in the ration formulation but need to extend the farm main forage area.

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# The Effect of Harvest Date and Fermentation Process on the Levels of Carotenoids and Tocopherols in Virginia Fanpetals (*Sida hermaphrodita*) Herbage and Silage

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## Abstract

The aim of this study was to determine the concentrations of  $\beta$ -carotene and tocopherols in Virginia fanpetals (*Sida hermaphrodita* R.) herbage and silage. Virginia fanpetals biomass was harvested at two dates. The average  $\beta$ -carotene content of herbage was 584.02 mg/kg DM.  $\beta$ -carotene concentrations were higher in second-harvest herbage and silage (P $\leq$ 0.01). The  $\beta$ -carotene content of silage was approximately 0.27 and 0.9 lower in comparison with first-harvest and second-harvest herbage, respectively. The average  $\alpha$ -tocopherol content of herbage was 72.85 mg/kg DM, and it was higher at the second harvest (P $\leq$ 0.01). Herbage contained high concentrations of  $\beta$ -tocopherol (52.01 mg/kg DM on average) and  $\gamma$ -tocopherol (29.29 mg/kg DM on average).  $\alpha$ -tocopherol loss during fermentation reached 0.2 and 0.18 in silage from the first and second harvest, respectively. The content of  $\beta$ -carotene,  $\alpha$ -tocopherol and vitamin E Eq was higher in second-harvest herbage than in first-harvest samples (P $\leq$ 0.01). The average losses of the analyzed ingredients in silage did not exceed 0.20 of their concentrations in herbage.

#### Introduction

Virginia fanpetals (*Sida hermaphorodita* R.) is a perennial plant that provides biomass rich in biologically active substances used in pharmacology. It can also be used as a honey crop, an energy crop for biogas production and, to a limited extent, as a source of fodder for livestock (Dinda et al., 2015). Before the biomass of Virginia fanpetals can be used as animal feed, it should be analyzed for the content of basic nutrients and biologically active compounds. Carotenoids and tocopherols supplied by green fodder are active antioxidants and biomarkers of nutritional status in livestock (Nozière et al., 2006).  $\beta$ -carotene is the most important provitamin A carotenoid. In the group of eight natural tocochromanols ( $\alpha$ -,  $\beta$ -,  $\gamma$ -,  $\delta$ -tocopherols and  $\alpha$ -,  $\beta$ -,  $\gamma$ -,  $\delta$ -tocopherol is the most active form of vitamin E.  $\alpha$ -tocopherol predominates in leaves, whereas grain and seeds are also abundant sources of  $\beta$ -,  $\gamma$ - and  $\delta$ -tocopherols. Feed preservation (drying, ensiling) leads to considerable losses (0.10-0.80) of provitamins and lipophilic vitamins (Nozière et al., 2006). The objective of this study was to determine the vitamin content of Virginia fanpetals (*Sida hermaphrodita*) herbage and silage and to test the research hypothesis that they can be valuable sources of carotenoids and tocopherols in ruminant nutrition.

### **Materials and Methods**

The experimental materials comprised Virginia fanpetals herbage from the first harvest (11 June 2015) and second harvest (23 September 2015), collected with a forage harvester equipped with a crusher (straw length - 10 mm). Herbage was ensiled in plastic 220 l containers. After 90 days, silage samples were collected with a probe along the entire length of the container. Samples of fresh herbage and silage (immediately after silo opening) were ground in the Retsch SK 100 mill for fibrous materials, and were assayed for the content of  $\beta$ -carotene (Rodriguez-Berlando de Quirós et Costa 2006) and tocopherols ( $\alpha$ ,  $\beta$ ,  $\gamma$ ,  $\delta$ ) (PN-EN ISO 6867: 2002). Vitamin E activity was expressed as the following equivalents:  $\alpha$ -T=1,  $\beta$ -T=0.4,  $\gamma$ -T=0.1,  $\delta$ -T=0.01 vitamin E Eq (mg/kg DM) = C<sub>1</sub>+0.4C<sub>2</sub>+0.1C<sub>3</sub>+0.01C<sub>4</sub>, C<sub>1</sub> content  $\alpha$ -T (mg/kg DM), C<sub>2</sub> content  $\beta$ -T (mg/kg DM), C<sub>3</sub> content  $\gamma$ -T (mg/kg DM), C<sub>4</sub> content  $\delta$ -T (mg/kg DM) (Eittenmiller et al., 1998). The results were analyzed statistically using STATISTICA ver. 12.0 software. The significance of differences between mean values was determined by Duncan's test.

#### **Results and Discussion**

There are no published data on the levels of carotenoids and tocopherols in animal feed made from Virginia fanpetals biomass. Therefore, we compared our results with the findings of other authors who analyzed vitamin content in the most common types of green fodder. The average  $\beta$ -carotene content of Virginia fanpetals herbage (584.02 mg/kg DM, Table 1) was similar to that reported for grasses and grass-legume mixtures (33-700 mg/kg DM) (Prache et al., 2003, Dunne et al., 2009), and higher than that reported for legumes (148-235 mg/kg DM) (Larsen et al., 2012).  $\beta$ -carotene concentrations were 0.18 higher in second-harvest herbage than in first-harvest herbage (P $\leq$ 0.01, Table 1). Carotenoid levels in herbage increase until the beginning of flowering, and decrease during the growing season. The average  $\beta$ -carotene content of silage (484.72 mg/kg DM) was approximately 0.27 and 0.9 lower in comparison with first-harvest and second-harvest herbage, respectively.  $\beta$ -carotene content was approximately 0.34 higher in second-harvest silage than in first-harvest silage (P $\leq$ 0.01).  $\beta$ -carotene loss during fermentation reached 0.18 on average. According to Nozière et al. (2006),  $\beta$ -carotene losses during the ensiling process may be as high as 0.80, but they usually do not exceed 0.20 of initial  $\beta$ -carotene concentrations in herbage. Average  $\alpha$ -tocopherol content (72.85 mg/kg DM) was higher in second-harvest herbage than in first-harvest herbage (P $\leq$ 0.01, Table 1); the noted values were also higher than those reported for clover (27 mg/kg DM) and grass-legume mixtures (55 mg/kg DM) (Larsen et al., 2012), and comparable with those observed in alfalfa (36-137 mg/kg DM) (Turner et al., 2002, Larsen et al., 2012).

## **BACK**

average content of  $\beta$ -tocopherol (52.01 mg/kg DM) and  $\gamma$ -tocopherol (29.29 mg/kg DM) in Virginia fanpetals herbage was considerably higher than in grasses and legumes (Larsen et al., 2012), and similar to their concentrations in sorghum ( $\beta$ -tocopherol - 87.5 mg/kg DM,  $\gamma$ -tocopherol - 37 mg/kg DM) (Chung et al., 2013). The content of  $\alpha$ -tocopherol (P $\leq$ 0.01) and vitamin E Eq (P $\leq$ 0.05) was higher in first-harvest silage than in second-harvest samples (Table 1).

		Herbage			Silage	
Parameter	1 <sup>st</sup> harvest	2 <sup>nd</sup> harvest	SEM	1 <sup>st</sup> harvest I	2 <sup>nd</sup> harvest	SEM
	n=5	n=5		n=5	n=5	
Dry matter (g/kg)	210 B	324 A	10.15	160 B	301 A	13.12
β- carotene	525.9 B	642.2 A	19.35	386.7 B	582.8 A	32.75
α-tocopherol	70.1 B	75.6 A	1.04	68.7 A	61.7 B	1.45
β-tocpherol	56.21 b	61.39 a	1.24	8.18 B	14.04 A	1.03
γ-tocopherol	33.5 A	25.1 B	1.64	32.6 A	25.3 B	1.37
δ-tocopherol	5.5 B	9.7 A	0.72	2.5 B	9.9 A	1.24
Total tocopherols	165.3	171.8	1.67	112.0	111.0	1.61
Vitamin E Eq	96.0 B	102.8 A	1.43	75.3 a	70.0 b	1.24

**Table 1** The content of  $\beta$ -carotene (mg/kg DM) and tocopherols (mg/kg DM) in Virginia fanpetals (*Sida hermaphrodita R*) herbage and silage from the first and second harvest

Significance levels:  $a,b - P \le 0.05 A, B - P \le 0.01$ 

 $\alpha$ -tocopherol loss during fermentation reached 0.2 and 0.18 in silage from the first and second harvest, respectively.  $\beta$ -tocopherol concentrations were substantially lower in silage than in herbage (Table 1). The average value of vitamin E Eq was approximately 0.27 higher in herbage than in silage (99.37 vs. 72.62 mg/kg DM) (Table 1).

#### Conclusions

Virginia fanpetals herbage from the second-harvest was a rich source of  $\beta$ -carotene and tocopherols. Second-harvest silage had a higher  $\beta$ -carotene content, a lower  $\alpha$ -tocopherol content and a lower vitamin E Eq value than first-harvest silage. The average losses of  $\beta$ -carotene and  $\alpha$ -tocopherol in silage did not exceed 0.20 of their concentrations in herbage. Virginia fanpetals herbage and silage contained high concentrations of  $\beta$ -,  $\gamma$ - and  $\delta$ -tocopherols, which affected the values of vitamin E Eq.

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# Ruminal Degradability of Dry Matter and Crude Protein from Virginia Fanpetals (*Sida Hermaphrodita* Rusby L.) Herbage and Silage

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## Introduction

Virginia fanpetals (*Sida hermaphrodita* Rusby) is a perennial plant of the family *Malvaceae*, grown mainly as an energy crop (Borkowska and Styk, 2006). Due to its high growth potential, low soil requirements and drought resistance, Virginia fanpetals has attracted attention as a potential valuable source of livestock fodder (Kwiatkowski, 2012). Previous research involving animals has focused on the use of Sida biomass in dry form (Tarkowski, 2006). There are no data available on the ruminal degradability of Sida herbage and silage.

The aim of this study was to determine the effect of ensiling fresh biomass of Virginia fanpetals on the degradability of silage dry matter (DM) and crude protein (CP).

#### **Materials and Methods**

The experimental materials comprised Virginia fanpetals (*Sida hermaphrodita* Rusby) herbage from the first harvest (budding stage, 11 June 2015), collected with a forage harvester equipped with a crusher. Herbage was ensiled in 220 l plastic containers, in the following treatments (each in triplicate): without additives (KO), with the addition of 2 kg of straw placed at the bottom of the container (KD), with the addition of inoculant (*Lactobacillus plantarum, Lactobacillus buchneri, Lactococcus lactis*) at 2.5 · 10<sup>8</sup> CFU/kg (KI). After 90 days, silage samples were collected with a probe along the entire length of the container. Herbage and silages samples were analyzed for proximate chemical composition (AOAC, 2005), the content of water-soluble carbohydrates (WSC) – by the anthrone method, NDF, ADF, ADL. Silage samples were assayed for the content of lactic, acetic and butyric acids - by high-performance liquid chromatography (HPLC), and ammonia nitrogen - by the Conway method.

The effective degradability (ED) of DM and CP, and degradation parameters (*a*, *b*, *c*) were determined by the in situ method proposed by Michalet-Doreau et al. (1987) on three non-lactating cows fitted with ruminal cannulas. The curves of DM and CP disappearance in the rumen were fitted to the model of Ørskov and McDonald (1979) at a ruminal outflow (k) of 0.06 h<sup>-1</sup> using non-linear regression procedures (SAS 1995). The ED of DM and CP and the digestibility rate constants (*a*, *b*, *c*) were calculated as: ED = a + (bc) / (c + k), where: *a*-instantly degradable fraction, *b*-slowly degradable fraction, *c*-rate of degradation (% h<sup>-1</sup>), k = 0.06 h<sup>-1</sup>.

#### **Results and Discussion**

The ensiling of Sida biomass and the applied additives contributed to an increase in the soluble fraction (*a*) and a decrease in the insoluble but potentially degradable fraction (*b*) of DM and CP (P<0.01), thus increasing the content of readily soluble fermentation products. The ensiling of Sida biomass enhanced the rate of DM degradation in the first 4 hours of incubation, which dropped below that noted in the raw material after 24 hours (P<0.05).

silage [g/kg D	DM]				
	Harbaga	Control	Drained	Inoculat	SEM
	Herbage	silage	silage	ed silage	SEM
DM [g/kg]	197	160	213	205	13.1
СР	199	193	175	207	7.72
WSC	73.0	6.00	23.0	10.0	4.37
NDF	403	423	394	369	23.4
ADF	308	371	339	303	18.5
ADL	38.0	61.0	56.0	55.0	2.27
pН		4,08	4.07	4.01	0.015
Lactic acid		125	109	112	2.77
Acetic acid		21,4	17.6	18.6	0.48
Butyric acid		9.29	7.84	8.21	0.50
N-NH <sup>3</sup> [g/kg	total N]	110	73.6	50.1	4.90

 Table 1
 Chemical composition of Virginia fanpetals herbage and silage [g/kg DM]

Throughout ruminal incubation, the rate of silage CP degradation was higher than that of herbage CP degradation. As a result, silage was characterized by higher ED values (P<0.01) at a similar rate of degradation. The highest ED of DM and CP (P<0.01) was noted in Sida biomass ensiled with the addition of inoculants. An increase in the DM content of Sida biomass enhanced DM and CP degradability in the first 16 hours of incubation in the rumen, whereas reduced degradability was observed during the next hours, relative to control silage. However, the lowest ED of DM and CP (P<0.01) was noted in control silage.

**BACK** 

degrad	dability paramet	ers [%]			degradability parameters [%]				
	Dry	matter					Dry matter		
	Herbage	Control silage	SEM			Control silage	Drained silage*	Inoculated silage	SEM
0	24.62B	31.46A	1.54		0	31.46Bb	34.17a	37.48A	0.89
2	32.35B	37.37A	1.19		2	37.37 Bb	40.37a	42.06A	0.77
4	35.45b	38.52a	0.78		4	38.52B	52.38A	48.97A	2.15
8	56.30	53.78	1.55		8	53.78C	59.74B	66.53A	1.91
16	61.05	57.95	1.07		16	57.95B	66.26Ab	72.34Aa	2.17
24	71.17a	67.99b	0.87		24	67.99B	67.08B	75.44A	1.37
48	76.08a	74.69b	0.35		48	74.69B	71.43C	78.75A	1.06
а	23.82B	30.88A	1.59		а	30.88C	34.60B	36.45A	0.83
b	54.38A	43.32B	2.51		b	43.32A	36.81B	42.13A	1.04
с	0.09	0.07	0.006		С	0.07Ba	0.13A	0.12b	0.01
ED	55.58	54.67	0.63		ED	54.67C	59.61B	64.74A	1.48
	Crud	e protein					Crude protein		
0	31.63B	68.51A	8.25		0	68.51A	68.48A	65.55B	0.53
2	44.32B	72.69A	6.35		2	72.69Ab	74.42Aa	69.97B	0.69
4	46.79B	74.03A	6.09		4	74.03B	79.72A	75.43B	0.88
8	69.34b	83.35a	3.42	_	8	83.35B	84.30b	86.78Aa	0.57
16	74.90B	84.79A	2.31		16	84.79B	85.50B	88.97A	0.66
24	84.26b	89.44a	1.29		24	89.44Ab	86.84B	91.10Aa	0.65
48	87.84B	90.97A	0.72		48	90.97a	88.83Bb	91.67A	0.48
а	31.71B	68.17A	8.16		а	68.17B	68.45A	64.88C	0.57
b	57.11A	21.23B	8.03	_	b	21.23B	18.37C	26.53A	1.20
С	0.11	0.12	0.007		С	0.12C	0.28A	0.17B	0.02
ED	67.95B	82.22A	3.23		ED	82.22C	83.54B	84.49A	0.34

**Table 2** The effect of ensiling on ruminaldegradability parameters [%]

 Table 3
 The effect of silage additives on ruminal degradability parameters [%]

\*-straw at the bottom of the container used for drainage; *a*-instantly degradable fraction; *b*-slowly degradable fraction; *c*-rate of degradation (%  $h^{-1}$ ); ED-effective degradability; SEM-standard error of the mean; <sup>a,b</sup>-significance at P<0.05; <sup>A,B,C</sup>-significance at P<0.01

## Conclusion

Virginia fanpetals biomass harvested in the budding stage is characterized by DM degradability similar to that of alfalfa and red clover harvested at the beginning of flowering, and higher CP degradability. Ensiling had no effect on DM degradability at a similar rate of degradation (parameter c), whereas it significantly increased the ruminal degradability of CP. The use of drainage and inoculant increased the ED of silage DM and CP.

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## Acknowledgements

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# Forage Production from Genetically Modified Crops

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## Introduction

Feeds are the important point of the entire food chain. Rapidly changing climate requires rapid introduction of new products that would meet the requirements for a good feed. According to Chrenková (2014), the main objective of the development of new products is high and stable yields with low need for resources such as water, nutrients and minerals. Genetically modified plants are the largest sector of biotechnology utilization in agriculture today. The plants most grown are soy resistant to herbicides, maize tolerant to herbicides and pests, cotton, oil rape, and sugar beet.

In 2006, Slovakia became the 21st country in the world to start utilizing GMPs. Slovak agriculture faces the same needs (plants tolerant to heat and drought, crops with added value) and issues (protection against pests and weeds) as other countries which exploit the advantages of GMPs in the world and in EU. Besides Slovakia, in Europe also Spain, France, Romania, Portugal, Germany, Czech Republic, and Poland engage in GMP (Křistková, 2009; James, 2011).

The advantage of growing Bt maize is particularly simple and effective protection against the European corn borer, which results in lower losses and thus higher yields, better health of vegetation, lower extent fungal infections, fewer mycotoxins, unlodging and unbroken plants. Attention is ever more paid also to the environmental aspects, which suggest lower impact on the environment (no insecticids and chemicals) (Křístková, 2009; Čeřovská, 2007). Growing Bt maize, however, is also accompanied by disadvantages: higher expenses connected to the purchase of seed, market issues, higher administrative expenses.

#### **Material and Methods**

The aim of the research was to study the agrotechnology of genetically modified maize in practice. We compared the growing process of the GM maize, conventional maize treated against European corn borer, and untreated isogenic maize, and the economy of growing these strains. The experiment was conducted at an agricultural company Paseka, zemědelská, a.s. in Czech Republic. The seat of the company is in Babice pri Šternberku. The experiments were conducted in 2009 and 2013.

The plant material was divided between two parcels, one of which was divided in two by an unsown planter row. Three hybrids were used: GM maize MON 810, conventional maize (chemically treated against European crop borer), and isogenic maize (without chemical treatment against European crop borer). Seed dressing was used in all three hybrids. Growing technology remained the same during both years of the experiment. There were minimal distances between the planting parcels, therefore all studied hybrids were subjected to the same weather conditions. The fore crop on both croplands in 2009 was common wheat, in 2013 hordeum was used. In autumn after the fore crop harvest, skimming was done with Horsch Terrano plow. The cropland was then fertilized with mineral fertilizers Amofos in a 100 kg.ha<sup>-1</sup> dosage (12 kg N and 52 kg P<sub>2</sub>O<sub>5</sub>) and potassium salts fertilizers in a 100 kg.ha<sup>-1</sup> dosage (60 kg K<sub>2</sub>O). In Spring, before sowing, ammonium nitrogen fertilizers were applied to the field. Directly before sowing, urea fertilizer in dosage 200 kg.ha<sup>-1</sup> (92 kg N) and NPK 15:15:15 in dosage 200 kg (30 kg N, 30 kg P<sub>2</sub>O<sub>5</sub>, 30 kg K<sub>2</sub>O) were applied to the cropland. Embedding was done the same day by Neptun compactor. All three hybrids were sown using Monosem plow. The plow was set to sow 80,000 seeds per ha for every hybrid. After sowing herbicide Guardian Extra was applied in 3.5 1.ha<sup>-1</sup> dosage. Insecticide was applied only to the conventional maize hybrid.

#### **Results and Discussion**

Searching for the opportunities to increase maize yield in practice leads to experimentation with planting density. For our experiment, we chose the distance of 75 cm between rows and 16,6 cm between the seeds in the row. The planting density was thus 80,000 individual plants per ha. One of the parameters we wanted to study in this experiment was one of the characteristics of the seed itself: seedling emergence.

For emergence determination, the number of plants for 10 meters of a row was used rather than the number of plants per 1m<sup>2</sup>. Comparing the two years, minimal difference was observed between the sown seed and the number of plants emerged of all three maize hybrids.

One of the parameters which forms the yield of a plant is the number of the rows of kernels on an ear. We compared the number of rows on an ear for 100 plants in row for each of the hybrids. The resulting average numbers of rows are presented in Table 1.

This parameter is to a significant degree limited by the conditions such as temperature, water supply, diseases and pests. These conditions were identical for all the studied hybrids during the experiment. Therefore the determined values are very similar.

The number of kernels per row on an ear is a parameter which forms at the ten leaf stage. Its main limiting factor is the lack of water. As in the number of rows, the number of kernels in a row on an ear was also determined on a sample of 100 plants from each studied hybrid (Table 1).

 Table 1
 The yield of biological material in years 2009 and 2013 (average values)

		2009		2013			
n=10	GMO	conventional	isogenic	GMO	conventional	isogenic	
Plants emerged in a row	59.9	59.6	60.2	59.8	59.3	59.6	
Kernel rows on an ear	14.2	14.0	14.3	14.3	14.3	14.1	
Kernels in a row on an	31.5	30.9	31.6	31.3	30.3	30.7	
Kernels on an ear	451.5	431.8	452.3	447.2	433.0	432.9	

The dependency of number of kernels per row on the water supply was confirmed in this experiment by comparing the results between the two years. The differences between the highest and lowest value determined for this parameter were in the first year 8, 7 and 12. In the second year those differences were 5, 4 and 5. The second year of the experiment had more favorable rain condition, which reflected in the higher number of kernels in a row.

The number of kernels per ear is the resulting parameter dependent on the number of rows per ear and kernels per row on an ear. The number of kernels on an ear indicates the conditions under which the maize is grown. Under optimal conditions (sufficient nutrients and water supply, optimal growth density) maize forms long ears covered in kernels to the tip with a high number of kernels. This is negatively affected by stress from the lack of nutrients and water or too dense growth. The results is then a short ear with empty tip. The number of kernels per row is also affected by pollination quality. As in the previous two parameters, the number of kernels per ear was also determined for sample of 100 plants for each of the three hybrids.

The differences in the average number of kernels per ear between the studied hybrids were minimal. The highest number of kernels per ear was determined in GM maize and isogenic maize in the first year and in GM maize in the second year. Yield is dependent not only on biological material, plant nutrition, irrigation, soil characteristics, weather conditions, but on the disease and pest infection rate as well. The interplay of these factors is significant and determines the yield of the hybrid per locality.

In year 2009 the highest yield (16.8 t.ha<sup>-1</sup>) was contributed by the isogenic material, the lowest yield (15.9 t.ha<sup>-1</sup>) by the conventional material. The highest yield in year 2013 was determined in the GM maize: 16.1 t.ha<sup>-1</sup>, which was 12.31 t.ha<sup>-1</sup> for 14% humidity. Conventional hybrid yielded 15.9 t.ha<sup>-1</sup> (12 t.ha<sup>-1</sup> for 14% humidity). Finally, for isogenic maize the yield was 14.8 t.ha<sup>-1</sup>, which was 11.2 t.ha<sup>-1</sup> for 14% humidity.

One of the most significant issues in growing maize are *Fusarium* infections, which under favorable conditions produce mycotoxins. The *Fusarium* infection rate and the rate of infection by European corn borer was determined on a sample of 100 plants in a row for every hybrid studied (Table 2).

Measured parameters	2009 2013					
	GMO	convention	isogenic	GMO	conventional	isogenic
Number of ears infected by	23	47	19	6	33	50
Fusarium alone	23	34	4	6	15	21
Fusarium + European corn borer	0	13	15	0	18	29
European corn borer alone	0	6	2	0	7	18
European corn borer total	0	19	17	0	25	47

 Table 2
 Mycoses evaluation in connection to European corn borer infection

The results suggest that the *Fusarium* infections are not always determined by the European corn borer infection. Conventional maize shown approximately the same number of ears affected by *Fusarium* infections accompanied by European corn borer infection as the isogenic hybrid, but almost eight times as many ears infected by *Fusarium* only as the isogenic maize. In both 2009 and 2013 no European corn borer infection was determined in GM maize. *Fusarium* infections was determined in the GM hybrid, which suggests that this hybrid is not entirely resistant to those infections. However, the rate of *Fusarium* infection was lower than the average rate in the other two hybrids.

High content of Fumonisin mycotoxin was determined in the isogenic maize (Table 3). Similar results were observed by Bakan et al. (2002), who determined 4 to 18 times lower rate of fungal infestation in Bt-maize compared to isogenic maize, which were both grown under natural conditions.

Table 3	Mycotoxin	Fumonisin	content	(µg/kg`	) in maize	kernels

	2009	2013
GMO	0	0
conventional	520	590
isogenic	1 250	1 630

Gene-based plant resistance belongs among the most economic and also the most ecologic methods of crop protection (Šudyová et al., 2011). When comparing the expenses for growing GM maize and conventional maize we considered seed, fertilizers, pesticides, and labor.

Compared to all other expenses, the seed purchase contributed the highest expense. GM maize seed was more expensive to purchase. One unit of GM maize is purchased for 152 €. Conventional maize was the least expensive to

purchase at the price of 147  $\in$  per unit. Both hybrids compared were fertilized by the same fertilizer from the same shipment and the same package. The total expenses for fertilizers were 106  $\in$  per ha. To all three hybrids the same herbicide, Guardian Extra in 3.5 1.ha<sup>-1</sup>, for 26  $\in$  was applied. Insecticide Inegro in dosage 0.6 1.ha<sup>-1</sup> for 41  $\in$  was used for the conventional hybrid only. Thanks to modern machinery and technology with high efficiency, the labor contributed the least significant expense. GM hybrid was not sprayed against European corn borer and therefore one less spraying, and one less water shipment, was necessary compared to the conventional hybrid as presented in Table 4. All other labor expenses were identical.

Item		GMO maize	Conventional maize
		Expe	nses in €/ha
Seed	Price for unit	152	147
Fertilizer	urea	44	44
	NPK 15:15:15	62	62
Chemicals	Guardian Extra	26	26
	Integro		41
Labor	skimming	3,6	3,6
	fertilizer distribution	6,5	6,5
	fertilizer distribution	6,5	6,5
	compacting	6,8	6,8
	sowing	28	28
	spraying	23	23
	water shipment	2,5	2,5
	spraying		23
	water shipment		2,5
	threshing	97	97
Expenses in	€/ha total	457,9	519,4

 Table 4
 Economic evaluation

The total sum of all expenses for the GM hybrid was  $457.9 \notin$ , which is  $61.5 \notin$  less compared to the conventional hybrid. These expenses were lower in the GM maize despite the higher initial expenses for obtaining seed. The total expenses for the conventional hybrid were  $519.4 \notin$ . These results are comparable also to those of Boroš (2008).

#### Conclusion

Minimal difference between the seed sown and plants emerged was determined in all three hybrids. The differences between the average number of rows per ear, kernels in row per ear, and kernels per ear were minimal between the hybrids. GM maize showed high yields. No European corn borer infection was determined in the GM maize. Plants with *Fusarium* infection were present among the GM maize, but the rate of this infection in GM maize was lower than the average *Fusarium* infection rate in the other hybrids compared.

In economic comparison of the conventional and GM maize hybrids we determined that the sum of all expenses for GM maize is  $457.9 \in$ , which is  $61.5 \in$  less than the sum of expenses for the conventional hybrid. These expenses were lower in the GM maize despite the higher initial expenses for obtaining seed. The total expenses for the conventional hybrid were  $519.4 \in$ .

## Acknowledgments

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# SESSION 2 – CONTROL OF FERMENTATION PROCESS, MICROBIOLOGY AND HYGIENIC QUALITY OF CONSERVED FEEDS

Oral presentations

<u>Global warming and forage preservation – forages and treatments</u> Marley, G.

<u>Effect of silage additives on fermentation process of clover-grass mixture</u> Rajčáková, Ľ., Gallo, M., Poláčiková, M., Britaňák, N.

<u>Re-ensiling of translocated triticale silage: to inoculate or not to inoculate?</u> Solórzano, L. C., Solórzano, L. L., Rodríguez, A. A.

<u>Effectiveness of various silage additives used in ensilage of sorghum grain and forage hybrid</u> Król, B., Słupczyńska, M., Sowiński J., Bodarski, R., Mońka ,M.

<u>Changes in chemical composition and quality parameters of sorghum grain variety</u> (12GS9014) whole plants ensiled with application of various silage additives Słupczyńska, M., Król, B., Sowiński, J., Bodarski, R., Mońka, M.

<u>Effects of seven formic acid based additives on grass silage fermentation and aerobic stability</u> Rinne, M., Kuoppala, K., Mäki, M., Seppälä, A., Jalava, T.

Effects of different sodium nitrite-containing additives on dry matter losses, fermentation pattern and biogenic amine formation in lucerne and cocksfoot silage Auerbach, H., Nadeau, E., Weiss, K., Theobald, P.

Improving of aerobic stability of maize silage by different additives Jambor, V., Vosynková, B.

Effects of dual purpose inoculants on fermentation parameters, aerobic stability and safety of whole crop maize silage after a short time of storage Jatkauskas, J., Vrotniakiene, V.

Oxygen barrier ('Silostop') stretch-wrap film improves hygienic quality of baled silage Lewis, D., Stein, K., Chamberlain, A. T., Banchero, C., Wilkinson, J. M.

# **Poster presentations**

Effect of *Lactobacillus kefiri* alone and in a silage inoculant blend on the fermentation quality of grass silage Boczonadi, A., Kesselring, J., Boeck, G., Schoendorfer, K., Hoeger, T., Schatzmayr, G.

<u>Effect of additives on Lucerne silage with high dry matter</u> Loučka R., Homolka P., Jančík F., Kubelková P., Tyrolová Y., Výborná A.

Fermentation of early cut whole crop field beans (Vicia faba) affected by silage additives Römer, G., Kalzendorf, C., Milimonka, A.

Effects of delayed filling and additive use on the quality of pressed sugar beet pulp ensiled in plastic bags Auerbach, H., Weber, G., Nadeau, E., Weber, U., Weiss, K., Potthast, C.

<u>Effect of silage additives on fermentation characteristics of maize silage</u> Tyrolová, Y., Bartoň, L., Loučka, R.

The effect of Sil-All Maize+ FVA on pastone silage Carcano, P., Hocke, N., Marley, G.

The possibility of wet maize corn conservation with chemical additive Bodarski, R., Hikawczuk, T., Słupczyńska, M., Król, B., Nałęcz, M., Mońka, M.

<u>A meta-analysis: effect of lime on the chemical composition, fermentation, and aerobic stability of sugarcane silages in Brazil</u> Rabelo, C. H. S., Härter, C. J., Mcallister, T. A., Reis, R. A.

<u>A meta-analysis: effect of heterofermentative inoculants applied at different dry matter</u> <u>contents on the fermentation patterns and aerobic stability of sugarcane silages</u> Rabelo, C. H. S., Härter, C. J., Reis, R. A.

# Global Warming and Forage Preservation – Forages and Treatments

**BACK** 

#### MARLEY, G.

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#### Introduction

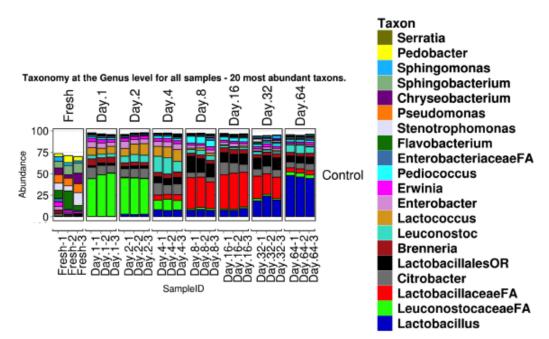
Climate change is an accepted fact irrespective of it being cyclical or as a direct result of greenhouse gases. Multiple recent articles (Ray, Olesen, Tao etc) continue to report on the proven impact of climate change on crop production with Ray reporting that between 32-39% of global yield variability is accounted for through climate variability, and that upto 60% of the yield variability in global breadbaskets are accounted for through climate variation, with over 50% of maize, rice, wheat and soybean production areas directly impacted by climate variation.

#### Fermentation

Enormous challenge exists in meeting human food production demands over the coming decades. Efficiency of food production is paramount to farm efficiency and, with crop production subject to climatic impact the price of cereals, milk, dairy and beef production is equally subject to climatic impact.

Fermentation is driven through a fluctuating microbial population that differentially dominates as the pH and atmosphere of the bunker changes. Chart 1 presents a typical metagenomic analysis of the epiphytic progression during ensiling of 20 grass clamps.

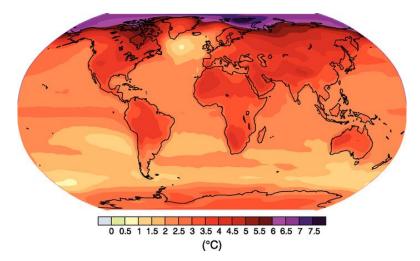
A rapid progression from *Leuconostocaceae FA* to *Lactobacillaceae FA* to finally *Lactobacillus* dominating the silage is observed



During the initial days of ensiling the forage and associated bacteria undergo a dramatic environmental change with forage switching from aerobic to anaerobic, increasing in temperature by  $10+^{\circ}C$  (dependent on density) and finally the acidity of the forage increasing 1000 fold – the dramatic environmental change dictating different bacterias dominating at differing stages of fermentation.

Ensiling temperatures vary across the globe with some countries silage production not likely to be impacted due to their cooler climate, however countries with ensiling temperatures currently above 27°C will be impacted by the ability of bacteria to survive and dominate in temperatures exceeding 40°C. This is already seen in some states in the US where ensiling temperatures are 35°C and forage temperatures increase by 10°C as oxygen is converted to carbon dioxide. The high temperatures immediately post ensiling are above the thermal death point of many epiphytic and inoculant lactic acid bacteria resulting in the more thermally stable spoilage bacteria dominating the initial fermentation and the production of highly butyric silage. Projections of temperature rises fluctuate depending on the exact model adopted though all models predict a minimum average 3°C rise over the coming 50 years across mainland Europe.

Increasingly in Central and Easter Europe silages that are subject to a limited fermentation are being observed (personal observation across Europe), with higher final pH, lower lactic acid, higher acetic acid, greater ammonia levels and also elevated levels of butyric acid where dry matter, treatment, compaction of forage and residual sugar levels would normally suggest a fuller fermentation with significantly lower butyric acid.



Thermal death points of bacterial species are strain specific but generally within a species the thermal death point of bacteria is within 2-3°C range. Bergey's quotes the following thermal death points for common silage fermentation bacteria:

Bacteria	Absolute Thermal	Growth at		
	Death Point °C	40°C	50°C	
L. plantarum	42°C	Yes	No	
L. casei		Yes	No	
L. brevis	<45°C	Yes	No	
L. buchneri	<45°C	Yes	No	
E. faecium	<45°C	Yes	No	
P. acidilactici	>60°C	Yes	Yes	
P pentosaceus	>60°C	Yes	Yes	

Most commonly used inoculant *Lactobacillus spp* have a thermal death points below 45°C, meaning that if average current ensiling temperature is in the region of 30°C with the increase in forage temperature during the initial ensiling period and the projected increase in global temperature the temperature of the forage will be inhibitory to lactic acid fermentation by epiphtic lactic acid bacteria and many inoculant bacteria resulting in more Clostridial silage, greater losses, reduced intake and reduced profitability.

## Conclusion

Farming is obliged to adapt to feed a growing population but it also needs to recognise the impact of global warming on the quality of silage that can be made. Global warming impacts crop growth but will also directly impact efficacy of fermentation and the ability of the farm to effectively produce home produced feed. The inoculant industry as a whole must react in order to ensure this issue is effectively communicated to the dairy and beef industry to ensure the ongoing safe and efficient production of silage.

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# Effect of Silage Additives on Fermentation Process of Clover-Grass Mixture

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## Introduction

In Slovakia, clover-grass mixtures are is grown mainly in the north. Due to its low requirements on soil quality and climate conditions, it represents a good alternative to growing alfalfa. However, the frequent rains during the harvest period, in particular during the first harvest, make the conservation of the mixture difficult. According to Herzog et al. (2014) the quality of the clover-grass fermentation process depends on the microbiology of the conserved forage as well as the level of its pressing. The necessity of ensuring quality fermentation process is brought forth also by Vrotniakiene et al. (2015), who examined the influence of microbial additive on the fermentation of alfalfa, clover, and clover-grass mixture. When ensilaging clover with low dry matter content, Tuori et al. (2012, a) recommends treating the silage with chemical additives as the bacterial additive treatment brought no positive results.

The objective of this study was to evaluate the options for the conservation of clover-grass mixture harvested at the end of the vegetation season.

#### Materials and method

The experiment was conducted on a mixture of 40 % clover and 60 % grass grown at 730 meters above sea level, from the late autumn harvest.

The harvest clover-grass mixture was wilted for 36 hours, chopped and after homogenization ensilaged in laboratory silos of 1.7 l content. The fermentation process during ensilaging was observed in one control variant (C) without treatment of the mixture and two experimental variants, in which the ensilaged matter was treated with additives:

T1 (biological silage additive, *Lactobacillus plantarum* DSM 3676 and 3677; the application rate was 2 l/t) and T2 (chemical additive, 24.4 % of sodium nitrite (E 250) and 16 % of hexamine; the application rate was 2.5 l/t).

During fermentation, the silages were stored in a dark room at temperature  $20 - 22^{\circ}$  C. The experiment finished 180 days after ensilaging and parameters of the fermentation process were determined in silage samples. Samples of clovergrass matter as well as silage samples were chemically analysed. The results were statistically processed by one-way analysis of variance, by the ANOVA multifactorial procedure and by the subsequent POST-HOC Tukey test.

#### **Results and Discussion**

Harvested mixture (Table 1) has lower dry matter content, good crude protein content, with lower content of crude fibre but higher content of total sugar and ash.

Item	g/kg DM	Item	g/kg DM							
Dry matter	276.20	Fat	21.39							
Crude protein	162.09	Ash	104.67							
Crude fibre	228.95	Total sugars	93.83							
ADF	290.79		MJ/kg DM							
NDF	434.28	NEL	6.1							

 Table 1
 Nutrition value in wilted clover-grass mixture

The fermentation process of the untreated clover-grass mixture can be evaluated as good (Table 2). It was characterized by lower pH, high lactic acid content, good acetic acid and butyric acid content, but also higher content of ammonium-N of total N.

The only positive impact of the bacterial inoculant application in comparison to the untreated silage was observed in the ammonium-N concentration. All other parameters of the fermentation process were on the same levels as in the untreated silage.

Application of the chemical additive compared to the untreated silage led to decrease of fermentation losses, high pH and high acetic acid content and lower content of lactic acid, butyric acid, and capronic acid, and decreased ammonium-N content of total N.

In nutrient content (Table 3), no significant differences were observed with the exception of the content of dry matter and total sugar.

Our results are similar to those presented by Tuori et al. (2012, b), who examined the influence of bacterial and chemical treatment on the fermentation process of red clover and alfalfa in different mixtures. Like us, these authors determined minimal differences between the untreated control variant and the matter treated with bacterial additive. High lactic acid content in the produced silages was likely caused by the higher sugar content in the conserved matter. Its levels reflected also in the lower pH. Chemical additive application led to a decrease of butyric acid content and the content of ammonium-N of total N. Similar results were observed for these parameters as in our previous study Gallo et al. (2008).

**BACK** 

	Contro	ol	Treatme	nt1	Treatment 2	
Item	$\frac{-}{x}$	SD	$\frac{-}{x}$	SD	$\frac{-}{x}$	SD
Losses of DM (%)	0.84 <sup>a</sup>	0.03	0.73 <sup>ab</sup>	0.18	0.72 <sup>b</sup>	0.04
pH	4.02 <sup>a</sup>	0.01	3.99 <sup>a</sup>	0.07	4.26 <sup>b</sup>	0.04
Acids (g/kg DM)						
- lactic	102.27 <sup>a</sup>	4.76	101.44 <sup>a</sup>	1.13	88.81 <sup>b</sup>	3.75
- acetic	18.93 ab	0.83	16.71 <sup>a</sup>	2.21	21.97 <sup>b</sup>	1.99
- propionic	0.21	0.11	0.09	0.02	0.15	0.04
- butyric + i.b.	0.75 <sup>a</sup>	0.11	0.73 <sup>a</sup>	0.21	0.32 <sup>b</sup>	0.09
- valeric + i.v.	0.32	0.05	0.29	0.16	0.32	0.15
- capronic + i.c.	0.23	0.13	0.05	0.02	0.04	0.00
VFA total (g/kg DM)	20.45 a	1.15	17.86 <sup>a</sup>	2.52	29.81 <sup>b</sup>	2.17
Alcohol (g/kg DM)	1.13	0.93	1.48	0.70	1.64	0.44
NH <sub>3</sub> -N of total N (%)	9.42 <sup>a</sup>	0.36	7.89 <sup>b</sup>	0.30	6.91 °	0.43

 Table 2
 Parameters of fermentation process in silages from clover-grass mixture

n = 6, DM – dry matter, VFA – voluntary fatty acids

Different letters <sup>a,b,c</sup> in the index numbers indicate significant (P <0.05) differences between

Item		Contr	ol	Treatmen	nt1	Treatment 2	
		$\frac{-}{x}$	SD	$\frac{-}{x}$	SD	$\frac{-}{x}$	SD
Dry matter	(g/kg FW)	261.42 a	3.30	266.82 a	1.43	270.71 <sup>b</sup>	3.43
Crude protein	(g/kg DM)	179.06	3.70	184.37	1.47	184.76	2.23
Crude fibre	(g/kg DM)	232.72	0.25	236.78	7.24	227.68	4.95
ADF	(g/kg DM)	288.46	10.72	285.40	2.40	272.80	8.92
NDF	(g/kg DM)	370.63	2.80	371.98	4.89	362.31	3.78
Fat	(g/kg DM)	31.98	0.68	31.15	0.69	32.12	0.97
Ash	(g/kg DM)	112.55	0.43	110.11	0.86	111.04	1.52
Total sugars	(g/kg DM)	13.99 <sup>a</sup>	0.42	20.21 <sup>b</sup>	0.40	17.26 <sup>ab</sup>	2.79
NEL	(MJ/kg DM)	5.89	0.01	5.90	0.01	5.92	0.02

 Table 3
 Nutrition value in silages from clover-grass mixture

n = 6, FW – fresh weight, DM – dry matter

Different letters <sup>a,b</sup> in the index numbers indicate significant (P <0.05) differences between

## Conclusion

The results of the study suggest that the fermentation process in conservation of clover-grass mixture harvested at the end of the vegetation season after wilting can be quite good. Application of bacterial inoculant treatment, compared to untreated matter, reduced the content of ammonium Nitrate of total Nitrate content, but it was unable to change the level of fermentation process. Application of chemical additive improved fermentation process of the silage matter, decreased the butyric acid content and the content of ammonium Nitrate of total Nitrate content.

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## Re-ensiling of Translocated Triticale Silage: to Inoculate or not to Inoculate?

**<u>BACK</u>** 

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#### Introduction

Silage has become a translocatable commodity as land scarcity increases near dairy farms. Today, it is a common practice to ensile crops near the location where they were produced and then transport them to the dairy farm for feeding on a daily basis. Silage may be transported upwards of 100 km to the farm where it will be fed to dairy animals. However, a question frequently asked by dairy producers is if the silage could be transported and re-ensiled at the dairy farm. The circumstances that may lead to re-ensiling at the dairy farm are several: 1) controlling forage inventories near-by as a consequence of sales/purchases of forages; 2) not having to depend on daily forage transportation; 3) removing the effect that inclement weather may play during transport and delivery of silage; 4) inventory consolidation to make room for new silage; 5) increase time available for other tasks during the busy harvest season and consolidate forage inventories during the cooler and less busy time of the year. The objective of this study was to determine the effect of re-ensiling triticale silage with or without the use of a homolactic bacterial inoculant (HBI) on nutritional and fermentation characteristics of the resulting silage.

#### **Material and Methods**

Whole plant triticale (x Triticosecale spp.) was chopped to a TLC of 20 mm and inoculated or not according to manufacturer's recommendation with HBI (supplying >9.1x1010 CFU/g containing *Lactobacillus plantarum*, *Enterococcus faecium*, *Lactococcus lactis*, *Pediococcus pentosaceus*, *Pediococcus acidilactici*). Triticale was fermented for 120 d at a temperature of 20-23°C using 3 L capacity PVC mini-silos fitted with two-way mechanics to vent gas. Four mini-silos per treatment were filled with about 2 kg of the crop at about 350g DM·kg<sup>-1</sup> FM and 52g·kg<sup>-1</sup> DM of soluble carbohydrates. After opening the silages, they were mixed within each treatment, sampled and the samples frozen at -18°C until analyzed. Silages were aerobically exposed (AE) for approximately 5 hours to simulate translocation and served as the test material for the present study.

After the 5 h aerobic exposure, half of the non-inoculated silage and half of the HBI silage were re-treated with inoculant as explained above, thus resulting in four experimental treatment combinations (First ensiling/Re-ensiling): 1) Non-inoculated/non-inoculated (N/N), 2) non-inoculated/HBI (N/HBI), 3) HBI/non-inoculated (HBI/N), inoculated/inoculated (HBI/HBI). The half of the silages that were not inoculated received a similar amount of water.

Silages were re-ensiled for 120 d at a temperature of 20-23°C using 0.946 L capacity glass mini-silos and vacuum sealed using a Food Saver FM2001 (Sunbeam Corp., Boca Raton, FL 33431 USA). Sixteen mini-silos (four/treatment) were filled with about 300 g of the silages at about 320 g DM·kg<sup>-1</sup> FM and contained no water soluble carbohydrates. Upon opening, nutritional characteristics were determined by Near Infra-Red Spectroscopy and fermentation characteristics were determined by wet chemistry at a commercial facility (Rock River Laboratory, Watertown, WI 53094 USA). Statistical analysis was performed using a completely randomized design (CRD). Tukey's Test was used for mean separation. Statistical significance was established at P<0.05.

#### **Results and discussion**

The DM content was similar (P>0.05) in the four silages after re-ensiling (average 308g DM·kg<sup>-1</sup> FM) and slightly wetter than the silages before the simulated relocation and consequent re-ensiling (average 316 g DM·kg<sup>-1</sup> FM). These results agree with those of Chen and Weinberger (2014) for wheat silages exposed to air for a similar time period prior to re-ensiling. Treatment did not affect (P>0.05) the contents of DM, starch, sugar, NFC and ash (Table 1), but HBI/N and HBI/HBI silages had higher (P<0.05) CP content, lower (P<0.05) ADF and aNDF contents compared with the silages from treatment combinations N/N and N/HBI. Chen and Weinberg (2014) reported no changes in NDF content of re-ensiled wheat silages exposed to air up to 48 h prior to re-ensiling, however, the present findings indicate that inoculation prior to first ensiling, but not at re-ensiling, was fundamental in preserving the protein fraction while decreasing the fibrous fractions. Both, the HBI/HBI and HBI/N silages had improved (P<0.05) contents of CP, ADF, and NDF compared with the N/N and N/HBI silages. Using HBI at first time of ensiling seems adequate to achieve the desired result. By contrast, the use of HBI at re-ensiling did not improve (P>0.05) any of the nutritional characteristics of triticale silage that had not been previously inoculated.

Re-ensiling resulted in triticale silages with lower (P<0.05) content of lactic acid (21.8 vs. 53.3 g·kg<sup>-1</sup> DM) and total VFA (75.5 vs. 91.9 g·kg<sup>-1</sup> DM) compared with the inoculated silage prior to aerobic exposure and re-ensiling (Table 2). By contrast to Kawamoto et al. (2011) who reported that the concentrations of lactic acid (% of DM) were increased after re-ensiling of Italian ryegrass irrespective of their DM content from 16 to 46 g·kg<sup>-1</sup> DM (Mild wilting) and from 4 to 29 g·kg<sup>-1</sup> DM (Heavy wilting), and with a consequent decline in pH. In the present case, re-ensiling resulted in silages with higher (P<0.05) contents of acetic acid (51.3 vs. 35.6 g·kg<sup>-1</sup> DM) and NH<sub>3</sub>-N (121.1 vs. 99.2 g·kg<sup>-1</sup> CP) and a higher pH (5.08 vs. 4.87) compared with the inoculated silage prior to aerobic exposure and re-ensiling, whereas similar (P>0.05) contents of propionic acid (2.6 vs. 2.9 g·kg<sup>-1</sup> DM) and ethanol (8.2 vs. 8.7 g·kg<sup>-1</sup> DM) were observed. Butyric acid was not detectable either before or after re-ensiling, which differs with the high levels reported by Chen and Weinberger (2014).

Treatment combinations did not affect (P>0.05) the contents of lactic, acetic, propionic and butyric acids nor total VFA (Table 3). However, Chen and Weinberger (2014) failed to find changes in pH, lactic acid, and acetic acid of reensiled wheat after up to 48 h of aerobic exposure. In that study ethanol content did not change significantly in silages that had been exposed to air prior to re-ensiling for a similar period as in the present study. These differences in the results between the two studies may be due to the higher moisture content of the wheat silage ensiled by Chen and Weinberger (2014). The HBI/HBI silage treatment resulted in the lowest NH<sub>3</sub>-N content which was lower (P<0.05) that of the N/N and N/HBI silages, but not the HBI/N silage. The HBI/N silage had a lower (P<0.05) NH<sub>3</sub>-N compared with the N/HBI silage but did not differ from the N/N silage. Inoculation before the first ensiling (HBI/N and HBI/HBI) resulted in lower (P<0.05) NH<sub>3</sub>-N (CP equivalent) and pH compared with the silages that were not inoculated before the first ensiling (N/N and N/HBI). Inoculation at first ensiling (HBI/N and HBI/HBI) tended (P<0.06) to reduce the content of ethanol compared with non-inoculation prior to the first fermentation. Re-inoculation with homolactic bacteria did not affect (P>0.05) the VFA contents of triticale silages that were re-ensiled. This lack of effect was probably due to the very low content of starch and the absence of WSC in the silages to be re-ensiled (Table 1), which would be expected to inhibit the development of the lactic acid bacteria provided by re-inoculation.

				Treat	ment				
ITEM	N/N		N/H	IBI	HB	HBI/N		HBI/HBI	
g∙kg⁻¹	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
DM <sup>2</sup>	313.8	15.6	297.7	5.9	313.0	2.3	306.4	4.4	0.08
СР	150.8 <sup>B</sup>	4.7	152.7 <sup>в</sup>	2.8	161.5 <sup>A</sup>	3.2	161.3 <sup>A</sup>	4.3	0.003
ADICP	6.7 <sup>AB</sup>	0.4	$7.0^{A}$	0.2	6.1 <sup>B</sup>	0.1	6.6 <sup>AB</sup>	0.5	0.04
ADF	424.7 <sup>A</sup>	7.2	421.9 <sup>A</sup>	4.3	409.0 <sup>B</sup>	4.1	406.4 <sup>B</sup>	4.3	0.001
aNDF	583.4 <sup>A</sup>	5.3	578.8 <sup>A</sup>	3.1	569.5 <sup>B</sup>	3.9	565.8 <sup>B</sup>	2.7	0.001
Lignin	38.5 <sup>A</sup>	1.2	39.1 <sup>A</sup>	2.2	33.0 <sup>B</sup>	2.2	35.9 <sup>AB</sup>	2.9	0.008
Starch	2.7	0.1	2.9	0.4	2.8	0.3	2.9	0.5	0.79
Sugar	2.7	0.1	2.6	0.1	2.6	0.1	2.7	0.1	0.86
NFC	61.6	8.8	67.5	7.1	70.6	7.9	77.2	5.5	0.07
Ash	174.5	7.8	171.4	3.1	167.2	9.6	165.1	5.8	0.28
10	ABC		11.00 D 0.0	-		2	11	1 D)	<b>(</b> 1 )

 Table 1
 Effect of inoculation or not with homolactic bacteria on nutritional parameters of re-ensiled triticale silage<sup>1</sup>

<sup>1</sup>Superscripts <sup>A,B,C</sup> within the same row differ P<0.05

<sup>2</sup>Fresh matter; all other DM basis

Table 2	Effe	ect of inocul	lation or no	t with ho	molactic	bacteria on t	he fe	ermentation of	f re-ensi	led tritical	e silage <sup>1</sup>	•

		Treatment									
ITEM	N/N		N/HBI		HB	HBI/N		HBI/HBI			
	Mean	SD	Mean	SD	Mean	SD	Mean	SD			
рН	5.18 <sup>A</sup>	0.03	5.23 <sup>A</sup>	0.04	4.96 <sup>B</sup>	0.04	4.94 <sup>B</sup>	0.04	0.01		
Lactic Acid <sup>2</sup>	18.8	0.7	16.7	1.6	24.3	14.6	27.2	3.7	0.23		
Acetic Acid <sup>2</sup>	53.7	5.6	57.0	3.21	41.3	24.3	53.0	3.5	0.36		
Propionic	3.2	1.0	2.7	0.4	2.0	1.3	2.4	0.5	0.37		
Acid <sup>2</sup>											
Butyric	ND		ND		ND		ND		-		
Acid <sup>2,3</sup>											
Ethanol <sup>2</sup>	9.9	2.2	10.0	1.6	5.8	3.5	6.9	1.2	0.05		
Total VFA <sup>2</sup>	75.6	7.1	76.3	4.4	67.6	40.2	82.6	7.5	0.79		
Ammonia, <sup>4</sup>	130.7 <sup>AB</sup>	13.1	136.6 <sup>A</sup>	9.4	110.9 <sup>C</sup>	7.8	106.3 <sup>BC</sup>	10.1	0.01		

<sup>1</sup>Superscripts <sup>A,B,C</sup> within the same row differ P<0.05

<sup>3</sup> g·kg<sup>-1</sup> DM basis

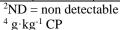
## Conclusions

Re-inoculation with HBI of simulated translocated triticale silage resulted in no important improvements in the nutritional and fermentative characteristics probably due to the lack of starch or water soluble sugars in the re-ensiled silage. The use of HBI at first ensiling improved CP and fiber levels. The results of this study emphasize the benefits of inoculation of the silage at first ensiling, which extended to the re-ensiling phase.

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# Effectiveness of Various Silage Additives Used in Ensilage of Sorghum Grain and Forage Hybrid

BACK

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#### Introduction

Sorghum is the one of the most important cereals on the world. This plant is use in food, feed and energy industry. Sorghum is resistant on lack of water what gives it possibility to grow on terrains with subtropical climate. It grown in India, China, African countries (Sudan, Nigeria, Egypt, RSA and Burkina Faso), USA and Mexico. In last years in European countries and also in Poland global warming is observed. That causes periodic draughts which decrease level of ground water. This is a threat for many crops, especially maize, that's why farmers has been looking for alternative plants which can get yield with satisfactory quality. Such alternative could be sorghum. The purpose of experiment were evaluation of chemical composition of grain sorghum variety (code NK251) ensiled with various silage additives and effect of used those additives on quality of obtained silages.

#### **Material and Methods**

As a material in experiment were used grain sorghum variety – code NK251 (during the harvest – brown grains; maturity: hard dough to fully ripe), grown in year 2015.

Raw material was ensiled in microsilos (about 1,5 kg green forage), silage additives were used in following scheme: 0 - control group, without additives (WA), 1 - bacterial additive (A1), which contained following bacteria: *Lactobacillus plantarum KKP/788/p*, *Lactobacillus buchneri KKP/593/p* and *Lactobacillus buchneri KKP/907/p*, 2 additive contained bacteria and enzymes (A2), preparation contained A1 and feed enzymes: endo-1,4-beta-glucanase and endo-xylanase, 3 - chemical and bacterial additives (A3) – additive D1 and formic acid add in amount of 3 ml/kg of raw material, 4 - contained 3 listed additives: bacterial, chemical and enzymatic(A4). Each experimental group contained 3 replications

The basic chemical composition was determined by conventional methods (AOAC 2007), structural carbohydrates, ADF and NDF - by the method of Georing and Van Soest (1970), the contents of lactic acid, acetic acid and butyric - Lepper method (AOAC 2007).

Test of aerobic stability of silage was also performed. From each tubes collected 200 g of silage for testing thermal stability under aeration. The samples were exposed to air for a period of 5 days (120 hours). The test was conducted by means of electronic, automatic thermometer LB-711. The temperature inside each silage samples was measured and recorded every hour.

All obtained data were evaluate by one factorial ANOVA, differences between groups were determined by Tukey test.

## **Results and Discussion**

In experiment no significant differences ( $p \le 0.05$ ) were observed in case content of dry matter, crude ash, crude fat and crude fiber in experimental groups. The highest content of CP ( $p \le 0.05$ ) was determined when in silage was used A3 (68.36 g\kg DM), while the lowest was obtained when silage was ensiled without additive and A1 (respectively 60.90 and 60.11 g\kg DM). In case of N-free extract significant higher ( $p \le 0.05$ ) content was determined when in silage used A2 (538.78 g\kg DM) in comparison with other groups. The lowest ( $p \le 0.05$ ) content of NDF was observed when in silage used A4 (453.21 g\kg DM) in comparison to other groups.

Application A3 and A4 in raw material decrease ( $p \le 0.01$ ) pH in silage below 4.20. In silages with A3 and A4 didn't butyric acid ( $p \le 0.01$ ) and registered significant higher content of acetic acid (respectively 30.33 and 39.26 g/kg DM). Addition of additives decrease content of lactic acid in silage, the lowest value ( $p \le 0.01$ ) as observed when A2 were used (25.39 g/kg DM). All silages were aerobic stable during the 5-day period of aeration.

#### Conclusions

In summary, the most effective additives to prepare silages from sorghum are A3 and A4 contained mainly chemical and bacterial substances.

Item	Dry matter, g/kg	Crude ash, g/kg DM	Crude protein, g/kg DM	Crude fiber, g/kg DM	Crude fat, g/kg DM	Nitrogen free extract, g/kg DM	NDF, g/kg DM	ADF, g/kg DM
WA	346.10 ± 9.50	42.15 ± 1.73	60.9 <sup>a</sup> ± 4.27	264.18 ± 21.17	31.90 ± 1.07	$600.84^{a} \pm 17.59$	$604.96^{a} \pm 32.78$	342.67 ± 15.76
A1	338.50 ± 17.03	47.03 ± 5.02	60.11 <sup>a</sup> ± 8.24	$266.75 \pm 41.60$	30.86 ± 3.43	$595.25^{a}$ ± 46.24	581.8 <sup>a</sup> ± 27.11	328.34 ± 61.33
A2	327.33 ± 15.04	49.51 ± 4.17	$63.72^{ab} \pm 5.52$	324.47 ± 24.18	23.51 ± 7.27	$538.78^{b} \pm 29.74$	628.95 <sup>a</sup> ± 31.98	367.55 ± 22.54
A3	334.03 ± 7.45	46.45 ± 3.66	68.37 <sup>b</sup> ± 2.01	$\begin{array}{c} 252.22\\ \pm 4.25\end{array}$	22.67 ± 2.67	$610.29^{a} \pm 9.50$	$\begin{array}{c} 588.80^{\mathrm{a}} \\ \pm 8.96 \end{array}$	$329.46 \pm 14.92$
A4	344.47 ± 21.88	41.62 ± 5.43	$64.19^{ab} \pm 1.67$	252.49 ± 34.09	23.41 ± 3.37	$\begin{array}{c} 618.28^a \\ \pm 41.23 \end{array}$	453.21 <sup>b</sup> ± 30.28	301.14 ± 17.44

 Table 1
 Content of basic nutrients in analyzed silages

Values in columns differ significant A,B when  $p \le 0.01$ ; a,b – when  $p \le 0.05$ 

 Table 2
 Content of silage acids, ammonia nitrogen and pH value of silages

Item	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Butyric acid, g/kg DM	N – NH3, % N <sub>total</sub>	рН
WA	$72.11^{Aa} \pm 64.32$	12.80 <sup>a</sup> ± 6.64	28.03 <sup>A</sup> ± 12.70	4.37 ± 1.37	$4.69^{\rm A} \pm 0.05$
A1	34.82 <sup>ABb</sup> ± 13.67	$16.97^{a} \pm 14.20$	$20.43^{\rm A} \pm 12.08$	$\begin{array}{c} 4.31 \\ \pm \ 0.81 \end{array}$	$\begin{array}{c} 4.71^{\mathrm{A}} \\ \pm \ 0.19 \end{array}$
A2	25.39 <sup>B</sup> ± 14.49	$24.63^{ab} \pm 10.86$	21.14 <sup>A</sup> ± 3.51	$5.04 \\ \pm 0.62$	$\begin{array}{c} 4.87^{\mathrm{A}} \\ \pm \ 0.07 \end{array}$
A3	$\begin{array}{c} 66.55^{\mathrm{Aa}} \\ \pm 4.62 \end{array}$	30.33 <sup>b</sup> ± 3.64	$\begin{array}{c} 0.00^{\mathrm{B}} \\ \pm \ 0.00 \end{array}$	3.74 ± 1.29	$\begin{array}{c} 4.17^{\mathrm{B}} \\ \pm \ 0.04 \end{array}$
A4	43.92 <sup>ABb</sup> ± 5.13	39.26 <sup>b</sup> ± 10.31	$\begin{array}{c} 0.00^{\mathrm{B}} \\ \pm \ 0.00 \end{array}$	$\begin{array}{c} 4.17 \\ \pm \ 0.05 \end{array}$	$\begin{array}{c} 4.20^{\mathrm{B}} \\ \pm \ 0.04 \end{array}$

Values in columns differ significant A,B when  $p \le 0.01$ ; a,b – when  $p \le 0.05$ 

Key words: sorgo, silage, additive, chemical composition, quality, aerobic stability

# Changes in Chemical Composition and Quality Parameters of Sorghum Grain Variety (12GS9014) Whole Plants Ensiled with Application of Various Silage Additives

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## Introduction

Sorghum is on fifth place among most important cereal in the world. This plant is applicable for example in the food, energy and feed industry. It is a plant resistant to drought, that's why it is widely cultivated in subtropical climates of India, China, African countries (Sudan, Nigeria, Egypt, South Africa, Burkina Faso), the US and Mexico, where water availability is a limiting factor in crop production. Recently, in European countries, including Poland a gradual warming of the climate and the occurrence of periodic droughts was observed, leading to a reduction in the groundwater. This is a threat to many corn crops, therefore researcher are looking for an alternative plants, which will enable in difficult conditions to get a high yield of satisfactory quality. The alternative could be sorghum. The aim of the study was to evaluate the chemical composition of varieties of grain sorghum code NK251 ensilage with using different additives and the effect of this additives on the quality of the obtained silage.

## **Material and Methods**

The material was a variety of sorghum code NK251 - grain sorghum (grains brown during harvesting, maturity wax to full), grown in 2015. Ensiled forage laboratory scale in mikrosilos (approx. 1.5 kg of green fodder) using silage additives in the following manner: 0 - control group, without additives (WA), 1 - bacterial additive (A1), which included bacterial strains: Lactobacillus plantarum KKP / 788 / p, Lactobacillus buchneri KKP / 593 / p and Lactobacillus buchneri KKP / 907 / p 2 - bacterial additive - enzyme (A2), comprising the addition of A1 and also enzymes: endo-1,4-beta- glucanase and endo-xylanase, 3 - addition of bacterial and chemical (A3) - appendix A1 and additional formic acid is added in an amount of 3 ml / kg of fodder additive and 4 (A4) was a combination of the three additives: bacterial, enzymatic and chemical. Each test was performed in triplicate. The basic chemical composition was determined by conventional methods (AOAC 2007), structural carbohydrates, ADF and NDF - by the method of Georing and Van Soest (1970), the contents of lactic acid, acetic acid and butyric - Lepper method (AOAC 2007).

We also performed a test aerobic stability of silage. From each tubes collected 200 g of silage for testing thermal stability under aeration. The samples were exposed to air for a period of 5 days (120 hours). The test was conducted by means of electronic, automatic thermometer LB-711. The temperature inside each silage samples was measured and recorded every hour.

## **Results and Discussion**

In experiment no significant differences ( $p \le 0.05$ ) were observed in case content of dry matter and crude ash between experimental groups. The lowest content of CP ( $p \le 0.05$ ) was determined in silage without additive (67.20 g\kg DM) in comparison to A1, A2 and A4 (respectively 74.36, 73.23 and 72.00 g\kg DM). Application in ensiled material A4 decreased ( $p \le 0.01$ ) content of crude fiber (228.38 g/kg DM) in comparison with other groups. Silage additives used in ensilage process decreased content of crude fat in silage, significant difference were determined between control group and silage with A4. In case of N-free extract significant higher ( $p \le 0.05$ ) content was determined when in silage used A4 (619.08 g\kg DM) in comparison with silage with A2 (573.17 g\kg DM). The lowest ( $p \le 0.01$ ) content of NDF was observed when in silage used A1 (446.82 g\kg DM) in comparison to control group, A3 and A4 (respectively 498.28, 503.40 and 487.71 g/kg DM).

Application A1 and A2 in raw material decrease ( $p \le 0.05$ ) pH in silage below 4.06. In silages with additives butyric acid wasn't observed ( $p \le 0.05$ ) also in those groups was determined higher concentration of acetic acid (significant difference were registered between control group - 21.33 g/kg DM and A4 – 33.15 g/kg DM). The highest concentration of lactic acid ( $p \le 0.05$ ) was observed in silage with A1 (103.80 g/kg DM) in comparison to A3 and A4 (respectively 91.22 and 84.58 g/kg DM). All silages were aerobic stable during the 5-day period of aeration.

## Conclusions

In summary, the most effective additives to prepare silages from sorghum are A1 and A4. First contained various strains of bacteria, second contained chemical, bacterial and enzymatic substances.

Item	SM, g/kg	PS, g/kg DM	BS, g/kg DM	WS, g/kg DM	TS, g/kg DM	BAW, g/kg DM	NDF, g/kg DM	ADF, g/kg DM
WA	$376.70 \pm 17.80$	$\begin{array}{c} 61.70 \\ \pm 2.91 \end{array}$	67.20 <sup>a</sup> ± 1.29	258.97ª ± 7.60	33.49 <sup>a</sup> ± 2.37	$578.65^{ab} \pm 10.18$	$\begin{array}{c} 498.28^{\rm Aa} \\ \pm \ 34.04 \end{array}$	$280.35^{ab} \pm 36.72$
A1	363.5 ± 16.74	$\begin{array}{c} 62.63 \\ \pm \ 6.36 \end{array}$	$74.36^{b} \pm 2.88$	$247.40^{ab} \pm 6.42$	$\begin{array}{c} 26.41^{ab} \\ \pm \ 4.85 \end{array}$	588.19ab ± 1.25	$446.82^{\text{Bb}} \pm 27.83$	265.81 <sup>b</sup> ± 21.12
A2	$365.77 \pm 17.20$	62.74 ± 1.47	73.32 <sup>b</sup> ± 4.51	$264.91^{a} \pm 29.37$	$\begin{array}{c} 25.88^{ab} \\ \pm 5.75 \end{array}$	573.17a ± 36.19	$462.67^{ABb} \pm 38.23$	$260.00^{b} \pm 24.84$
A3	368.27 ± 18.46	62.06 ± 7.53	$71.13^{a.b} \pm 5.92$	250.91ª ± 66.06	$\begin{array}{c} 26.16^{ab} \\ \pm 4.45 \end{array}$	589.73ab ± 81.27	$\begin{array}{l} 503.4^{\mathrm{Aa}} \\ \pm 90.82 \end{array}$	292.19ª ± 55.29
A4	381.97 ± 12.91	59.77 ± 5.58	72.00 <sup>b</sup> ± 2.36	228.38 <sup>b</sup> ± 15.52	$20.78^{b} \pm 3.96$	619.08b ± 19.49	$\begin{array}{c} 487.71^{\rm Aa} \\ \pm \ 10.07 \end{array}$	$266.77^{ab} \pm 14,68$

 Table 1
 The content of the basic nutrients analyzed in silages

 $\overline{A,B}$  – values in the strongly differ significantly (P $\leq$ 0,01) a,b – values in the significantly different (p  $\leq$  0.05)

 Table 2
 The content of silage acids, ammonia nitrogen and pH of sorghum silages

Item	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Butyric acid, g/kg DM	N – NH3, % N <sub>total</sub>	рН
WA	95.35 <sup>ab</sup> ± 5.20	21.33 <sup>a</sup> ± 6.42	5.20a ± 5.65	$5.91 \\ \pm 0.73$	$\begin{array}{c} 4.20^{\mathrm{ab}} \\ \pm \ 0.09 \end{array}$
A1	103.80 <sup>a</sup> ± 4.79	$\begin{array}{c} 28.70^{ab} \\ \pm 3.83 \end{array}$	$\begin{array}{c} 0.00b\\ \pm \ 0.00\end{array}$	$5.80 \\ \pm 0.60$	$\begin{array}{c} 4.04^{b} \\ \pm \ 0.02 \end{array}$
A2	$92.80^{ab} \pm 7.77$	$\begin{array}{c} 27.86^{ab} \\ \pm 8.59 \end{array}$	$\begin{array}{c} 0.00b\\ \pm \ 0.00\end{array}$	$5.52 \\ \pm 0.61$	$\begin{array}{c} 4.06^{b} \\ \pm \ 0.02 \end{array}$
A3	91.22 <sup>b</sup> ± 6.27	$27.91^{ab} \pm 3.47$	$\begin{array}{c} 0.00b\\ \pm \ 0.00\end{array}$	$5.56 \\ \pm 0.76$	$\begin{array}{c} 4.10^{ab} \\ \pm \ 0.02 \end{array}$
A4	84.58 <sup>b</sup> ± 11.02	33.15 <sup>a</sup> ± 13.39	$\begin{array}{c} 0.00b\\ \pm \ 0.00\end{array}$	4.94 ± 1.57	$\begin{array}{c} 4.20^{\mathrm{a}} \\ \pm \ 0.04 \end{array}$

a,b – values in the significantly different ( $p \le 0.05$ )

Key words: sorgo, silage, additive, chemical composition, quality, aerobic stability

# Effects of Seven Formic Acid Based Additives on Grass Silage Fermentation and Aerobic Stability

BACK

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## Introduction

Formic acid (FA) as silage additive has several benefits. Rapid drop of pH in the beginning of ensiling reduces proteolysis and growth of harmful bacteria (McDonald et al 1991). Lactic acid fermentation is restricted which in turn has several benefits such as higher silage intake (Huhtanen et al. 2007), higher production of microbial protein in rumen (Jaakkola et al. 2006) and higher milk protein and fat content (Huhtanen et al. 2003). FA treated silages have also had better aerobic stability than silages made with homofermentative lactic acid bacteria strains (Saarisalo et al. 2006, Seppälä et al. 2013, Seppälä et al. 2016). Further, in automatic milking systems, silage fermentation quality seems to be the most important feed parameter to explain differences in milking frequency (Puumala et al. 2014). Blends of organic acids and their salts are used as silage additive to combine beneficial effects of different acids and some buffering (sodium or ammonium salts) is typically used to reduce corrosivity and odour of the product. In this trial seven commercial FA-based blends were compared to evaluate differences between them.

#### **Materials and Methods**

A third cut of timothy-meadow fescue (proportions 65 and 32 % of fresh matter) grass was prewilted overnight, precision chopped and ensiled in 12 l silos to compare efficacy of silage additives in Jokioinen, Finland. The tested silage additives included seven blends of organic acids (Table 1) and their salts, the major component being FA. Application level was 5 l/t. A control treatment without additive was included. Four replicate silos were filled for each treatment to measure fermentation quality and aerobic stability. Ensiling time was three months (10 Sep-10 Dec 2015). Analytical methods were the same as described by Seppälä et al. (2016).

Table 1	Blends and products tested in the trial (provided by Eastman Chemical Company). Additionally a control
treatment	without additive was included.

Name	Active ingredients
AIV 2 Plus	formic acid, ammonium formate
Blend1	formic acid, sodium formate, propionic acid, sodium bentzoate
Blend2	formic acid, sodium formate, propionic acid
AIV 2 Plus Na	formic acid, sodium formate
AIV Ässä Na	formic acid, sodium formate, propionic acid, potassium sorbate
Blend3	formic acid, sodium formate, lactic acid
Blend4	formic acid, sodium formate

#### Results

The grass dry matter prior to ensiling was 242 g/kg, and it contained ash 102, crude protein 182, water soluble carbohydrates (WSC) 99, neutral detergent fiber 501 g/kg DM, and buffering capacity was 5.8 g lactic acid/100 g DM. Yeast and mould counts were high (6.03 and 6.41 log cfu/g) but count of lactic acid bacteria (LAB) was only 4.38 log cfu/g.

Tested additives reduced ammonium N (control vs additive treated silages: 66 vs 37 g/kg total N), lactic acid (99 vs 45 g/kg dry matter (DM)) and acetic acid concentrations (25 vs 13 g/kg DM) in the silage, which was reflected as improved silage DM intake index (99 vs. 91, Huhtanen et al. 2007) and higher WSC concentration (87 vs. 22 g/kg DM) in the additive treated silages. The strongest restrictive effect on fermentation was obtained when products AIV 2 Plus Na and AIV Ässä Na were used, while the latter also resulted in low ethanol content, lowest yeast counts and longest aerobic stability of the silage. In general, additive treatments improved aerobic stability of the silages (175 vs. 104 hours).

### Discussions

Grass material had fermentation coefficient of 38 which means that the grass was moderately ensilable (Weissbach and Honig 1996). Despite low number of epiphytic LAB, the control silage showed fermentation which was dominated by lactic acid. However intake potential of the control silage was inferior compared to additive treated silages due to high amounts of fermentation acids.

Comparing AIV 2 Plus and AIV 2 Plus Na which differ only in the type of buffering (ammonium formate vs. sodium formate) shows that the silages made with those additives differ from each other only in terms of ammonium content, which reflects difference in their constituents. Inclusion of propionic acid in AIV Ässä Na shows that in addition to the ability to restrict lactic acid fermentation that additive has ability to restrict also ethanol production,

which was reflected as the lowest yeast count. WSC content in AIV Ässä Na treated silage was slightly higher than in grass prior to ensiling probably as a consequence of degradation of cell wall carbohydrates in acidic environment (McDonald et al 1991).

 Table 2
 Fermentation and microbial quality and aerobic stability of silages treated with different formic acid based additives.

	Control	AIV2 Plus	Blend1	Blend2	AIV 2 Plus Na	AIV Ässä Na	Blend3	Blend4	SEM	P-value
Dry matter (DM), g/kg	240	241	241	243	239	240	241	242	1.1	0.1567
рН	4.22	4.27	4.20	4.20	4.21	4.22	4.17	4.34	0.023	0.0007
Ammonium-N, g/kg total N	66.1	43.1	41.1	38.7	30.8	36.9	34.5	36.9	1.40	<.0001
WSC, g/kg DM	22.3	85.5	93.8	73.9	86.6	115.1	75.6	77.1	5.44	<.0001
Ethanol, g/kg DM	2.7	19.4	11.4	17.1	21.0	7.2	23.7	18.1	2.45	<.0001
Lactic acid, g/kg DM	98.8	38.2	57.1	54.7	32.2	42.1	48.4	41.5	2.18	<.0001
Acetic acid, g/kg DM	25.4	12.1	12.4	13.4	11.9	11.6	12.7	13.8	0.68	<.0001
Propionic acid, g/kg DM	0.46	1.16	2.72	1.71	0.45	5.17	0.44	0.45	0.108	<.0001
Butyric acid, g/kg DM	0.36	0.49	0.34	0.33	0.41	0.37	0.29	0.33	0.039	0.0482
Silage DM Intake index	91	100	97	98	101	99	99	99	0.3	<.0001
Formic acid, g/kg DM	0	16.6	14.4	15.0	16.7	13.4	15.4	19.0	0.52	<.0001
Yeasts (log <sub>10</sub> cfu/g)	4.0	4.6	4.0	4.4	4.7	3.4	4.6	4.4	0.22	0.0055
Moulds (log <sub>10</sub> cfu/g)	2.1	0.9	2.1	1.8	2.8	2.0	2.8	0.6	0.61	0.143
Aerobic stability, hours	104	181	183	192	140	197	166	169	15.2	0.0013

WSC=water soluble carbohydrates, VFA = volatile fatty acids

## Conclusions

Tested additives restricted fermentation, improved silage intake potential and prolonged aerobic stability. Changing type of buffering (sodium formate vs. ammonium formate) had no other effect on silage quality than the change in ammonium content due to a difference in the composition of the additive.

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# Effects of Different Sodium Nitrite-Containing Additives on Dry Matter Losses, Fermentation Pattern and Biogenic Amine Formation in Lucerne and Cocksfoot Silage

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BACK

#### Introduction

Sodium nitrite (SN) has been widely used in Europe as a component of silage additives that strongly inhibits clostridia. As SN is rapidly converted to nitric oxides and ammonia during the early fermentation phase, this compound is applied in combination with other chemicals, which prevent clostridia development during later stages of the fermentation process. Those substances can be hexamethylene tetramine (HMT), sodium benzoate and potassium sorbate (Bader, 1997). The aim of this study was to test as to whether the most important co-substance of SN in chemical silage additives - HMT - can be replaced by a combination of formate with either sodium benzoate or potassium sorbate in difficult-to ensile materials, such as low dry matter (DM) lucerne and cocksfoot, without compromising the inhibiting effects on clostridia even for extended periods of anaerobic storage.

#### **Materials and Methods**

Forages were cultivated on the research farm of University Nürtingen-Geisslingen, Germany, mowed by a plot harvester before budding (lucerne, Medicago sativa L.) and before heading (cocksfoot, Dactylis glomerata L.) respectively, in early June 2015. After raking by hand in the field, forages were wilted under a roof for about one day and subsequently chopped by a stationary chopper to about 30 mm theoretical cutting length. After additive application, three replicates of 1.5 L glass jars per treatment were filled and stored at 20 °C for 314 days (lucerne) and 313 days (cocksfoot), respectively. In addition to a control treatment (CON) and the commercially available product Xtrasil classic (XC, 245 g/kg sodium nitrite, 165 g/kg hexamethylene tetramine), two proprietary new blends composed of sodium nitrite and formate/sorbate (XFS) or formate/benzoate (XFB) were tested. All products were obtained from KONSIL Europe GmbH and applied at 3 l/t. All additives were diluted with tap water to give an application rate of 10 ml/kg fresh forage which was manually sprayed onto the forage. The control treatment received 10 ml of tap water per kg fresh forage. Chemical analysis of the fresh herbage was performed according to the official German standards for feed evaluation. The DM of silages was measured and corrected for the loss of volatiles during drying (Weissbach and Kuhla, 1995). An HPLC method was used to analyze lactic acid, and volatile fatty acids and alcohols were determined by GC. Biogenic amines were detected by GC-MS/MS. Losses of DM during fermentation were calculated according to Weissbach (2005). Statistical analyses were performed by using the procedures GLM and REG of SAS. Differences between least-square (LS) means were tested by Tukey's test, and significance declared at P < 0.05.

#### **Results and Discussion**

Both forages had typical protein and fibre values for first cut materials (Table 1). However, ash content exceeded the recommended maximum content of 100 g/kg DM, and water-soluble carbohydrate concentrations were much lower than previously reported by Weissbach and Auerbach (2012). Especially the markedly increased buffering capacity, which was most likely caused by the high ash content due to raking by hand, rendered the forages difficult-to-ensile as the fermentability coefficients were below 35 (Weissbach and Auerbach, 2012).

Table I Cha	<b>Table 1</b> Characteristics of the resh forages at ensning (n=5, means and standard deviation in brackets)									
Forage	$DM^1$	$CP^2$	$CF^3$	$CA^4$	WSC <sup>5</sup>	$BC^{6}$	FC <sup>7</sup>			
species	(g/kg)	(g/kg DM)	(g/kg DM)	(g/kg DM)	(g/kg DM)	(g LA/kg DM)				
Lucerne	231 (0.4)	244 (5.3)	251 (8.9)	182 (6.6)	42 (0.9)	102 (3.5)	26 (0.6)			
Cocksfoot	200 (3.0)	187 (9.6)	274 (7.1)	132 (2.3)	40 (13.6)	99 (2.7)	23 (0.9)			

**Table 1** Characteristics of the fresh forages at ensiling (n=3, means and standard deviation in brackets)

<sup>1</sup>dry matter; <sup>2</sup>crude protein; <sup>3</sup>crude fibre; <sup>4</sup>crude ash; <sup>5</sup>water-soluble carbohydrates; <sup>6</sup>buffering capacity; <sup>7</sup>fementability coefficient=DM+8\*WSC/BC

Fermentation losses were reduced by silage additives in both forage species, and differences between additive treatments were not observed (Table 2). This is in line with observations on other sodium nitrite-containing additives (Reuter and Weissbach, 1991; Knicky and Spörndly, 2010). Untreated silages underwent excessive clostridial fermentations as demonstrated by high concentrations of butyric acid due to anaerobic decomposition of lactic acid, as well as by extremely high ammonia levels (Kaiser et al., 1997). The activity of clostridia was largely restricted by the additives with butyric acid concentrations never exceeding 3 g/kg DM, which is the threshold value for good silage quality set by the German Agricultural Society (DLG, 2006). The formation of acetic acid remained unaffected by treatment in lucerne silage but the additives increased its concentration in cocksfoot silage. Ethanol formation in lucerne silage was suppressed by all treatments, whereas the additive XC increased its concentration when compared with untreated cocksfoot silage. Decarboxylation of amino acids to biogenic amines by clostridia was strongly reduced by

additive use, and the resulting values were always lower than typical concentrations of less than 5 g/kg DM in well-fermented silages (Richardt, 2012).

Treatment	$DML^1$	pН	$NH_3-N^2$	Lactate	Acetate	Butyrate <sup>3</sup>	Ethanol	Amines <sup>4</sup>
	(%)		(% total-N)	(g/kg DM)	(g/kg DM)	(g/kg DM)	(g/kg DM)	(g/kg DM)
				Lucerne (A	Aedicago sativ	a L.)		
CON	12.3ª	6.23 <sup>a</sup>	36.0 <sup>a</sup>	1.6 <sup>b</sup>	34.0	93.0ª	11.0 <sup>a</sup>	6.4 <sup>a</sup>
XC	6.6 <sup>b</sup>	4.88 <sup>b</sup>	12.6 <sup>b</sup>	74.1 <sup>a</sup>	39.0	2.0 <sup>b</sup>	5.7 <sup>b</sup>	2.4 <sup>b</sup>
XFS	6.6 <sup>b</sup>	4.84 <sup>b</sup>	13.1 <sup>b</sup>	74.6 <sup>a</sup>	35.5	$0.6^{b}$	5.9 <sup>b</sup>	2.4 <sup>b</sup>
XFB	6.6 <sup>b</sup>	4.85 <sup>b</sup>	13.5 <sup>b</sup>	71.8 <sup>a</sup>	35.7	0.2 <sup>b</sup>	5.4 <sup>b</sup>	2.4 <sup>b</sup>
SEM	0.12	0.047	1.58	2.18	1.70	3.49	0.52	0.39
Significance	***	***	***	***	ns	***	***	***
-				Cocksfoot (L	Dactylis glome	rata L.)		
CON	11.1 <sup>a</sup>	5.85 <sup>a</sup>	23.0 <sup>a</sup>	2.1 <sup>b</sup>	18.1 <sup>b</sup>	54.4 <sup>a</sup>	10.2 <sup>b</sup>	14.1 <sup>a</sup>
XC	7.5 <sup>b</sup>	5.00 <sup>b</sup>	11.9 <sup>c</sup>	39.7 <sup>a</sup>	33.2 <sup>a</sup>	0.1 <sup>b</sup>	12.6 <sup>a</sup>	2.3 <sup>b</sup>
XFS	7.5 <sup>b</sup>	4.94 <sup>b</sup>	14.2 <sup>b</sup>	37.7 <sup>a</sup>	36.6 <sup>a</sup>	0.5 <sup>b</sup>	11.1 <sup>ab</sup>	2.0 <sup>b</sup>
XFB	7.5 <sup>b</sup>	4.90 <sup>b</sup>	14.8 <sup>b</sup>	38.9ª	36.7ª	$0.0^{b}$	11.2 <sup>ab</sup>	2.6 <sup>b</sup>
SEM	0.03	0.097	0.19	0.84	1.15	1.72	0.37	0.50
Significance	***	***	***	***	***	***	*	***

Table 2	Effects of sodium nitrite-containing additives on DM losses, fermentation pattern, aerobic stability and
biogenic	amine concentrations in lucerne and cocksfoot silage (n=3; data presented as LSmeans)

<sup>1</sup>dry matter losses; <sup>2</sup>corrected for the addition of N by the additives;, <sup>3</sup>sum of n- and iso-butyric acid, n- and i-valeric acid and n-caproic acid; <sup>4</sup>total biogenic amines reflecting the sum of putrescin, cadaverin, histamine, phenylethylamine and tyramine concentrations; values in columns within forage species bearing different superscripts differ (P < 0.05, Tukey's test); \*P < 0.05, \*\*P < 0.01, \*\*\*P < 0.001, ns not significant.

Across forage species, there existed a strong polynomial relationship between the concentrations of butyric acid and ammonia-N (y=0.16x<sup>2</sup>+0.91x+13.3, R<sup>2</sup>=0.97, P<0.001). Moreover, biogenic amine concentrations were closely related with those of butyric acid (lucerne: R<sup>2</sup>=0.91, P<0.001; cocksfoot: R<sup>2</sup>=0.99, P<0.001) and ammonia-N (lucerne: R<sup>2</sup>=0.86, P<0.001; cocksfoot: R<sup>2</sup>=0.90, P<0.001) respectively, but the slopes and intercepts for the linear models differed between lucerne and cocksfoot silage.

#### Conclusions

Fermentation quality of lucerne and cocksfoot silages ensiled at low DM was very poor and elevated concentrations of biogenic amines were detected in untreated silages. The use of sodium nitrite-containing additives – regardless of the co-substances in the mixture - resulted in excellent fermentation quality and also reduced biogenic amine formation to normal levels even after extended periods of storage for about ten months. It seems possible to replace HMT by combinations of formate/sorbate or formate/benzoate in silage additive preparations without compromising their inhibitory effect on clostridia growth. More studies, including large scale ensiling trials, are under way.

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# Improving of Aerobic Stability of Maize Silage by Different Additives

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## Introduction

The hygienic quality of maize silage is important for animal health, animal production and food quality (Knicky M. 2009). On quality of maize silage was influenced by different factors. Especially for maize silage is difficult to improve aerobic stability. When the silo is opened for feeding and the silo face is poorly managed, yeasts, molds develop and cause heating. Meanwhile bacterial and fungal deterioration occurs with the risk of mycotoxin production. Organic acids have been used extensively as forage and grain preservatives (Kung at al., 2003). Nowadays, heterofermentative lactic acid bacteria like Lactobacillus buchneri (LB) are very popular since they have proven their efficacy to increase the stability of silage (Holzer at al., 2003). In the last decade, the use of silage inoculants containing LB strains increased steeply, targeting high quality silages with proper aerobic stability during the feedout phase. However, the benefits of LB inoculation seems to be dose-dependent (Kleinschmit and Kung, 2006), at least for silage preservation. Short aerobic stability in silages with higher DM content is associated with the growth of undesirable yeasts and moulds and ultimately a reduction in crop nutrients (Woolford, 2005). The antimicrobial properties of Sodium benzoate, Potassium sorbate and Sodium nitrite are very well known because they are used as additives in the conservation of a variety of feeds and foods. Woolford (1978) characterizes potassium sorbate in forage conservation as being effective in inhibiting spore-forming bacteria, yeasts and molds a pH range from 3 to 6. Sodium benzoate inhibits similar bacteria, although these are reduced at higher pH values. Today, different commercial products with a different composition and mode of action are on the market and farmers need to be informed about their effectiveness. The aim of this experiment was to determinate of aerobic stability for different additives for conservation of maize silage on dairy farms.

## **Materials and Methods**

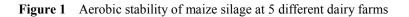
At May 2016 we choose from 5 different dairy farms maize silages. Each farmer used during harvest different additives. Farm 1 Lactic Acid Bacteria (LAB) + Lactobacilus Buchneri (*LB*), Farm 2 Lactic Acid Bacteria (LAB), Farm 3 Lactic Acid Bacteria (LAB) + Lactobacillus buchneri (LB), Farm 4 Lactic Acid Bacteria (LAB) + Sodium benzoate, Farm 5 Lactic Acid Bacteria + Sodium benzoate. Application dose of each additive was according recommendation of producer. The samples (n=3) we removed from the face of silo at the same day and transported immediately to the laboratory. All samples we placed to the apparatus for determination of aerobic stability. Each sample had 3 replicates and determination of temperature was every 15 minute for 7 days resp. 168 hours. Maize silage samples were analysed for fermentation quality (pH, lactic, acetic, propionic and butyric acid) using an IONOSEP 2001 analyser (RECMAN - laboratory systems, Ostrava, Czech Republic) according to Kvasnička (2000). Dry matter (DM) was obtained from drying of silage at 60°C for 24 hours. Dried material was subsequently milled to pass through a one-millimetre sieve for laboratory analyses. NDF was assayed with a heat stable amylase and expressed inclusive of residual ash (aNDF) according to Mertens (2002). DM, ash, crude fibre (CF), and CP (N x 6.25) were determined as described by AOAC (2005). OM was calculated as DM minus ash. Degradability of OM (OMD) and NDF (NDFD) was measured by *in sacco* method (in vitro) in the fistule rumen of dairy cows (McDonald, 1981).

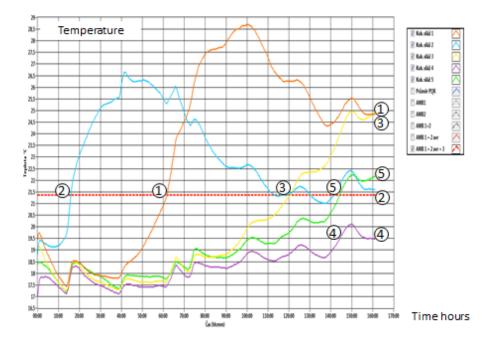
#### **Results and Discussion**

At the Table 1 you can see results of quality of 5 maize silages from different dairy farms. The content of organic nutrients is similar for all maize silages. At that Table is not difference, pH of all silages was from 3.54 to 3.65 within low difference. But content of lactic acid (LA) was lower for silages treated by LAB + L. buchneri, silages treated by LAB + Sodium benzoate had content of LA 3.43 and 3.49 % FM. Content of Acetic acid(AA) was at all silages with low range 0.8 to 0.99 % FM. Content of AA was at all silages under 1 % FM. Some authors declared content of acetic acid is too high it may decrease intake of DM, but we did not tested intake of DM. For us was very important at this experiment to monitoring aerobic stability of maize silages. Aerobic stability, according Figure 1, was very different at different samples. Very low stability we can see in sample 1 and 2 which was treated by LAB and LAB + LB. Maize silage treated by LAB had lowest content of AA (0.8 % FM) but this level of AA indicate some technology problems during daubing of silage on farm. We can see large differences for ratio of LA/VFA, but silage treated by LAB and Sodium benzoate had higher numbers. Silages treated by biological additives had this ratio lower. Results of OMD and NDFD were between samples very different and you can see also different content of NEL. We can see that farmers which used the program for the selection of silage hybrids according NDFD have higher NDFD (farm 1 and 4). Next point for good quality of maize silage is ratio of structure fibre which is determined by Penn State Particle Separator (PSPS). The highest TLC (Theoretical Long of Cat) we determined for maize silage in farm 3 (29 % on 18 mm) and lowest TLC for silage farm 5 (3.21 % on 18 mm).

## Conclusion

According results of aerobic stability we can see that maize silage treated by different additives had different results. The lowest stability was for silages treated by biological additives LAB. Follow is LAB + LB with different results. The longest stability is for chemical + LAB additives. For maize silage, except aerobic stability, is very important to determinate digestibility of NDF, structure of fibre with PSPS and content of fermentation acids.





**Table 1** Characteristics of maize silage quality at different dairy farms.

N = 3	1	2	3	4	5		
Dry matter (DN), %	34.48	30.96	36.44	34.31	33.91		
Soluble DM, % DM	24.11	14.61	20.15	18.28	26.86		
Crude protein, % DM	8.34	9.6	9.75	6.95	8.4		
Crude fibre, % DM	16.19	20.57	19.91	18.29	20.86		
NDF, % DM	38.25	43.63	40.56	41.2	39.61		
Starch, % DM	43.08	25.61	26.54	35.08	28.61		
Ash, % DM	3.35	4.06	4.15	3.21	3.38		
NFE, % DM	68.83	62.63	62.9	68.25	64.04		
NFC, % DM	46.77	39.56	42.24	44.94	45.29		
OM, % DM	96.65	95.95	95.85	96.79	96.63		
NEL, MJ/kg	6.43	6.24	6.24	6.42	6.31		
pH	3.61	3.65	3.54	3.63	3.59		
Lactic acid (LA), % FM	2.33	2.02	2.85	3.43	3.49		
Acetic acid, % FM	0.99	0.8	0.91	0.9	0.87		
Propionic acid, % FM	0.1	0.02	0.04	0.03	0.04		
Voluntary fatty acid (VFA), % FM	1.09	0.82	0.95	0.93	0.91		
LA/VFA	2.1	2.5	3	3.7	3.8		
NH3, % FM	0.02	0.02	0.02	0.02	0.02		
N-NH <sub>3</sub> , %	4.2	3.14	3.51	4.13	3.15		
Formol. titration, % FM	0.08	0.06	0.13	0.09	0.07		
N - NH <sub>2</sub> , %	14.9	10.6	20.3	20.9	12.6		
Proteolysis, %	19.1	13.7	23.8	25.0	15.8		
OMD (in vitro)	81.3	76.66	75.84	80.56	74.48		
NDFD (in vitro)	72.3	67.69	63.37	73.06	53.98		
Aerobic stability, in Hours	52.6	16.3	121	more 168	145		
Penn State Particle Separator (PSPS) in % of 0.5 kg							
19 mm	6.4	10.8	29.0	7.41	3.21		
8 mm	66.8	59.8	49.0	66.13	65.86		
4 mm	20.8	22.6	16.0	20.24	23.9		
Bottom	6.0	6.8	6.0	6.21	7.03		

# Dedication: MZe NAZV QJ1510391

## References

The list of references is available at the authors and/or at the conference secretary.

# Effects of Dual Purpose Inoculants on Fermentation Parameters, Aerobic Stability and Safety of Whole Crop Maize Silage after a Short Time of Storage

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## Summary

This work aimed to determine the effect of SiloSolve inoculants (FC, AS 200 and AS 100) to whole crop maize on weight loss, fermentation, aerobic stability and microbial composition at storage (fermentation) periods of 2, 4, 8, 16 and 32 days. Inoculant treatments increased acidification rate in whole crop maize silage, resulting in a significant pH drop, and significant total acids and lactic acid concentration. This implies that inoculants accelerated fermentation. All inoculant treatments reduced significantly butyric acid, ethanol and ammonia-N formation, and weight loss during fermentation and during exposure to air time. Yeast and mold cont was decreased significantly by application of silage additives in all storage periods as a consequence of restriction of aerobic deterioration/improving aerobic stability.

## Introduction

Microbial additives containing lactic acid bacteria (LAB) are commonly used for silage preservation to achieve a rapid pH drop through organic acid production, and to improve aerobic stability, by inhibiting spoilage moulds and yeasts (Tabacco et al., 2011). The homofermentative lactic acid bacteria (*Lactobacillus plantarum*, *Lactococcus lactis*) rapidly produce lactic acid which helps to preserve forage mass, however, might decrease aerobic stability of silage. A heterofermentative LAB (*L. buchneri*) has been shown to convert lactic to acetic acid with good antifungal properties and thus improve the stability of silages when they are exposed to air (Muck, 2012). Recently, a novel lactic acid bacteria *Lactococcus lactis* O224 DSM11037 was found to be superior in oxygen scavenging and fasting reducing pH (Hindrichsen et al., 2012). Combining heterofermentative LAB with homofermentative LAB allows gaining positive attributes when active silages fermentation and when silages are exposed to air, respectively. The objective of this study was to evaluate the effectiveness of combining *Lactobacillus buchneri* with the new *Lactococcus lactis* for the fermentation parameters and aerobic stability at even short ensiling times.

#### **Materials and Methods**

Silages were prepared from whole crop maize containing 344 g/kg dry matter and 30.2 g/kg water soluble carbohydrate on fresh matter basis, respectively. Forage was treated the following: no additive (Control), SiloSolve FC containing *Lactobacillus buchneri* DSM22501 and *Lactococcus lactis* DSM11037 (FC), SiloSolve AS 200 containing *Lactobacillus buchneri* DSM22501, *Lactobacillus plantarum* DSM16568 *and Enterococcus faecium* DSM 22502 (AS 200) and SiloSolve AS 100 containing *Lactobacillus buchneri* DSM22501(AS 100), Chr, Hansen A/S, Denmark. The target application rate was 150 000 cfu/g forage. The untreated (Control) received the appropriate amount of sterile water. Forage was ensiled in 3 l laboratory silos and each treatment was replicated 5 times. Silages were analyzed on day 2, 4, 8, 16, and 32 of storage at 20 °C. Aerobic stability (AS) of silages was determined from 7 day aerobic challenge period after each fermentation time (day 2, 4, 8, 16, and 32 of storage) and was determined by monitoring the temperature increase in silages stored aerobically in insulated PVC-tubes at 20 °C. Aerobic stability is determined by the amount of time it takes to exceed the ambient temperature with more than 3 °C. Data were statistically analyzed as a randomized complete block by using the GLM procedure of SAS.

#### **Results and discussion**

Compared to control silage dry matter content was higher (P<0.05) in inoculant treated silages beginning day 8 of fermentation and highest dry matter content gave FC and AS 200 silages (Table 1). Inoculants resulted in improved fermentation and better preserved silages with significantly (P<0.05) lower pH and significantly (P<0.05) lower yeast and mold counts. The highest (P<0.05) pH drop at day 2 and at day 4 of storage was observed in FC and AS 200 silages. However, treatment FC gave the lowest pH value at 32 of storage. Treatments FC and AS 100 showed lowest yeast and mold count at day 32 of the storage. When compared with the Control all inoculants treatments reduced (P<0.05) weight loss beginning day 8 of fermentation. On average of the different fermentation periods inoculants reduced (P<0.05) weight losses from 30% to 51%. Inoculant treated silages resulted in a significantly enhanced aerobic stability, on average by 49% (P<0.05), at all ensiling periods compared to Control. The best aerobic stability was observed in silages treated with inoculants FC and AS 100. The decreased number of yeast and mold of inoculated silages compared with control silage supports the finding that the aerobic stability has improved by inoculation. AS100 treatment showed the lowest lactic acid and highest lactic acid concentrations compared with Control and other inoculants treatments. Concentration of butyric acid, alcohols, ammonia-N was lower (P<0.05) in the inoculated silages compared with the control silage. These findings are in agreement with the results presented in other studies in which *Lactobacillus buchnery* was used alone or in combination with homolactic bacteria (Schmidt and Kung, 2010).

**BACK** 

Ensiling time	Factor	Treatment	DM, g/kg	рН	Yeast, log <sub>10</sub>	Mold, log <sub>10</sub>	Weight loss, g/kg	AS, hours
	1	Control	340	1 55	cfu/g	cfu/g	47.82	22.4
2 day				4.55 <sub>2,3,4</sub>	5.082,3,4	3.49 <sub>2,3,4</sub>		32.4 <sub>2,3,4</sub>
2 day	2	FC	339	4.391,3,4	4.321,3,4	$2.52_1$	41.51	49.21
	3	AS 200	340	4.36 <sub>1,2,4</sub>	4.84 <sub>1,2,4</sub>	2.461	43.8	46.81
	4	AS 100	340	4.401,2,3	4.64 <sub>1,2,3</sub>	2.631	44.3	50.41
	1	Control	338	4.422,3,4	4.72 <sub>2,3,4</sub>	3.12 <sub>2,3,4</sub>	77.7 <sub>2,3,4</sub>	42.0 <sub>2,3,4</sub>
4 day	2	FC	338	4.221,3,4	3.631	2.411	54.51	55.21,4
	3	AS 200	338	4.181,2,4	3.76 <sub>1,4</sub>	2.461	55.41	55.21,4
	4	AS 100	338	4.25 <sup>1,2,3</sup>	3.51 <sub>1,3</sub>	2.401	58.91	66.01,2,3
	1	Control	325 <sub>2,3,4</sub>	4.24 <sub>2,3,4</sub>	4.63 <sub>2,3,4</sub>	2.86 <sub>2,3,4</sub>	105.7 <sub>2,3,4</sub>	69.6 <sub>2,3,4</sub>
8 day	2	FC	3381	4.051	2.47 <sub>1,3,4</sub>	1.971	61.1 <sub>1</sub>	98.4 <sub>1,3,4</sub>
	3	AS 200	3321	4.031,4	2.74 <sub>1,2,4</sub>	1.871	60.7 <sub>1,4</sub>	92.40 <sub>1,2,4</sub>
	4	AS 100	3281	4.051,3	2.22 <sub>1,2,3</sub>	1.951	66.41,3	115.2 <sub>1,2,3</sub>
	1	Control	319 <sub>2,3,4</sub>	4.11 <sub>2,3,4</sub>	3.93 <sub>2,3,4</sub>	2.71 <sub>2,3,4</sub>	117.7 <sub>2,3,4</sub>	78.0 <sub>2,3,4</sub>
16 day	2	FC	339 <sub>1,4</sub>	3.951,3,4	1.341	1.781	64.51,4	109.21
	3	AS 200	3291,4	3.981,2	1.481	1.741	62.61,4	120.01
	4	AS 100	323 <sub>1,2,3</sub>	3.99 <sub>1,2</sub>	1.401	1.621	69.6 <sub>1,2,3</sub>	115.21
	1	Control	316 <sub>2,3,4</sub>	4.01 <sub>2,3,4</sub>	3.32 <sub>2,3,4</sub>	2.67 <sub>2,3,4</sub>	125.7 <sub>2,3,4</sub>	88.0 <sub>2,3,4</sub>
32 day	2	FC	3381,4	3.891,3,4	1.061,.3	1.451,4	68.61,3	146.4 <sub>1,3</sub>
	3	AS 200	3281,4	3.92 <sub>1,2</sub>	1.46 <sub>1,2,4</sub>	1.51 <sub>1,4</sub>	62.0 <sub>1,2,4</sub>	124,81,2,4
	4	AS 100	322 <sub>1,2,3</sub>	3,93 <sub>1,2</sub>	1.00 <sub>1,3</sub>	1.06 <sub>1,2,3</sub>	68.0 <sub>1,3</sub>	142.8 <sub>1,3</sub>

 Table 1
 Silage parameters after different ensiling periods, ensiled with or without additives

Factor nr. 1- different (P<0.05) from factor nr 2, 3, 4 and etc.; Factor nr. 2- different (P<0.05) from factor nr 1, 3, 4, 5 and etc.

## Conclusions

The SiloSolve inoculants were efficient at reducing pH, weight loss, yeast and mold counts at all the investigated ensiling periods in this study. The SiloSolve products significantly enhanced aerobic stability of whole crop maize silage compared with ordinary fermented silage.

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#### Oxygen Barrier ('Silostop') Stretch-Wrap Film Improves Hygienic Quality of Baled Silage BACK

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#### Introduction

Baled silage is normally conserved by wrapping wilted forage with multiple layers of stretch-wrap film of 25µm thickness. The introduction of oxygen barrier (OB) stretch-wrap film (Borreani and Tabacco, 2005, 2008, 2012) offers more effective protection from oxidative losses and undesirable microorganisms such as yeasts, moulds and listeria in the outer layer of the bale than the same number of layers of standard polyethylene (PE) stretch wrap film. In a meta-analysis of 10 paired comparisons, mean total loss of dry matter (DM) during storage of baled silage was 76.8 g/kg for PE film compared to 45.6 g/kg for OB film (Wilkinson and Fenlon, 2014).

The aim of the research reported here was to test the hypothesis that use of OB film improves preservation and hygienic quality of silage in both outer and central areas of the bale.

#### **Material and Methods**

The trial was undertaken on a commercial farm in Gippsland, Victoria, Australia. A crop of perennial ryegrass (*Lolium perenne*) was wilted for 24 hours after mowing, baled with a round baler (John Deere 960) on 7 November 2014 and inoculated at baling with Trilac 3300 silage inoculant (Lallemand Animal Nutrition Ltd, Australia). Bales were wrapped in the field within 1 hour of baling with a McHale 991 bale wrapper before being transported and stacked at the farm. The wrapper applied 55% stretch to the film during wrapping. Bales were wrapped individually with either 4 layers of standard PE film of 25 µm thickness (PE) or 4 layers of OB film of 25µm thickness (SS, 'Silostop', B Rimini Ltd, London, UK). Samples (2 per bale) were collected from 5 bales wrapped with PE and 5 bales wrapped with SS on 22 June 2015 after 227 days storage. Samples were taken (by corer 650 mm length and 50 mm diameter) from outer layer of the centre of the side of each bale, from 0 to 200 mm depth, and also from the centre of the bale. The central core was aimed to be in the centre of the bale from all directions and was the section of core 400 mm to 600 mm depth. The samples (WSC), neutral detergent fibre (NDF) acid detergent fibre (ADF), lignin, ash, yeasts and moulds. Statistical analysis was by two-way ANOVA with type of film (PE v SS) and depth of sampling (outer v centre) as main effects. Yeast and mould counts were log (10) transformed prior to statistical analysis.

#### **Results and Discussion**

No holes were found in the film in any bales. Mean values for composition and hygienic quality of silage stored under PE or SS in the outer or centre of bales are in Table 1. There were no differences between type of film or depth of sampling in silage DM or pH value. Ash concentrations were lower for SS bales than PE bales at both depths of sampling (P=0.09), indicating lower loss of organic matter from bales wrapped with SS film than from those wrapped with PE film. This was confirmed by higher concentrations of residual WSC (P=0.08) and a trend of lower concentration of lignin (P=0.18) in silage wrapped with SS film than in silage wrapped with PE film. Crude protein was lower for bales wrapped in SS than PE film and lower in centre than outer layers. A similar trend was seen for  $NH_3-N$ . These differences and those in NDF content might have reflected changes in crop composition during the baling process since bales were wrapped with SS after those wrapped with PE.

Counts of yeasts were lower for bales wrapped with SS than PE film in both outer and centre layers (P=0.01), despite the small number of bales (5) per treatment, confirming the hypothesis that OB film improved hygienic quality throughout the bale by restricting ingress of oxygen not only into the outer layer but also into the centre of the bale. Mould counts were similar at log 3 cfu/g for all samples.

The results reported here confirm the results of previous comparisons between OB and PE film with baled ryegrass silage. Borreani and Tabacco (2005) found lower counts of yeasts and clostridia spores in samples taken from both the periphery (0 to 200 mm depth) and centre (210 to 550 mm depth) of bales of Italian ryegrass (*L. multiflorum*) silage (300 g DM/kg) wrapped in 6 layers of OB film compared to 6 layers of PE film after 120 days storage. Borreani and Tabacco (2012) recorded a lower yeast count in Italian ryegrass silage of similar DM concentration to that of the present

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study (598 g DM/kg) stored for 140 days in bales wrapped with 4 layers of OB film (oxygen permeability 32  $\text{cm}^3/\text{m}^2/24\text{h}$  at 20°C, 1 bar and 0.65 RH) than in bales wrapped in 4 layers of PE film (oxygen permeability 7989  $\text{cm}^3/\text{m}^2/24\text{h}$  at 20°C, 1 bar and 0.65 relative humidity (RH); yeast counts were 2.59 log cfu/g for OB film and 3.17 log<sub>10</sub> cfu/g for PE film, P<0.01). In the experiment of Borreani and Tabacco (2012), loss of DM during the storage period was 55 g/kg for bales wrapped with 4 layers of OB film compared to 80 g/kg for bales wrapped with 4 layers of PE film (P<0.001).

 Table 1
 Mean composition and hygienic quality of silage stored in bales wrapped with four layers of either standard polyethylene (PE) or oxygen barrier (SS) film.

		Туре	of film		s.e.m.	Significance of effects		
	I	PE		SS		Type of film	Depth of sampling	Film x Depth
Depth of sampling	Outer	Centre	Outer	Centre				
DM (g/kg FW)	522	531	557	576	9.53	NS	NS	NS
pH	4.94	4.89	4.80	4.89	0.021	NS	NS	NS
Ash (g/kg DM)	86.2	88.7	80.7	77.8	1.41	0.09	NS	NS
WSC (g/kg DM)	57.7	66.4	70.9	76.1	1.63	0.08	NS	NS
CP (g/kg DM)	160	150	144	125	1.75	0.01	0.003	NS
NH <sub>3</sub> -N (g/kg total N)	29.8	22.1	24.3	17.7	1.26	NS	0.10	NS
NDF (g/kg DM)	512	526	522	560	6.85	0.08	0.04	NS
ADF (g/kg DM)	344	349	345	362	3.51	NS	0.10	NS
Lignin (g/kg DM)	60.6	57.1	56.5	54.9	1.57	0.18	NS	NS
Yeasts (Log <sub>10</sub> cfu/g FW)	4.94	5.58	3.80	3.36	0.343	0.01	NS	NS

FW = Fresh weight. NS = Not significant at P=0.2.

#### Conclusions

Hygienic quality of silage, assessed by yeast count, was higher in both outer and central areas of bales wrapped with oxygen barrier (SS) film than in bales wrapped with the same number of layers of standard polyethylene (PE) film. Ash concentration was lower and residual WSC was higher in silage wrapped with SS film than for silage wrapped with PE film, indicating lower loss of organic matter from bales wrapped with SS film than from those wrapped with PE film.

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## Effect of *Lactobacillus Kefiri* Alone and in a Silage Inoculant Blend on the Fermentation Quality of Grass Silage

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#### Introduction

Multi-strain silage inoculants containing homo- and heterofermentative Lactic Acid Bacteria (LAB) are commonly used worldwide to improve silage quality. A good balance between lactic acid and acetic acid induced by a wellbalanced ratio of homofermentative and heterofermentative silage inoculant strains is essential to achieve both palatability and aerobic stability. *Lactobacillus kefiri* DSM 19455 is a novel heterofermentative strain proprietary to Biomin Holding GmbH, Austria. The aim of this trial was to better characterize the effects of *Lactobacillus kefiri* DSM 19455, a novel heterofermentative strain, alone and in a three-strain silage inoculant blend on the fermentation quality of grass silage.

#### **Materials and Methods**

Meadow grass, harvested in April 2015 was inoculated with *L. kefiri* DSM 19455 alone or with Biomin<sup>®</sup> BioStabil Plus (BIOMIN, Getzersdorf, Austria), comprising a blend of *L. kefiri* DSM 19455, *L. brevis* DSM 23231 and *L. plantarum* DSM 19457 strains. At ensiling 15 kg of raw material per group were taken from the total available material (weighed into plastic bag) and distributed on a plastic film. The silage additive was evenly sprayed on the raw material and afterwards mixed by hand. Mini-silos (plastic buckets with tightly closing lid) with a volume of 1.8 L and 5.8 L were fully filled and compacted with hydraulic pressure to a density of 2 kg DM/5 L. Uniform density was achieved by closely monitoring the packing pressure, which was 3 bar for small sample silos (1.8 L) and 6 bar for large sample silos (5.8 L). Immediately after packing, the plastic bag lining was sealed airtight with a cable binder and the buckets closed with their according lids. Airtight test silos were stored at 22 ( $\pm$  2) °C. In addition, three fresh forage samples were analyzed for pH and fermentation parameters at 7, 46, and 92 days and additionally for nutritional composition at 46 and 92 days with 5 replicates each per treatment per time point. ANOVA and orthogonal contrasts were calculated using IBM SPSS 19.0 software. The level of significance was 0.05 for all evaluations.

#### **Experimental treatments**

T1: Negative control (no additives, but same amount of water)

**T2**: 2 x 10<sup>5</sup> cfu/g forage *L. kefiri* DSM 19455.

**T3**: 2 x 10<sup>5</sup> cfu/g forage *L. plantarum* DSM 19457, *L. brevis* DSM 23231 and *L. kefiri* DSM 19455 (Biomin<sup>®</sup> BioStabil Plus, BIOMIN, Austria).

#### **Results and Discussion**

The initial pH of the grass substrate used for ensiling was typical for meadow grass. Microbiological enumeration of herbage showed that the number of yeasts and molds was typical for the epiphytic microflora characteristics in Austrian conditions for first cut meadows (3.18 log cfu/g FM). The forage had low protein content (drought stress) and low fibre content (early harvest) together with high sugar content (early harvest). High crude ash content indicates soil contamination.

Nutritional composition of the silage after 92 days of fermentation is shown in Table 1. Inoculation with T3 led to significantly higher energy values (+ 0.28 MJ NEL) than T1, whereas *L. kefiri* (T2) did not influence the silage energy content. Dry matter losses in first cut silage (grass 1) were as expected: T3 preserved DMc; control and *L. kefiri* alone did not.

	Unit	T1 -	T2 -	T3 - BioStabil	p-value of the
		Control	L. kefiri	Plus	ANOVA
Dry matter, DM	g/kg FM	$307.00 \pm 2.45$	$304.80\pm5.36$	$328.20 \pm 7.26$	0.008
Crude protein	g/kg DM	$141.00 \pm 3.74$	$138.20 \pm 1.79$	$133.60 \pm 2.41$	0.013
Crude fat	g/kg DM	$34.40 \pm 1.34$	$32.00 \pm 1.22$	$32.40 \pm 0.55$	0.033
Crude fibre	g/kg DM	$249.80 \pm 12.79$	$240.40 \pm 12.74$	$226.40\pm6.80$	0.019
Crude ash	g/kg DM	$108.40\pm2.70$	$112.60 \pm 5.50$	$100.60 \pm 2.30$	0.004
Ammonia N	g/kg total N	$8.34\pm0.98$	$6.76 \pm 1.41$	$5.23 \pm 0.43$	0.001
pH after 7 days		$4.76\pm0.05$	$4.19\pm0.04$	$4.08\pm0.24$	< 0.001
pH after 92 days		$4.38\pm0.35$	$4.40\pm0.03$	$4.03\pm0.03$	< 0.001
ME	MJ/kg DM	$10.24\pm0.14$	$10.26 \pm 0.13$	$10.62 \pm 0.11$	0.001
NEL	MJ/kg DM	$6.15 \pm 0.11$	$6.12 \pm 0.13$	$6.43 \pm 0.11$	0.001
DMc losses	g/kg DMc	$50.85\pm10.47$	$49.55 \pm 11.28$	$36.94 \pm 18.42$	0.053

Table 1 Averages of nutritional composition, pH and DM loss after 92 days of fermentation in the treatment groups

T2 and T3 significantly lowered the pH of the silage already after 7 days compared to T1 (p < 0.001). The grass was easy to ensile, therefore the spontaneous fermentation in T1 acidified the silage quite well, so that finally after 91 days only T3 silage was significantly more acidic than the other two treatments (p < 0.001) (Table 1). T3 treatment led to pronounced lactic acid fermentation with moderate acetic acid levels, whereas T2 formed high amounts of acetic acid. Butyric acid was only present in T1, however in none of the inoculated groups T2 or T3 (Table 2).

 Table 2
 Fermentation parameters after 92 days of ensiling in the treatment groups

Metabolite [g/kg DMc]	T1 - Control	T2 - L. kefiri	T3 - BioStabil Plus	p-value of ANOVA
Lactic acid	$78.50 \pm 10.28$	$22.66 \pm 5.20$	$100.38 \pm 10.24$	0.000
Acetic acid	$24.02 \pm 3.88$	$84.09\pm8.78$	$19.39 \pm 3.21$	0.000
Propionic acid	$6.94\pm3.90$	$0.00\pm0.00$	$0.00\pm0.00$	0.007
Ethanol	$42.07\pm3.40$	$7.67 \pm 0.56$	$4.89\pm4.48$	0.009
Butyric acid	$2.74\pm3.20$	$0.00\pm0.00$	$0.00\pm0.00$	0.032
1,2-Propanediol	$0.00\pm0.00$	$17.07\pm0.98$	$0.00\pm0.00$	0.001
1-propanol	$0.00\pm0.00$	$0.00\pm0.00$	$0.00\pm0.00$	n.a.
Succinic acid	$2.04\pm4.57$	$0.30\pm0.68$	$5.10\pm0.92$	n.a.

#### Conclusion

In conclusion, *Lactobacillus kefiri* DSM 19455 was demonstrated to be a typical heterofermentative silage bacterium, producing high levels of acetic acid. The blend of *L. kefiri* DSM 19455, *L. brevis* DSM 23231 and *L. plantarum* DSM 19457 at 2 x  $10^5$  cfu/g forage significantly improved the fermentation quality and energy contents of grass silage and prevented fermentation losses during fermentation.

#### Effect of Additives on Lucerne Silage with High Dry Matter

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#### Introduction

The dry matter of lucerne silage has great importance for the development and result of the fermentation process. An optimal dry matter of ensilaged fodder is a natural preservative. The mown matter should not be left in place more than two days; the fodder should not become wet due to rain. With increasing time of stay of the matter in the field, the negative epiphyte microflora gets more activated, the fermentation process and the nutrient contents gets more endangered (due to breathing, growth of proteolysis, ashes, etc.), and the health soundness of silages gets impaired. If the fodder dries too much, the risk of losses rises both in the field and due to inappropriate fermentation and to mould infestation. As soon as the withered lucerne starts losing leaves due to loss of water, the most precious items grown will stay in the field. The ensilaging ability of lucerne is stated to be ideal with a dry matter of 40 % (Merchen and Sattern, 1983). Makoni et al (1993) found that withering increases the proportion of more soluble fractions of nitrogen substances, but at the same time also the proportion of chloroplast. Fermentation in anaerobic environment leads to change the content of nutrients, primarily of nitrogen substances and fibre. Proteolytic changes of lucerne silages, aimed at reduction of degradation of proteins under use of different silage agents were studied for example by Guo et al (2008).

The experiment was aimed at finding out the effect of selected preservatives on the result of nutritional values and of the fermentation process at ensilaging lucerne with dry matter of about 60 %.

#### **Materials and Methods**

The lucerne, after 2 days of withering at sunny weather, was mown by a cutter with 3 and 1.5 cm TLC, with dry matter oscillating between 58.6 and 60.1 %. Penn State Particle Separator with sieve mesh of 19 mm and 8 mm was used to ascertain the actual length of the chops. 5 variants in three repetitions were stored in special bags, with 10 kg of chops from each variant. The control variant was without any preservative; the test variants had 4 preservatives applied in the chops: (M) formic acid in a dose of 4.5 l/t, (A) Albit (Polybetahydroxybutyricacid) in a dose of 1 l/t, (C) Alicin (garlic extract) in a dose of 60 g/t and (F) Formasil (bacterial-enzymatic *P. pentosaceus, L. plantarum,* beta-glucanase, xylanase) in a dose of 100 g/t. The Thermochron sensor, calibrated for measurement of temperature each 60 minutes, with accuracy of 0.065°C, was inserted into each bag. Subsequently, the bags were put into the ensilaged matter in the bunker silo, covered with a layer of silage and tamped down by a tamper with wagon wheels.

After 4 months of storage in the bunker silo, the bags were removed from the silage, weighed and transported to the laboratory for standard-method chemical analyses (AOAC, 1995). Stability was measured according to Ranjit and Kung (2000). The results were statistically processed under use of the Statistica var. 10 (StatSoft, USA) program, by the ANOVA multifactorial procedure and by the subsequent POST-HOC Tukey test.

#### **Results and Discussion**

The physically efficient fibre (peNDF) was established at 31.9 % in the longer chops and at 21.3 % in the shorter chops.

There were no significant differences in nutritional values between the variants tested (P>0.05). The situation was similar in the fermentation indicators, except for lactic acid – the significantly highest amount (P<0.05) of lactic acid was produced in silages treated with formic acid and with shorter chops. The differences between the control silages without preservatives and the silages with preservatives were not significant. The results are in accordance with the results presented by Guo et al (2008).

The pH values for the fermentation process without development of butyric acid were at a critical limit of 5.3 (Weisbach et al, 1974) with the high level of dry matter. But they did not exceed it markedly in no variant, which had the positive effect of detecting no or only trace amounts of butyric acid, under 0.03 %. The acidity values of the water extract were relatively high (1697 mg KOH/100g), showing that although the dry matter was high, the fermentation did take place. Merchen and Sattern (1983) compared lucerne silages with dry matter of 40 % and 66 %. In silages with 66 % of dry matter, almost all quality indicators were worse, except for the NH3-N content. In silages with 66 % of dry matter, they achieved a similar pH value result to that of our experiment (5.40 vs. 5.33).

The differences between the variants could be found particularly when evaluating the aerobic stability. In silages with longer chops, the aerobic stability was 33 hours shorter in average (48 hours in silages with longer chops, against 81 hours in silages with shorter chops). The lowest aerobic stability was found in silages with longer chops without additives (26 hours) and with formic acid (30 hours), while the highest stability was found in silages with longer chops, preserved by Alicin (60 hours) and with Albit (81 hours). When silages were made with shorter chops, the temperature differences between silages with different additives were minimal. Fermentation indicators were established for all variants after 144 hours. The silage with shorter chops without preservative showed worse (P<0.05) results of aerobic

stability for lactic and acetic acid and for ammoniac nitrogen that acts as an indicator of increased proteolysis. The silages with longer chops showed worse results of aerobic stability in all variants and in all indicators. That leads to the conclusion that silages with higher dry matter, primarily those with longer chops (peNDF 31.9 %), must be fed as soon as possible, otherwise their quality will significantly decrease.

Index	V	В	FA	А	С	F	SEM	D/K	Ad	D/K*Ad
рН	D	5.37	5.32	5.34	5.33	5.29	0.01	0.725	0.014	0.311
	Κ	5.37	5.28	5.32	5.35	5.33				
AWE	D	1648	1692	1692	1714	1714	9.16	0.727	0.124	0.246
(mg KOH/100g)	Κ	1671	1735	1714	1671	1693				
LA	D	1.14 <sup>a</sup>	$1.17^{ab}$	1.14 <sup>a</sup>	1.11 <sup>a</sup>	1.08 <sup>a</sup>	0.28	0.148	0.009	0.063
(% FM)	Κ	1.11 <sup>a</sup>	<b>1.47</b> <sup>b</sup>	1.12 <sup>a</sup>	1.10 <sup>a</sup>	1.14 <sup>a</sup>				
VFA	D	0.39	0.41	0.42	0.39	0.38	0.01	0.342	0.121	0.317
(% FM)	Κ	0.40	0.51	0.38	0.40	0.40				
LA/VFA	D	2.92	2.85	2.71	2.85	2.84	0.07	0.983	0.976	0.926
	K	2.78	2.88	2.95	2.75	2.85				
N-NH <sub>3</sub>	D	29.0	28.1	28.3	28.2	31.5	0.99	0.981	0.617	0.502
(mg N/100 g)	Κ	26.9	33.0	28.5	27.7	29.2				

 Table 1
 Indicators of silage fermentation

V = variant, D = longer chop, K = shorter chop, B = control without additive, FA = formic acid, A = Albit, C = Alicin, F = Formasil, SEM = standard error of means, D/K = longer/shorter chop; Ad = additive, X = S variant for K has not been established; AWE = Acidity of water extract; LA, Lactic acid; FM = fresh matter, VFA = Volatile fatty acids; Different letters <sup>a,b,c</sup> in the index numbers indicate significant (P <0.05) differences between

#### Conclusion

When ensilaging lucerne with high dry matter (about 60 %), the results of nutritional values and of fermentation indicators were comparable between the variant without preservative and with selected preservatives. It can be stated that the main preservative (preservation agent) consisted in the higher content of dry matter. The aerobic stability of the silages with longer chops without preservatives and with formic acid was lower (26 and 30 hours, respectively) than with other preservatives (43-81 hours).

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## Fermentation of Early Cut Whole Crop Field Beans (Vicia Faba) Affected by Silage Additives

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#### Introduction

Grain legumes can provide ruminants as well as monogastrics with farm grown protein and reduce the global warming potential. The ripening of grain legumes under humid climate conditions is uneven and results in considerable field losses or the harvested grains may require expensive drying. These problems could be overcome by ensiling as well as the feeding value might be improved (Doblado et al. 2003). However, the grain legumes are characterized by a low sugar buffer capacity ratio (S/BC) (Anonymus, 2014, Gefrom et al., 2013). Consequently, the ensilability of whole crop grain legumes seems to be difficult. On the other hand, fermentation is also affected by the DM content and a low BC and a sufficient high DM may result in a good fermentation coefficient. The fermentation can be enhanced by adding molasses and LAB but butyric acid might be still high at low plant nitrate contents (Heinritz et al., 2012, Gonzales et al., 2012). We analysed the effect of different silage additives on the fermentation of whole crop field beans.

#### **Materials and Methods**

Field beans (Vicia faba L.) were grown on loamy soil in the North West of Bremen. The plant material was harvested at the 14<sup>th</sup> of August 2014. The beans reached yellow ripeness (DM 37.0 %, ingredients in % DM: crude protein 22.5, crude fibre 17.8, WSC 3.5, starch 27.7, nitrate 0.54). The buffer capacity was 4.2 g LA/100 g DM, the WSC/BC ratio 0.8 and the fermentation coefficient 44. Immediately after harvest the plant material was chopped and ensiled in 2.5-L glass jars in a randomized block design with 3 replications. The jars were stored for 103 days at a constant temperature of  $25^{\circ}$ C. The treatments were: C = control (sprayed with 100 ml tap water), LABho = homofermentative LAB with  $1x10^5$  cfu/g forage applied with 100 ml/g forage, HMT = chemical additive (hexamethylene tetramin and sodium nitrite, 3 l/t), LABho/he = combination of homo- and heterofermentative LAB with 2.5  $10^5$  cfu/g forage, applied with 100 ml/g forage, BeSo = chemical additive (benzoate, sorbate, acetate, 2 l/t). Chemical analysis of the plant material was performed according to the official German standards for feed evaluation (Anonymus, 2011). Silage DM was measured and corrected for loss of volatiles during drying (Weißbach and Kuhla, 1995). The pH-value was analysed by a pH-electrode. Determination of the fermentation acids was done by HPLC for lactic acid and by GC for volatile fatty acids and alcohols. Ammonia was analysed photometrically. The DM losses during fermentation were calculated according to Weißbach (2005). Aerobic stability was measured by the temperature method (Honig, 1990). Data were evaluated by using procedure MIXED of SAS. Differences among means were compared at a significant level of  $P \le 0.05$ .

#### **Results and Discussion**

After 103 days of fermentation, all silages were of a good fermentation quality. The DM losses were reduced by the HMT and LABho/he treatments (Tab. 1). Fermentation losses during the anaerobe phase typically come from the metabolism of the groups of the enterobacteria and clostridia. Because of the low butyric acid concentration in the silages, acetic acid fermentation probably contributed to these losses most. In case of HMT, the low fermentation losses corresponded to a low acetic acid concentration in the silage. With LABho/he on the other hand, we found the highest amount of acetic acid and low fermentation losses. That may also have come from a heterofermentative metabolism of the lactic acid bacteria (LAB) in the inoculant. In this experiment, we did not analyse 1,2 propanediole to describe a possible *Lactobacillus buchneri* type effect (Weiß and Krause, 2011). However, also a heterofermentative metabolic pathway gives more DM losses compared to a homofermentative lactic acid fermentation. Therefore, it's not clear why high acetic acid concentrations corresponded with low DM losses in case of the LABho/he treatment.

The pH value was lowest in the HMT silage which corresponded with the highest amount of lactic acid and lowest content of acetic acid (Tab. 1). Not in line with the data in literature were the inoculant treatments (Gefrom et al., 2013; Gonzales et al, 2012) resulting in higher pH levels and lower lactic acid concentrations, compared to the control. These treatments with the highest pH value also had the highest acetic acid concentrations. The control and all tested treatments showed quite low contents of butyric acid (<0.1 % of DM). Heinritz et al. (2012) found quite high amounts of butyric acid in grain legume silages but low amounts of nitrate (<2 g/kg DM). The plant material used for ensiling was characterized by a sufficient nitrate concentration (>4.4 g/kg DM). The strong positive impact of nitrate on butyric acid free silages and the related fermentation pathway was described by Kaiser et al. (1997).

**BACK** 

	pH value	Lactic acid	Acetic acid	Butyric acid	NH3-N of total N	DM losses
			(% of DM)		(%)	(%)
Control	4.40a	5.16a	1.57a	0.08a	4.46a	5.33a
LABho	4.93b	3.21b	2.67b	0.00b	4.77a	5.73a
HMT	4.23c	6.51c	1.49a	0.00b	3.37b	4.17b
LABho/he	5.40d	1.71d	3.17c	0.08ab	4.27a	4.23b
BeSo	5.30e	2.04e	3.08c	0.00b	3.62ab	6.37a

 Table 1
 Effect of different additives on fermentation acids, pH, NH<sub>3</sub>-N of total N and DM-losses of a field bean (*Vicia faba*) whole crop silage

HMT=chemical additive hexamethylene tetramin and sodium nitrite; LABho=homofermentative lactic acid bacteria; LABho/he=homo and heterofermentative lactic acid bacteria; BeSo=chemical additive, benzoate, sorbate, acetate; different letters show significant differences at P <= 0.05

The ammonia-N in silage was only affected by the HMT additive (Tab. 1). If we consider that proteolysis only occurs up to a minimal pH of 5 (Seyfarth et al., 1989), the fast and long-lasting (103 days) low pH may have affected protein break down. However, the ammonia-N concentrations in the other treatments were around 4 and did not really indicate any severe protein break down.

As expected the aerobic stability of the silages (data not shown) was lowest in the HMT treated silage. But also in the LABho/he treatment which reached as well 5 days the aerobic stability was unexpected low. Also in the other silage treatments, temperature was stable only for 6.5 days.

#### Conclusions

Whole crop field beans can be preserved by ensiling. The DM losses are at an acceptable level and can be reduced by using a suitable additive such as HMT. With HMT as an additive, the highest lactic acid and lowest acetic acid levels were found in the silages as well as the lowest DM losses. The pH value was low and no butyric acid was formed when the HMT was applied. An undesirable fermentation can be prohibited by suitable additives, but there is a risk of heating and hygienic deterioration.

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## Effects of Delayed Filling and Additive Use on the Quality of Pressed Sugar Beet Pulp Ensiled in Plastic Bags

**BACK** 

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#### Introduction

Pressed sugar beet pulp (PSBP) is frequently ensiled in plastic bags and represent a valuable feed component of feed rations for ruminants. However, its high susceptibility to fungal infestation during feed-out, especially in summer, often results in low stability and poor hygienic quality (Kalzendorf, 2007). Therefore, especially the use of chemical additives containing salts of sorbic, benzoic and and/or propionic acids, has attracted significant attention in order to control fungal growth (Weber et al., 2006b; Potthast et al., 2014). In the sugar beet processing season, PSBP is continuously produced but, sometimes, legal restrictions to transport on weekends require storage on-site before dispatch. Therefore, the aim of this study was to test the effects of delayed bag filling and additive use on the quality and stability of PSBP ensiled in plastic bags.

#### Materials and methods

The PSBP (about 50 t per treatment) were obtained from the sugar plant of Südzucker AG in Zeitz, Germany, in the late processing season in 2014. The material for the treatment with delayed filling (DEL) was produced on 13 Dec., 11.30 pm to 12 pm, followed by the production of the treatment with delayed filling plus additive (DELADD) on 14 Dec., 0.30 am to 1.30 am, to which 2 l/t of Xtrasil excel HD (KONSIL Scandinavia AB, containing 250 g/l sodium benzoate, 150 g/l potassium sorbate, 50 g/l ammonium propionate) were added by spraying during transport from the extraction site to the storage place in the sugar plant. These materials were loosely piled up in the sugar factory until dispatch. The material for treatment FRESH was produced in the morning of 15 Dec just before all PSBP was dispatched to the trial site in Kleinbautzen, Germany, where they were pressed into plastic bags (diameter: 2.7 m; length: about 8 m; plastic thickness: 215 µm) by a rotor bagger (RT8000, BAG Budissa GmbH). For each treatment, samples for routine chemical and microbiological analyses were taken during bag filling (n=5) and upon opening after 223 days on 13 July 2015 (n=3 from upper layer, 20 cm below surface; n=3 from lower layer, 1 m above ground). Fermentation variables and sugar were determined by HPLC, and the temperature method by Honig (1990) was employed to evaluate aerobic stability (ASTA). The extent of aerobic deterioration (AD) was expressed as the accumulated differences (K) in temperature between sample and ambient measured in 2 h-intervals for 336 hours. Statistical analysis was performed by using the procedures GLM, REG and CORR (Pearson correlation coefficient) of SAS. Differences between least-square (LS) means were analysed by Tukey's test. Significance was declared at P < 0.05, tendency to significance at 0.05<*P*<0.10.

#### **Results and Discussion**

Data on chemical composition of PSBP presented in Table 1 compare well with those by Weber et al. (2006a). The higher sugar concentration may have been caused by differences in the extraction technology. Differences in chemical composition between treatments were small and may be associated with batch-to-batch variation. On the contrary, the higher yeast count in DEL compared to FRESH should be attributed to their growth during storage on-site, which could be prevented by additive use in DELADD.

Table I Char	<b>Table 1</b> Characteristics of sugar beet pulp at bug mining (ii=5, Ebineans given in grkg Divi aness stated otherwise)										
Treatment	$DM^1$	Crude	Crude	Crude	NFC <sup>2</sup>	Sugar <sup>3</sup>	Metabolizable	Yeasts	Moulds		
		ash	protein	fibre			energy	(log <sub>10</sub>	cfu/g)		
DEL	207 <sup>b</sup>	55	85ª	185 <sup>a</sup>	380	157 <sup>ab</sup>	12.2	5.68ª	1.76		
DELADD	207 <sup>b</sup>	57	84 <sup>b</sup>	177 <sup>b</sup>	382	174 <sup>a</sup>	12.2	4.33 <sup>b</sup>	1.70		
FRESH	219 <sup>a</sup>	57	83 <sup>b</sup>	183 <sup>a</sup>	364	147 <sup>b</sup>	12.2	4.64 <sup>b</sup>	1.98		
SEM	1.6	0.9	0.4	0.6	5.6	6.3	0.01	0.210	0.126		
P-value	< 0.001	ns	< 0.01	< 0.001	0.090	$<\!0.05$	ns	< 0.001	ns		

	Table 1	Characteristics of suga	r beet pulp at	t bag filling (n=	=5, LSmeans give	en in g/kg DM	unless stated otherwise)
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<sup>1</sup>dry matter; <sup>2</sup>non-fibre carbohydrates, <sup>3</sup>sum of sucrose, glucose, fructose, maltose, lactose, xylose; galactose, arabinose, ribose, mannitol, values in columns bearing different superscripts differ, ns not significant.

With the exception of the yeast count, all parameters measured in PSBP silages were affected by treatment (Table 2). Silages of treatment FRESH showed the lowest fermentation intensity which was accompanied by the largest sugar concentration, and DELADD reduced the extent of sugar degradation compared with DEL. This may be explained by the likely higher temperature of FRESH at bagging although temperature was not measured. The generally low fungal counts in PSBP silages can be attributed to the extended storage length (Middelhoven and van Baalen, 1988). Concentrations of alcohols were highest in DEL, which likely was caused by yeast development during storage before

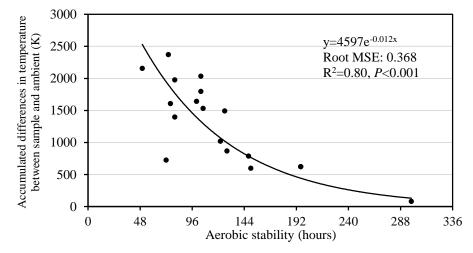
bagging (Table 1) and in the early phases of fermentation. Use of the chemical additive improved ASTA at a similar magnitude as reported by Weber et al. (2006b) and Potthast et al. (2014), and concomitantly restricted AD. These two variables were highly negatively correlated (r=-0.89, P < 0.001, Figure 1). Sampling location had variable effects on the tested parameters. Yeast count was higher, and a strong trend towards impaired ASTA was observed in the upper bag layer, which substantiates observations by Weber et al. (2006a). The lower compaction of PSBP silage at this location (Weber et al., 2006a) may result in greater porosity available for gas exchange through the plastic during storage.

 Table 2
 Effects of delayed filling, additive use and sampling location on the quality of sugar beet pulp ensiled in

 Image: the second secon

plas	tic bags a	and stored	for 223 day	ys (data pre	sented as LS	means in g	/kg DM <sup>1</sup> u	inless state	plastic bags and stored for 223 days (data presented as LSmeans in g/kg DM <sup>1</sup> unless stated otherwise)											
Main	pН	Lactate	Acetate	Ethanol	Propanol	Sugar <sup>2</sup>	Yeasts	Moulds	ASTA <sup>3</sup>	$AD^4$										
effects							(log <sub>10</sub>	cfu/g)	(hours)	(K)										
Treatment <sup>5</sup>																				
DEL	3.74 <sup>b</sup>	24.9ª	20.7 <sup>a</sup>	14.8 <sup>a</sup>	3.5 <sup>a</sup>	1.3°	2.66	1.93	110 <sup>ab</sup>	1413 <sup>ab</sup>										
DELADD	3.80 <sup>ab</sup>	23.6 <sup>a</sup>	15.7 <sup>b</sup>	1.9 <sup>b</sup>	$0.8^{b}$	47.2 <sup>b</sup>	1.90	2.05	172ª	612 <sup>b</sup>										
FRESH	3.81ª	11.6 <sup>b</sup>	10.8 <sup>c</sup>	4.5 <sup>b</sup>	$0.0^{b}$	82.5 <sup>a</sup>	2.31	2.48	86 <sup>b</sup>	1864 <sup>a</sup>										
SEM	0.018	2.07	0.94	1.18	0.48	6.55	0.250	0.151	18.2	128.4										
P-value	< 0.05	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	ns	0.056	< 0.05	< 0.001										
Location <sup>6</sup>																				
upper	3.77	22.4	17.7 <sup>a</sup>	6.6	2.5 <sup>a</sup>	38.9 <sup>b</sup>	2.78 <sup>a</sup>	2.21	100	1405										
lower	3.79	17.6	13.8 <sup>b</sup>	7.5	0.4 <sup>b</sup>	48.4 <sup>a</sup>	1.80 <sup>b</sup>	2.10	145	1188										
SEM	0.015	1.69	0.77	0.96	0.39	5.34	0.204	0.124	14.8	104.9										
<i>P</i> -value	ns	0.070	< 0.01	ns	< 0.01	< 0.05	< 0.01	ns	0.052	ns										

<sup>1</sup>corrected for the loss of volatiles during drying; <sup>2</sup>sum of sucrose, glucose, fructose, other monomers not detected, <sup>3</sup>aerobic stability, <sup>4</sup>aerobic deterioration, <sup>5</sup>n=6, <sup>6</sup>n=9, values in columns within treatment or location bearing different superscripts differ, ns not significant.





#### Conclusions

Treatment of PSBP ensiled in plastic bags with Xtrasil excel HD can generally be recommended. It alleviates the negative effects of intermediate storage and delayed filling on the quality of PSBP silage. More so, it improves aerobic stability and restricts the extent of deterioration during feed-out over that of freshly ensiled PSBP.

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### Effect of Silage Additives on Fermentation Characteristics of Maize Silage

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#### Summary

The aim of this study was to evaluate the effect of chemical and microbiological additives on fermentation characteristics in corn silage at 1, 3, 5, 10 and 90 days of ensiling. The following additives were used: commercial bacterial inoculant (2 g/t) containing homofermentative lactic acid bacteria *Lactobacillus plantarum, Lactobacillus paracasei* and *Pediococcus pentosaceus*, and chemical additive containing formic acid, propionic acid, ammonium formate and benzoic acid (4 l/t). Both additives positively influenced the fermentation process in corn silage. Their application resulted in a decreased pH in the treated compared to untreated silage at 1 day of ensiling (P<0.05). Significant pH reductions (P<0.05) were particularly observed between days 1 and 3 and between days 3 and 10 for all treatments. The highest content of water soluble-carbohydrates (WSC) was preserved in the silage treated with the chemical additive (P<0.05) at 3, 5, 10 and 90 days of ensiling. The highest and thus the most desirable value of the lactic to acetic acid ratio was obtained at 90 days of ensiling in the group with the bacterial additive (P<0.05).

Keywords: maize silage; lactic acid bacteria; formic acid; fermentation dynamics

#### Introduction

Maize silage is a highly digestible and palatable feed source valued for its nutritional composition. Ensiling is a method of long-term preservation and storage of fresh plant material under anaerobic and acidic conditions. Various types of microbial additives can be used to improve silage fermentation (Reich and Kung 2010). Chemical additives are added to ensiled forages to prevent or reduce the growth of such undesirable microorganisms as yeast or moulds, which are responsible for aerobic deterioration in silages. Chemical-based additives are useful for improving fermentation during unfavourable climatic conditions.

The objective of this study, therefore, was to evaluate the effects of bacterial and chemical additives on the number of LAB and on fermentation indicators in whole maize silage at 1, 3, 5, 10 and 90 days of fermentation.

#### **Material and Methods**

Maize (Ronaldinio hybrid; FAO 240/250) was harvested at whole-plant DM content of approximately 33.6% and chopped using a conventional forage chopper to average length of ca 12 mm for ensiling in a conventional silo. Approximately 60 kg of forage was randomly collected, it was thoroughly mixed, and pre-ensiling samples were taken for analyses. Three piles each containing approximately 20 kg of forage were prepared and treated without any additive (C), with a commercial biological inoculant (B), or with a chemical additive (CH). The bacterial inoculant, added at 2 g/t of forage, contained the homofermentative LAB *Lactobacillus plantarum, Lactobacillus paracasei*, and *Pediococcus pentosaceus* at total concentration 1.5 x 10<sup>11</sup> cfu/g of inoculant. The inoculant (0.04 g) was diluted in 80 ml of distilled water and applied by spraying onto the 20 kg forage during mixing. The chemical additive containing formic acid (42.5%), propionic acid (10.0%), ammonium formate (30.3%), and benzoic acid (2.2%) was applied at the rate of 80 ml per 20 kg of forage (4 l/t). The equivalent amount of water was applied to the untreated control forage. Chopped forage samples (700 g; n = 25 from each treatment) were packed into polyethylene bags (300 × 400 mm), vacuum sealed using a VacSy<sup>®</sup> system, and stored in a tempered dark room at +20°C. Silages (n = 5 from each treatment) were analysed for fermentation quality after 1, 3, 5, 10 and 90 days of preservation.

#### **Results and Discussion**

All maize silages were well preserved as indicated by the low pH and by the fact that no butyric acid was detected in either control or treated silages. The fermentative characteristics of maize are presented in Table 1.

Interactions between treatment and day of fermentation were detected for all the observed characteristics except for DM, thereby indicating that changes over time depended on the additive used. Significant differences in pH values between treatments were only found at day 1, whereas these were similar at any other time of fermentation. At day 1, the pH was highest for C, followed by the B treatment, and the lowest pH was detected for CH. This confirmed that added acids are more effective than is natural fermentation, because acidification occurs almost immediately after adding the additive (Charmley 2001). The pH gradually decreased from day 1 to day 10 and then remained constant. Significant pH reductions (P < 0.05) were particularly observed between days 1 and 3 and between days 3 and 10 for all treatments. The pH values at day 1 correspond with the different concentrations of lactic and acetic acids in the respective treatment groups.

The concentrations of lactic acid gradually increased over time, reaching their highest values at day 90 in all treatment groups. The most rapid elevation was detected between day 3 and day 5, during which time the levels of lactic acid almost doubled. At days 1 and 5, the concentrations of lactic acid from highest to lowest were for CH, B and C. At day 90, this was highest for B.

The lactic acid : acetic acid ratio is a good efficiency indicator for silage fermentation. In the present study, this ratio increased over time in all treatments and the highest value of 3.62 (P < 0.05) was observed for the bacterial inoculant-treated silage at day 90 of fermentation.

The WSC concentrations were rapidly reduced in C and B silages during the first 10 days of fermentation (P < 0.05), whereas these remained unchanged for CH. In agreement with Meeske et al. (2002), bacterial additive had no effect on WSC during fermentation, indicating that WSC were utilized at the same rate in untreated and inoculant-treated maize silages. The WSC concentrations were higher (P < 0.05) in CH compared to C and B at days 3, 5, 10 and 90 of fermentation. As with our study, treating maize silages with the chemical-based additives has been shown to increase residual WSC concentrations, thus suggesting partial inhibition of fermentation (Kleinschmitt et al. 2005; Da Silva et al. 2015).

	Treatment		D	Day of ensil	ing			Signi	ficance	
		1	3	5	10	90	SEM	Т	D	TxD
pН	С	$4.97^{aX}$	4.35 <sup>b</sup>	4.06 <sup>c</sup>	3.91°	3.93°	0.05	***	***	***
	В	$4.68^{aY}$	4.17 <sup>b</sup>	4.01 <sup>bd</sup>	3.78 <sup>cd</sup>	3.83 <sup>cd</sup>				
	СН	$4.34^{\mathrm{aZ}}$	4.20 <sup>b</sup>	4.10 <sup>b</sup>	3.84 <sup>c</sup>	3.84 <sup>c</sup>				
Lactic acid	С	$12.8^{aX}$	18.9 <sup>a</sup>	35.4 <sup>bX</sup>	47.6 <sup>c</sup>	57.6 <sup>dX</sup>	1.49	***	***	***
(g/kg of	В	$17.4^{aXY}$	20.0 <sup>a</sup>	39.4 <sup>bXY</sup>	45.4°	87.5 <sup>dY</sup>				
DM)	СН	20.3 <sup>aY</sup>	21.9ª	44.0 <sup>bY</sup>	49.0 <sup>b</sup>	59.4 <sup>cX</sup>				
Acetic acid	С	$14.7^{aX}$	18.5 <sup>bX</sup>	21.9°	24.2°	24.2 <sup>cX</sup>	0.72	***	***	**
(g/kg of	В	$18.5^{aY}$	22.2 <sup>bY</sup>	22.5 <sup>b</sup>	22.9 <sup>b</sup>	24.8 <sup>bXY</sup>				
DM)	СН	21.4 <sup>aY</sup>	23.0 <sup>aY</sup>	23.7 <sup>ab</sup>	25.6 <sup>bc</sup>	29.0 <sup>cY</sup>				
LA/AA	С	0.88ª	1.03 <sup>a</sup>	1.62 <sup>b</sup>	1.97°	$2.32^{dX}$	0.05	***	***	***
	В	0.95ª	0.90 <sup>a</sup>	1.75 <sup>b</sup>	1.98 <sup>b</sup>	3.62 <sup>cY</sup>				
	СН	0.95ª	0.96 <sup>a</sup>	1.85 <sup>b</sup>	1.92 <sup>b</sup>	$2.08^{bX}$				
WSC (g/kg	С	74.1ª	45.2 <sup>bX</sup>	30.4 <sup>cX</sup>	22.2 <sup>cX</sup>	7.7 <sup>cX</sup>	2.49	***	***	***
of DM)	В	71.2ª	51.3 <sup>bX</sup>	14.9 <sup>cY</sup>	15.6 <sup>cX</sup>	9.1 <sup>dX</sup>				
	СН	79.3ª	76.1 <sup>aY</sup>	86.9 <sup>aZ</sup>	85.6 <sup>aY</sup>	49.5 <sup>bY</sup>				

 Table 1
 Characteristics of maize silage at 1, 3, 5, 10 and 90 days of fermentation

T = significance of treatment, D = day of fermentation, T x D = interaction between T and D; n.s. not significant; \*\* P<0.01, \*\*\* P<0.001; <sup>a, b, c, d</sup> Values within a row with different superscripts differ at P<0.05

 $^{X, Y, Z}$  Values within a column with different superscripts differ at P<0.05

#### Conclusion

Both treatments decreased pH of silage at day 1 of fermentation. All silages were well fermented with pH < 4.0 after 10 days of fermentation. Addition of bacterial inoculant increased the concentration of lactic acid and improved the lactic acid : acetic acid ratio at day 90 of fermentation. The chemical additive containing formic acid, propionic acid, ammonium formate, and benzoic acid did not affect the concentration of lactic acid and increased the concentration of acetic acid compared to untreated silage at day 90 of fermentation.

#### Acknowledgment

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## The Effect of Sil-All Maize+ FVA on Pastone Silage

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### Introduction

High mositure corn grain (HMCG) is harvested globally as a high energy content feed for dairy, beef and pigs. Pastone is harvested significantly earlier than HMCG at a dry matter in the region of 55% which gives a high energy silage on a reduced plant growth cycle. Processing methodology of grain to make pastone and HMCG is comparable.

Pastone is prone to microbial issues through fermentation and feed out to a greater extent than HMCG due to its higher water activity making effective treatment of pastone at ensiling critical.

#### Material and methods

Pastone was produced at 54% dry matter (46% mositure content) for ensiling into a concrete bunker. 80Kg of untreated pastone was separated and hommogenised on clean plastic. Pastone was split into 40Kg aliquots. Untreated pastone was sprayed with 400ml of tap water using a hand held sprayer (Control). Treated pastone was sprayed with 400ml of Sil-All Maize+ FVA (diluted to apply the commercial application rate of 250,000cfu/g forage). Mixing off the pastone was undertaken both during and after treatment.

3 bags of treated (T = treated) and 3 bags of untreated pastone (C = Control) were produced with coloured silks to identify treatment. Between 7 and 8 Kg of pastone was ensiled in 'onion bags' (weight recorded) and positioned centrally in the bunker across a horizontal plane at a height of circa 1m. Bags were sandwiched with 1m of untreated pastone above and below. Bags were subject to the compaction of the bunker as the bunker was filled, with conditions typical of the bunker (compaction, standard sheeting with single layer white on black plastic, oxygen penetration etc). On de-siling bags were weighed and individually homogenized and sampled for microbial, nutritive and digestibility profile.

#### **Results and discussion**

Ensiled bags were reached 9 weeks post ensiling and were dug out of the pastone bunker at the same time. Bags were weighed post ensiling and sampled for microbial and fermentation profile.

Bags had variously lost weight and gained weight through storage. This can only be attributed to bags variously adsorbing effluent. Extrapolating this to the fermentation profile of the bags it is clear that bags fermentation profile will be diluted by control effluent entering the bags reducing the visible impact of treatment on the fermentation profile.

Treated and untreated pastone was well fermented with no difference in the spoilage microbial profile (Table 1). Very low yeast and mould counts coupled with the observed stability of the pastone during feeding out negated the need to undertake aerobic stability assessments.

	Microbial Cou	unt (colony formi	ng units per g forage)
	Yeast	Mould	Clostridia
C1	<100	<100	2300
T1	<100	8100	400
C2	<100	<100	4
T2	<100	<100	3

Table 1Pastone	Microbial	profile
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Treatment with Sil-All Maize+ typically resulted in higher levels of lactic acid and lower levels of acetic acid, indicating a more efficient homolactic fermentation profile. The maximum LA:AA ratio was shown with the C3 and T3 paired bags with the SII-All Maize+ treated pastone having a significantly lower level of acetic acid and a ratio of LA:AA in excess of 50:1 compared with the control sample ratio of 7.3:1. (Table 2). All silages were well fermented with no butyric acid, low levels of ammonia and low, stable pH.

	Lactic Acid	Acetic Acid	LA:AA	Butyric Acid	Ammonia	рН
C1	3.19	0.86	3.7	0	1.7	3.9
T1	3.67	0.36	10.2	0	2.4	3.8
C2	2.9	1.45	2.0	0	2	3,8
T2	2.68	1.65	1.6	0	2	3.7
C3	3.65	0.5	7.3	0	2	3.8
T3	4.55	0.09	50.6	0	2.3	3.9

**Table 2** Fermentation Profile (%Dry Matter)

Table 3 presents the relative NDF-Digestbility (NDF-D) of treated and control pastone. Digestibility of forage reduces from time of cutting until time of reaching a stable pH. Faster pH fall stops the drop in NDF-D more rapidly

	NDF-Digestibility %								
	12hr	24hr	30hr	48hr					
Control	30.15	44.99	56.48	73.64					
Sil-All Maize+	31.88	46.67	59.97	79.03					
Difference	+1.73%	+1.68%	+3.49%	+5.39%					

30hr NDF-D is most reflective of the impact on the dairy animal (being the average time that forage remains in the rumen of a high producing dairy animal).

Milk production has been show to increase by 0.231 of milk per cow per day for each 1% unit in NDF-D by Michigan State University (Kendall et al). Pastone only accounts for a proportion of the animal diet however all farms are required to maximise the recovery of feed value from the ensiled forage and treatment with Sil-All Maize+ better maintained NDF-D by 3.49% compared to untreated pastone, representing a further 0.8l milk per cow per day.

Combing the fermentation data and digestibility data it can be strongly suggested that the speed of fermentation was faster with Sil-All Maize+ FVA, driven through a homolactic fermentation (shown by the higher LA, lower AA and elevated LA:AA), resulting in a faster final pH and greater NDF-D (as analysed). The control pastone was more heterolactic in its fermentation profile (higher AA and lower LA:AA). This strongly suggests that more dry matter will also have been protected through inoculation with Sil-All Maize+.

#### Conclusion

Pastone can be made without the use of inoculant when good ensiling and desiling practises are adopted but the use of Sil-All Maize+ under 'on farm' ensiling and feed out conditions results in a more homolactic fermentation that likely preserves more of the ensiled dry matter, does result in a faster fermentation of the pastone and better maintained digestibility of the pastone that would equate to an additional 0.81 of milk per cow per day.

Keywords: pastone, fermentation, digestibility, milk production, Sil-All Maize+

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#### The Possibility of Wet Maize Corn Conservation with Chemical Additive

BACK

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#### Introduction

Maize (*Zea mays*) due to the comprehensive possibilities of use and to huge yield potential is of vital economic importance plants in the world. Due to the high demand for fodder, interest in the production of silage from corn grain as feed useful in feeding cattle and pigs increase. From an economic point of view, to acidify minimize production costs associated with the preparation of a grain, because it is substantially cheaper than by drying it. It is important, to emphasize the fact, that in the Polish conditions to the aggregating the grain which is appropriable to storage is impossible for climatic reasons - always have to be pre-preserve. For maintenance of grain corn may be used silage additives in order to improve ensiling, silage quality, reduce losses of organic ingredients and improve aerobic stability.

The aim of the study was to evaluate the effectiveness of the drying and grinding wet grain corn using a chemical preservative preparation.

#### **Material and Methods**

The material consisted of Carmen corn varieties, which was collected with water content to 331 g/kg. The grain was ensiled on a laboratory scale in gas-tight tubes (1.5 l). Half of the grain before ensiling was grinding. There were four variants of silage: whole corn grain without the addition of chemical, whole corn grain with a chemical additive (Agro-Sil Corn - basic components: lactic acid, formic acid and acetic acid) kibbled corn without the addition of chemical additive.

The basic chemical composition was determined by conventional methods (AOAC 2007), structural carbohydrates, ADF and NDF - by the method of Georing and Van Soest (1970), the contents of lactic acid, acetic acid and butyric - Lepper method (AOAC 2007).

We also performed a test aerobic stability of silage. From each tubes collected 200 g of silage for testing thermal stability under aeration. The samples were exposed to air for a period of 5 days (120 hours). The test was conducted by means of electronic, automatic thermometer LB-711. The temperature inside each silage samples was measured and recorded every hour.

In order to determine the loss of dry matter and protein in the time of maintenance of whole corn grain to the film tube (production conditions) at three points (top center and bottom left and right) during the filling six plastic bags was inserted, each with a weight of 1 kg (three maize and without the addition of three - plus). During the dialing grain from sleeve bags the silage was analyzed for their content of dry matter and protein, the difference in the amount of dry matter and protein before ensilage and after loss of weight was calculated and the percentage losses.

#### **Results and Discussion**

Content of dry matter was not different between ensiling grain and starting material (Tab. 1). In this study there was no effect of treatments (grinding and additive) on the content of dry matter and protein. Silage prepared without the addition of chemical compared to the silage with the chemical additive contained less crude fat. The highest content of crude ash was observed in silage grain kibbled without the addition of a chemical preservative. In the silages made with the addition of chemical compounds reported a higher concentration of nitrogen-free extract (Tab. 2). Higher crude fiber content was in silage grain kibbled. In addition, the silage compared to silage with whole grain contained a lower fraction of NDF. Lower pH value was recorded in the silage grain kibbled before ensilage (Tab. 3). Silage prepared with a chemical additive was characterized by a higher content of lactic acid and acetic acid. Clearly less nitrogen in ammonia nitrogen was observed in silage kibbled. All silages were aerobic stable during the 5-day period of aeration. During the analyses the lower losses of dry matter and protein in silage made with chemical additive were observed (Fig. 1).

#### Conclusions

In summary, the chemical preservative Agro-Sil Corn was effective ensilage additive which was supporting the maintenance process of wet corn grain, particularly after the initial grinding.

Key words: maize corn, chemical additive, chemical composition, quality, aerobic stability

 Table 1
 Dry matter and non-carbohydrates content in ensiled maize corn

	Nutrients								
Kind of silage	Dry matter (g·kg <sup>-1</sup> )	Crude protein (g·kg <sup>-1</sup> DM)	Crude fat (g·kg <sup>-1</sup> DM)	Crude ash (g·kg <sup>-1</sup> DM)					
W-0	658,7 ±7,7	98,3 ±2,7	43,3 <sup>ab</sup> ±1,4	14,7 <sup>ab</sup> ±0,2					
W-I	668,5 ±10,4	95,6±1,7	42,8 <sup>ab</sup> ±1,2	13,9ª ±0,5					
K-0	674,6 ±4,4	100,2 ±0,9	48,9ª ±2,2	16,0 <sup>b</sup> ±0,4					
K-I	686,0±6,8	96,4±1,7	39,4 <sup>b</sup> ±5,5	15,2 <sup>ab</sup> ±0,2					

Different letters document statistical differences in each column:  $a,b - P \leq 0,05$ W-0 whole grain without additive, W-I whole grain with additive, K-0 kibbled grain without additive, K-I- kibbled grain with additive 
 Table 2
 Carbohydrates content in ensiled maize corn

	Nutrients								
Kind of silage	N-free extract (g·kg <sup>-1</sup> DM)	Crude fibre (g·kg <sup>-1</sup> DM)	NDF (g·kg <sup>-1</sup> DM)	ADF (g·kg <sup>-1</sup> DM)					
W-0	815,9ª ±5,9	27,9ª ±4,5	226,5 <sup>A</sup> ±6,4	22,1 ±2,7					
W-I	827,6 <sup>b</sup> ±1,6	20,1 <sup>b</sup> ±0,8	278,6 <sup>A</sup> ±9,3	24,7 ±4,5					
K-0	809,0ª ±0,5	26,0 <sup>ab</sup> ±0,8	113,9 <sup>B</sup> ±2,9	26,1 ±2,1					
K-I	822,7 <sup>b</sup> ±4,2	26,3 <sup>ab</sup> ±1,7	126,5 <sup>B</sup> ±6,7	23,7 ±0,8					

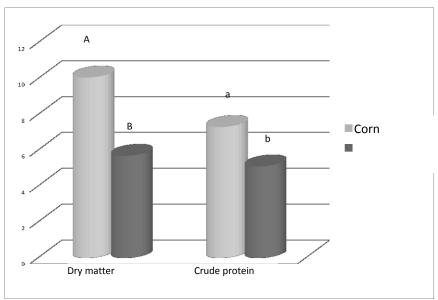
Different letters document statistical differences in each column: a,b – P≤0,05, A,B – P≤0,01

W-0 whole grain without additive, W-I whole grain with additive, K-0 kibbled grain without additive, K-I- kibbled grain with additive 
 Table 3
 Quality of ensiled maize corn

	Parameters									
Kind of silage	рН	Acetic acid (g·kg <sup>-1</sup> DM)	Lactic acid (g·kg <sup>-1</sup> DM)	Buttyric acid (g·kg <sup>-1</sup> DM)	N-NH3 %N <sub>total</sub>					
W-0	$4,41^{a} \pm 0,04$	32,9ª ±4,1	$14,9^{a}\pm 2,9$	0,4 ±0,4	30,03 <sup>a</sup> ±8,62					
W-I	4,30ª ±0,12	54,8 <sup>ab</sup> ±6,5	23,3 <sup>ab</sup> ±3,5	$0,0 \pm 0,0$	21,19 <sup>ab</sup> ±5,54					
K-0	$4,08^{b}\pm0,02$	66,0 <sup>b</sup> ±3,5	24,2 <sup>ab</sup> ±9,1	$0,0 \pm 0,0$	$21,90^{ab}\pm 1,34$					
K-I	3,95 <sup>b</sup> ±0,27	76,5 <sup>b</sup> ±4,3	35,1 <sup>b</sup> ±7,9	$0,0 \pm 0,0$	14,59 <sup>b</sup> ±3,18					

Different letters document statistical differences in each column: a,b - P≤0,05

W-0 whole grain without additive, W-I whole grain with additive, K-0 kibbled grain without additive, K-I- kibbled grain with additive



Different letters document statistical differences: a,b-P≤0,05, A,B-P≤0,01 Figure 1 Dry matter and crude protein losses during ensiling process of whole maize grain (%)

## A Meta-Analysis: Effect of Lime on the Chemical Composition, Fermentation, and Aerobic Stability of Sugarcane Silages in Brazil

BACK

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#### Summary

Our objective was to undertake a meta-analysis to investigate the impact of lime on the fermentation and aerobic stability of sugarcane silages in Brazil. The database was compiled by 77 treatments from 27 studies that assessed sugarcane silage treated with lime. For each gram of lime added ethanol concentration and dry matter (DM) losses decreased by 4.27 and 6.24 g/kg of DM, respectively. Neutral detergent fiber content was linearly reduced by addition of lime leading an increase on the *in vitro* DM digestibility (13.3 g/kg DM for each gram of lime added). Aerobic stability of silages increased by 4.88 h/g of lime added. Lime is a helpful additive to improve fermentation and aerobic stability of sugarcane silages.

#### Introduction

In Brazil, 21.5% of dairy farms use sugarcane silage as a feedstuff (Bernardes and Rêgo, 2014). Sugarcane has many desirable ensiling traits including a suitable dry matter (DM) content at ensiling even during the dry season, a high concentration of water-soluble carbohydrates (WSC) and low buffering capacity, which ensures a rapid pH decline (Freitas et al., 2006). However, sugarcane silages may display DM losses up to 30% (Pedroso et al., 2005) due to the production of high amounts of ethanol (Kung and Stanley, 1982). Thus, lime (calcium oxide) has been assessed to avoid the alcoholic fermentation in sugarcane silage. Lime is an alkaline additive that inhibits the growth of yeasts and reduces DM losses (Cavali et al., 2010) while, increasing the hydrolysis of cell-wall carbohydrates (Daniel et al., 2013a). A great number of studies used lime to improve the fermentation and aerobic stability of sugarcane silages, but a meta-analytical investigation of these data has not been undertaken. Consequently, there is no consensus regarding the effectiveness of these additives. Therefore, our objective was to undertake a meta-analysis to investigate the impact of lime on the fermentation and aerobic stability of sugarcane silages in Brazil.

#### **Material and Methods**

The database to examine the impact of lime on the ensiling of sugarcane silage consisted of data collected from 27 studies, representing a total of 77 treatments. The rate of lime applied to freshly harvested sugarcane ranged from 0 to 20 g/kg. A minimum of two studies per each treatment was the prerequisite for keeping the dependent variables in the final database. Experiments were excluded if silages were ensiled for less than 30 d. The chemical composition of silages was reported on a DM basis and yeast population was reported on a log<sub>10</sub> basis. Some calculates were made when data were lacking in the publications. Hemicellulose content was calculated as neutral detergent fiber (NDF) minus acid detergent fiber (ADF), whereas cellulose content was calculated as ADF minus lignin. Aerobic stability was measured in the same manner for all studies, and was defined as the number of hours required for the temperature of aerobically exposed silage to increase 2°C above ambient temperature.

Data on lime were analyzed as mixed models regressing the variables against the levels of lime application using the MIXED procedure of SAS (v. 9.4 SAS Institute Inc., Cary, NC). When significant, the effect of level of lime applied was tested for linear, quadratic and cubic effects by orthogonal polynomial contrasts using the CONTRAST statement of MIXED procedure. The study effect was also included in the model through the RANDOM statement (St-Pierre, 2001). Differences were declared significant at  $P \le 0.05$  and trends discussed at  $0.05 > P \le 0.10$ .

#### **Results and Discussion**

Effects of addition of lime on chemical composition, fermentation, and aerobic stability of sugarcane silage are given in Table 1. Lime did not affect the concentrations of WSC (overall mean 79.8 g/kg of DM), acetic acid (overall mean 30.8 g/kg of DM), ammonia-N (overall mean 69.4 g/kg of total nitrogen), and the population of yeasts (overall mean 4.68 cfu/g of fresh silage). Otherwise, addition of lime linearly increased the silage pH due its alkaline nature. The addition of lime also linearly increased the concentration of lactic acid (P < 0.01). There was a linear reduction (P < 0.05) in the concentration of propionic acid, whereas butyric acid quadratically increased (P < 0.01). For each gram of lime added on sugarcane silage, the concentration of ethanol decreased (P < 0.01) 4.27 g/kg of DM, while gas and DM losses decreased (P < 0.01) by 6.2 g/kg of DM. Ethanol reduction may be associated to changes on fermentative pathway (homolactic against alcoholic fermentation), once the production of lactic acid increased and lime may change the osmotic potential of silage, which is a mechanism to inhibit spoilage microorganisms. Aerobic stability of sugarcane silage increased by 4.88 h/g of lime added. Although data about the yeasts growth during aerobic exposure are lacking in Brazilian literature, aerobic stability of silages likely increased due to a reduction in yeast population caused by the decreased water activity from addition of lime.

Despite of chemical composition, lime is an inorganic material rich mainly in calcium oxide, and as expected, its addition linearly increased (P < 0.01) DM and ash contents of sugarcane silage. Crude protein (CP) content was linearly reduced (P < 0.01) while addition of lime increased, likely because the dilution effect caused by the additive. Lime reduced (P < 0.01) NDF, ADF, hemicellulose, cellulose and lignin contents. As a consequence, *in vitro* DM digestibility

enhanced (P < 0.01) by 13.3 g/kg of DM for each gram of lime added on fresh sugarcane prior to ensiling. Although NDF content linearly decreased by addition of lime in our study, the net hydrolysis of fiber was not observed once the hemicellulose content expressed on NDF basis was not changed. Moreover, the reduction of lignin content by lime may be explained by dissolving of a part of lignin from alkaline treatment (Van Soest, 1987).

Table 1 - Effect of addition of lime (g/kg of fresh sugarcane) on the fermentation and chemical composition (data are
given in g/kg of DM, unless otherwise stated) of sugarcane silage as estimated by a mixed model regression analysis <sup>1</sup> .

Item	Intercent	SEM <sup>2</sup>	Slope				<i>P-</i> -	<i>P</i> -value	
	Intercept	SEIVI	Linear	SEM	Quadratic	SEM	Linear	Quadratic	
Fermentative profile									
Gas loss	174.6	18.75	-6.23	1.29			0.0012	0.2504	
DM losses	232.1	26.18	-6.24	1.54			0.0055	0.1661	
Lactic acid	35.8	10.71	3.27	0.57			0.0013	0.8524	
Propionic acid	15.03	6.48	-0.88	0.30			0.0262	0.8515	
Butyric acid	2.7	2.29	-0.88	0.70	0.17	0.08	0.0021	0.0452	
Ethanol	94.2	33.74	-4.27	0.81			0.0073	0.5023	
pН	3.49	0.07	0.076	0.01			< 0.0001	0.7762	
Aerobic stability, h	74.6	23.54	4.88	1.81			0.0364	0.3353	
Chemical composition									
DM, g/kg as fed	246.3	9.49	2.19	0.41			0.0003	0.2801	
Ash	48.6	6.66	3.92	0.43			< 0.0001	0.6348	
СР	33.7	1.58	-0.39	0.07			< 0.0001	0.1081	
NDF	654.2	14.89	-8.35	0.82			< 0.0001	0.1941	
ADF	421.5	12.32	-4.85	0.78			< 0.0001	0.7057	
Hemicellulose	227.5	12.52	-3.71	0.72			0.0002	0.5092	
Cellulose	333.5	21.84	-4.08	0.99			0.0018	0.3092	
Lignin	76.6	3.35	-0.91	0.24			0.0020	0.5485	
IVDMD <sup>3</sup>	516.2	27.73	13.33	1.72			< 0.0001	0.3652	

<sup>1</sup>Only significant regressions are shown (P < 0.05). <sup>2</sup>Standard error of the mean. <sup>3</sup>In vitro dry matter digestibility.

#### Conclusions

Lime is a suitable additive for sugarcane ensiling because reduce DM losses during fermentation and improve aerobic stability of silages, along with additional gain on silage digestibility.

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## A Meta-Analysis: Effect of Heterofermentative Inoculants Applied at Different Dry Matter Contents on the Fermentation Patterns and Aerobic Stability of Sugarcane Silages

BACK

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#### Summary

A database compiled with 107 treatment means from 29 studies that assessed sugarcane silage treated with *Lactobacillus buchneri*, *L. brevis*, *L. kefiri* and *L. hilgardii* at different dry matter (DM) contents was used to evaluate the impact of heterofermentative inoculants on the fermentation and aerobic stability of sugarcane silages in Brazil. Treatments were classified in a 2 (two DM contents: moderately low DM (ranged from 250 to 295 g DM/kg as fed) and normal DM (ranged from 300 to 370 g DM/kg as fed))  $\times$  2 (with or without inoculant) factorial design. In both DM contents, inoculation increased (P < 0.01) acetic acid and reduced (P < 0.05) ethanol concentration in sugarcane silages. Inoculation increased (P < 0.05) the aerobic stability of sugarcane silages compared to untreated silages. There were no interactions between heterofermentative inoculants and DM content at which the sugarcane was ensiled.

#### Introduction

Silage inoculants composed of heterofermentative lactic-acid bacteria (LAB) have been added to sugarcane at ensiling with the goal to reduce dry matter (DM) losses and improve aerobic stability of silages. For example, *Lactobacillus buchneri* enhances the aerobic stability in a wide variety of silages (Kleinschmit and Kung, 2006) due its capacity to convert lactic acid into acetic acid and 1,2-propanediol (Oude Elferink et al., 2001). Acetic acid has an antagonistic effect on the growth of yeasts (Moon, 1983), and reduces the production of ethanol, along with improvements on the aerobic stability of sugarcane silages (Ávila et al., 2012). The DM at which forage crops are ensiled also has profound effects on silage fermentation because a lack of moisture in dry forages restricts the overall fermentation process (Hu et al., 2009). However, the effect of DM content at which sugarcane is ensiled on the effect of silage inoculants is unknown. Therefore, our objective was to evaluate through a meta-analysis the impact of heterofermentative inoculants applied at the different DM contents on the fermentation patterns and aerobic stability of sugarcane silages in Brazil.

#### **Material and Methods**

The database compiled for this study consisted of 107 treatment means from 29 studies (peer-reviewed articles and two theses) that assessed sugarcane silage treated with heterofermentative inoculants either alone or combined. We included L. buchneri, L. brevis, L. kefiri and L. hilgardii in the same database because the data in this review indicated that sugarcane silages treated with these bacteria had similar fermentation patterns. The treatments considered in this study were classified into four categories in a 2 (sugarcane ensiled at the moderately low DM and normal DM content) × 2 (sugarcane ensiled with (inoculated) or without silage inoculant (untreated)) factorial arrangement. The fresh sugarcane classified into the category moderately low DM ranged from 250 to 295 g DM/kg as fed, whereas the normal DM category ranged from 300 to 370 g DM/kg as fed, respectively. All experiments stored the silos between 30 to 140 d. The aerobic stability was measured in the same manner for all studies, and was defined as the number of hours required for the temperature of sugarcane silages increase 2°C above the ambient temperature after exposure to air. A minimum of two studies per each treatment was the prerequisite for keeping the dependent variables in the final database. All data were analyzed as a mixed model in a  $2 \times 2$  factorial arrangement using the MIXED procedure of SAS (v. 9.4 SAS Institute Inc., Cary, NC). Differences between means were determined using the LSMEANS statement of the P-DIFF option, which differentiates means based on Fisher's F-protected at least significant difference test. The study effect was included in the model through the RANDOM statement (St-Pierre, 2001). Differences were declared significant at  $P \le 0.05$  and trends discussed at  $0.05 > P \le 0.10$ .

#### **Results and Discussion**

All comparisons of means between sugarcane silages untreated and inoculated at the different DM contents for chemical composition, fermentation and aerobic stability are in Table 1. Inoculation increased (P < 0.01) the concentration of acetic acid by 38.2% in sugarcane silages. Inoculation reduced (P < 0.05) by 22.1% the concentration of ethanol in sugarcane silages. Untreated silages were stable for 25.6 h but inoculation increased (P < 0.05) improved aerobic stability by an additional 7.5 h. The heterofermentative LAB investigated in this meta-analysis were used in order to increase the concentration of acetic acid by anaerobic conversion of lactic acid when the primary fermentation ended (Oude Elferink et al., 2001). The antifungal effect (Moon, 1983) of acetic acid explains the reduction in yeasts counts and ethanol concentration, and the enhanced aerobic stability of sugarcane silages by using the inoculants. The crude protein (CP) tended (P < 0.10) to be reduced by 22.5% in sugarcane ensiled at the normal DM content compared to moderately low DM. The untreated silage produced at the normal DM content than untreated silage at the moderately low DM (more 32 g/kg of DM). Inoculation also increased (P < 0.05) the *in vitro* DM digestibility (IVDMD) coefficients of sugarcane silages by 4.3%.

Item	Moderate	ly low DM	Norm	al DM	<b>CEM</b>	<i>P</i> -value			
Item	Untreated	Inoculated	Untreated	Inoculated	SEM	DM	Ι	$\mathrm{DM}  imes \mathrm{I}$	
Fermentation profile									
Effluent, kg/t of FM1	105.7	87.4	40.0	39.1	18.78	0.0651	0.1138	0.1472	
Gas loss	286.7	209.7	225.6	214.9	55.65	0.7380	0.1481	0.2689	
DM loss	270.0	255.8	237.0	215.9	36.58	0.4739	0.2456	0.8192	
$WSC^2$	37.6	36.8	82.8	79.3	18.18	0.0973	0.3860	0.5813	
Lactic acid	34.9	32.7	28.4	31.2	6.44	0.6565	0.8651	0.1769	
Acetic acid	25.9	34.5	27.2	38.9	5.45	0.6994	0.0007	0.5702	
Lactic acid: acetic acid	1.73	1.21	1.46	0.98	0.45	0.6689	0.0564	0.9290	
Propionic acid	5.09	5.48	2.07	1.58	1.58	0.1036	0.9572	0.6225	
Butyric acid	0.62	1.95	0.47	0.38	0.80	0.4137	0.2519	0.1946	
Ethanol	121.2	102.4	81.6	55.6	21.32	0.1341	0.0414	0.7399	
pН	3.50	3.49	3.59	3.48	0.06	0.6565	0.0020	0.0094	
Ammonia-N, g/kg of TN <sup>3</sup>	63.9	59.2	59.0	62.0	22.65	0.9741	0.8876	0.4984	
LAB <sup>4</sup> , cfu/g	8.05	8.13	4.20	5.39	0.91	0.0118	0.1251	0.1784	
Yeasts, cfu/g	6.03	4.04	5.38	5.26	0.52	0.6334	0.0271	0.0488	
Aerobic stability, h	13.5	18.7	37.7	47.5	4.20	< 0.0001	0.0480	0.5209	
Chemical composition									
DM, g/kg as fed	234.8	241.5	269.1	284.0	11.01	0.0160	0.0004	0.1552	
Ash	52.8	54.2	33.5	33.4	8.20	0.1156	0.5740	0.5188	
СР	41.3	39.6	31.4	31.3	3.34	0.0653	0.3069	0.3842	
NDF	623.8	633.1	655.8	639.4	19.69	0.4863	0.6313	0.0870	
ADF	421.2	422.4	430.3	414.9	20.15	0.9775	0.2160	0.1484	
Lignin	-	-	85.4	88.6	12.46	-	0.4483	-	
IVDMD <sup>5</sup>	494.3	506.7	477.2	506.7	36.23	0.8716	0.0419	0.3969	

 Table 1
 Effects of DM content and silage inoculant (I) on the chemical composition, fermentation patterns and aerobic stability of sugarcane silages (data are given in g/kg of DM, unless otherwise stated).

<sup>1</sup>Fresh matter. <sup>2</sup>Water-soluble carbohydrates. <sup>3</sup>Total nitrogen. <sup>4</sup>Lactic acid bacteria. <sup>5</sup>In vitro dry matter digestibility.

#### Conclusions

The data in this meta-analysis suggest that the effects of heterofermentative LAB on fermentation and chemical composition are similar for sugarcane ensiled at the moderately low and normal DM content.

#### Acknowledgments

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## SESSION 3 – CONSERVED FEEDS IN NUTRITION OF ANIMALS

## **Oral presentations**

<u>Selection and nutritive value of different hybrids of maize silage</u> Jambor, V., Vosynková, B., Loučka, R.

<u>Risks of feeding silages with low physically effective neutral detergent fibre</u> Loučka, R., Homolka, P., Jančík, F., Kubelková, P., Tyrolová, Y., Výborná, A., Illek, J., Jambor, V., Takahashi, J., Okamoto, M.

<u>A new approach related to digestible and undigestible NDF content of different forages</u> Orosz, S., Davies, D. R., Szemethy, D., Szabó, J., Podmaniczky, T.

## **Poster presentations**

Mineral content (Ca, P, K, Na, Mg, S, Mn, Zn, Cu) of the most important forages in Hungary (2013-2015) Orosz, S., Simon, Á., Szabó, J., Podmaniczky, T. Davies, D. R., Szemethy, D., Könyves, L.

Corn silage processing score in Hungary between 2013-2015 Orosz, S., Davies, D. R., Szemethy, D., Szabó, J., Podmaniczky, T.

Effect of storage length on the maize starch degradability Férard, A., Peyrat, Uijttewaal, A., Meslier, E.

Effect of crimped maize grain ensiled with high moisture grains of transgenic Bt maize in fattening bulls

Chrenková, M., Pomikalová, S., Chrastinová, Ľ., Polačiková, M., Formelová, Z., Rajský, M., Mlyneková, Z.

<u>The level of rumen fermentation in heifers during transition from winter to pasture feeding</u> Žitňan R.

## Selection and Nutritive Value of Different Hybrids of Maize Silage

**BACK** 

#### JAMBOR, V.<sup>1</sup>, VOSYNKOVÁ, B.<sup>1</sup>, LOUČKA, R.<sup>2</sup>

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#### Introduction

Numerous studies have evaluated the impact of advancing maturity, mechanical processing, application of additives on nutritive value and digestibility of maize silage (Andrae et al., 2001, Thomas et al., 2001, Jensen et al., 2005, Filya and Sucu, 2010, Hetta et al., 2012). Loučka and Jambor (2009) published differences in the content of selected nutrients and digestibility of whole plants in five hybrids with the same earliness (medium early FAO 300). Cherney et al. (2004) monitored fermentation characteristics of maize hybrids, selected to vary in dry matter (DM), crude protein (CP) and crude fibre digestibility (CFD), whereas Russell et al. (1992) demonstrated that the differences in the nutritional value of silage were not influenced only by individual hybrids, but also by methods of tillage and agro-technical measures.

The aim of our study was to compare the differences in the content of organic matter degradability (OMD) and neutral detergent fibre degradability (NDFD) for two maize hybrids grown for silage in farm conditions.

#### **Material and Methods**

At condition of agriculture cooperative Zašová (VZOD) we planted two hybrids of maize (Kairo and Ronaldinio) for testing of nutritive value and quality. Maize (planted on 15th May) was harvested at silage maturity on 25th September by the same harvestor at the same day and pressed into a big plastic bag with the capacity of 200 tone of fresh matter (FM) for each plastic bag. On January next year we take away by hand sampler 5 samples from each plastic bag. Samples were taken according to the methods in Commission Regulation (EC) No 152/2009 (2009). Silage samples were analysed for fermentation quality (pH, lactic, acetic and butyric acid) using an IONOSEP 2001 analyser (RECMAN - laboratory systems, Ostrava, Czech Republic) according to Kvasnička (2000). Dry matter (DM) was obtained from drying of silage at 60°C for 24 hours. Dried material was subsequently milled to pass through a one-millimetre sieve for laboratory analyses. NDF was assayed with a heat stable amylase and expressed inclusive of residual ash (aNDF) according to Mertens (2002). DM, ash, crude fibre (CF), and CP (N x 6.25) were determined as described by AOAC (2005). OM was calculated as DM minus ash. Degradability of OM (OMD) and NDF (NDFD) was measured by *in sacco* method in the fistule rumen of dairy cows (McDonald, 1981).

#### **Results and Discussion**

In the table 1 can see results of nutritive value of maize silage in plastic bags. The average values of DM, CP, CF and aNDF in both silages had a minimum difference.

Sample	DM	СР	CF	aNDF	Starch	OMD	NDFD	NEL	NEL tab.
1	28.4	8.9	22.0	44.3	27.5	71.7	50.7	6.32	6.79
2	28.9	8.7	21.2	43.9	27.6	73.5	54.6	6.45	6.80
3	29.8	8.7	21.4	44.8	27.2	75.0	56.4	6.49	6.81
4	28.9	9.0	20.1	43.7	26.7	73.2	51.3	6.40	6.81
5	30.4	8.6	21.1	44.0	31.1	74.0	55.3	6.47	6.81
Avg	29.3	8.8	21.2	44.1	28.0	73.4	53.6	6.43	6.80
SD	0.82	0.16	0.69	0.43	1.76	1.19	2.52	0.07	0.01

 Table 1
 Nutrition value of maize silage, prepared from hybrid Kairo

Sample	DM	СР	CF	aNDF	Starch	OMD	NDFD	NEL	NEL tab.
1	34.1	9.2	20	43.9	34.8	61.7	38.1	6.1	6.81
2	30.8	9.3	21.9	45.3	31.7	59.9	35.6	5.95	6.78
3	33.3	9.4	20.7	43.9	34.1	63.4	37.2	6.04	6.79
4	31.7	9.2	22.3	46.4	31.5	62.5	45.8	6.19	6.78
5	31.2	9.2	19.5	48.7	31.5	61.7	44.6	6.27	6.81
Avg	32.2	9.3	20.9	45.6	32.72	61.8	40.3	6.11	6.79
SD	1.43	0.1	1.20	2.01	1.60	1.30	4.62	0.13	0.02

Content of starch is significantly higher (P<0.05) with hybrid Ronaldinio (32.2 % DM) and lower with hybrid Kairo (29.28 % DM). NDFD was significantly higher (P<0.05) with hybrid Kairo (53.65%) compared to hybrid Ronaldinio (40.27%). Significantly higher OMD was with Kairo than hybrid Ronaldinio.

This is reflected in the value of calculating the concentration of power in both silages netto energy of lactation (NEL), see Table 3. In calculating the energy value of maize silage in agricultural laboratories commonly used CFD of table value of 69%. If these tables digestibility of CF was used in both silages resulting concentration of power NEL had minimal differences. These minor differences are due to the fact that the content of organic nutrients in both silages was minimal. In our case, where we have established laboratory values of NDFD, the difference was 13.38%, and the resultant value was significantly (P<0.05) higher 6.43 NEL MJ.kg<sup>-1</sup> DM. NEL of hybrid Kairo, compared with hybrid Ronaldinio, have had 6.11 MJ.kg<sup>-1</sup> DM.

Hybrid	DM	СР	CF	aNDF	Starch	OMD	NDFD	NEL	NEL tab.
Kairo	29.3	8.78	21.2	44.1	28.0ª	73.4 <sup>b</sup>	53.6 <sup>b</sup>	6.43 <sup>b</sup>	6.80
Ronaldinio	32.2	9.26	20.9	45.6	32.7 <sup>b</sup>	61.8 <sup>a</sup>	40.3 <sup>a</sup>	6.11 <sup>a</sup>	6.79

 Table 3
 Nutritive value of maize silage made from hybrid Kairo and Ronaldinio

<sup>a,b</sup> Means in the same column with different superscripts differ (P < 0.05).

#### Conclusion

The result of this experiment shows, that content of CP, CF, aNDF of maize silage of two silage hybrids is similar, but we determined that content of starch, OMD and NDFD was statistically different (P<0.05) at this farm experiment. We can calculate, if the maize silage from hybrid Kairo had a higher energy content (6.43 MJ.kg<sup>-1</sup> DM) than Ronaldinio (6.11 MJ.kg<sup>-1</sup> DM). Cows which have intake of maize silage (10 kg of DM) can produce from this maize silage 20.6 kg resp. 19.6 kg of milk per day. For maize silage from Kairo hybrid it is 1 kg of milk more than from maize silage from hybrid Ronaldinio. According Oba and Allen (1999) 1% NDFD increase intake of 0.25 kg of DM. Increasing of intake of maize silage increase the energy intake and it means also higher production of milk.

#### Dedication: MZe NAZV QJ1510391

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#### Risks of Feeding Silages with Low Physically Effective Neutral Detergent Fibre

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#### Introduction

The structural fibre given by the particle size has a significant effect not only on the fermentation processes of fodder preservation (the shorter and more eroded particles, the better the fermentation result) but also on the motor activity of the rumen. The physically effective fibre (peNDF) with particle sizes on a sieve of 8 and 19 mm was examined with the help of the Penn State Particle Separator (PSPS) in holstein dairy cows by Beauchemin and Yang (2005). They confirmed that the particle length is a reliable indicator of the rumination period, but it may not necessarily reduce the rumen acidosis. The review of prof. Forbes from the University of Leeds (Forbes, 2007) also gives evidence that the digestion of ruminants is not as simple as often presented. The rumen of dairy cows is a place of complicated digestion processes that increase the total acidity. If the acid contents in the rumen exceeds a specific limit (often set at pH 5.8), so called acidosis may occur and negatively affect the feed intake, the microbial metabolism, the production and quality of milk and the general health condition of the animals, including limb diseases (laminitis). The risk of acidosis can be reduced by higher proportion of physically efficient neutral detergent fibre (peNDF).

The rumen acidosis can be allegedly reduced by using ruminal mechanical stimulating (RMS) brushes (patent EP0609045A2) called Rumenfibe (RF). It is a brush used to stimulate the rumen motor activity.

The study was aimed at ascertaining the changes of pH in the rumen of dairy cows fed by total mix ratio (TMR) with higher proportion of physically effective neutral detergent fibre (peNDF) and in finding out whether the changes of pH would be influenced by application of RF in the rumen of the dairy cows.

#### **Materials and Methods**

The experiment included 26 Holstein dairy cows with average yield of 9335 kg milk; 4 of them had a eCOW probe (Devon, Ltd., Great Britain) in their rumen, implemented via esophagus, to perform pH measurements each 15 minutes. 2 dairy cows had a rumen cannula; they had also the pH continuously measured all along the experiment. In the middle of the first experiment period, one half of the dairy cows (RF group) got the artificial RMS device that was inserted into their rumen with the help of a special probe, another cows ((RF group) were without RMS.

The cows were stabled in an experimental barn with tensometric feeding troughs connected to a computer system with the respective software. So the consumption of fodders was continuously monitored all along the experiment, individually for each cow. The lying boxes had wood shavings on the floor.

The experiment took 19 weeks, including 2 preparatory weeks. 4 total mix ratios (TMRs) constituted the material for the cow experiment. The TMRs consisted of alfalfa silage, maize silage, corn cob mix silage, brewer's grains, mashed grains, energy and mineral agent and straw with particles 20 mm long. The main differences between the TMRs consisted in the proportion of brewer's grains (6 kg in TMR1 and TMR2, 10 kg in TMR3 and TMR4) and straw (0 kg in TMR1 and TMR4, 1 kg in TMR2 and TMR3).

The individual TMR components and the actual TMRs were analysed in a chemical laboratory with the help of standard methods (AOAC, 1995), 3x in each experiment period. The preparatory and the first experiment period ran with the same TMR. The particle length in the TMR was measured with the help of the Penn State Particle Separator (PSPS) with 8 and 19 mm sieves. The physically effective fibre (peNDF) was determined by multiplying the content of the neutrally detergent fibre in dry matter and the % residue on the 8 and 19 mm sieves (pef).

Additionally to the continuous measurement of pH in the cow rumen, the rumen liquid was taken in each period with the help of a probe, the amount and quality of the milk were measured every day, ethological monitoring took place in each period, and the health condition of the cows and their reproduction indicators were monitored all along the experiment. This article is focused on the evaluation of pH in the rumen.

The results were processed by the SAS program, with and without the regression method, and also by the Statistica 10 (StatSoft, USA), by the ANOVA procedure and by the subsequent POST-HOC Tukey test.

#### **Results and Discussion**

The percentage of chaff with particles over 8 mm (pef) of all TMRs amounted to 35%; the calculated peNDF (pef x NDF) of TMR1 amounted to 14.9, that of TMR2 to 15.1, and that of TMR3 to 15.5, which are values below the acidosis limit. The average pH of TMR1 was 6.0 (Standard Deviation 0.24), that of TMR2 was 6.02 (SD 0.22), that of TMR3

was 6.11 (SD 0.21) and that of TMR4 was 6.19 (SD 0.18). The proportion of time in which the pH values got below 5.8 was 22 % for TMR1, 19 % for TMR2, 33 % for TMR3, and 8.4 % for TMR4.

In each period, the pH was slightly higher in the RF group than in the control C group. For the whole time of the experiment, the difference between the RF and the C amounted to 0,14 pH. More importantly, during the whole time of the experiment, the pH of the RF group dropped under 5.8 on average only in 13 per cent, while that of the C group C in 25.5 %. The greatest difference between the periods and groups could be found when comparing the period with TMR3 and TMR4 when the feeding ratio contained about 10 kg brewers' grains (as against mere 6 kg in the preceding period). TMR3 contained additionally 1 kg straw. If there was no straw in the feeding TMR3 and 4, the difference between the RF and C groups was higher (than that of TMR3). But as at the time of feeding TMR3 and 4, the cows were in advanced lactation already and their yield was approximately the half, as compared to the beginning of the experiment, the pH dropped under 5.8 also in the control group only in 2.5 % of cases, and thus the effect of the RF on the fodder consumption and milk production was not significant.

The passage to the new TMR did not significantly influence the pH; rather on the contrary, after changing the fodder, the pH increased and in the second half of each period, it decreased.

The cows with the rumen cannula had an average pH of 6.09 in the C group, while 6.30 in the RF group. During the whole time of the experiment, the pH of the RF group dropped under 5.8 on average only in 3 per cent, while that of the C group C in 14.8 %. The percentage of decrease of pH under 5.8 was always higher in the control group than in the RF group. The difference between the RF and C group was 0.2 pH. The development of the pH changes in the monitored experiment periods was more balanced in the RF group than in the control group (SD 0.23 vs. 0.33).

The results of measurement of pH in the rumen of RF dairy cows cannot be compared with literature, as the scientific literature does not describe any experiment with measurement of pH in a rumen with RF. But there are results, described in patent EP0609045A2, that document the effect of RF on the indicators of yield and health of high-yield dairy cows in Japan. For dairy cows with lower yield, fed with a structural feeding ratio, the RF application into the rumen has probably no sense. Experiments performed in Thailand (Angthong et al., 2012) with the local beef race, Thai native cattle (*Bos indicus*) did not confirm the benefits of application of RF. Positive results were found in holstein bulls (Horiguchi and Takahashi, 2002, 2004; Christopherson et al., 2008).

#### Conclusion

The risks of feeding silage with low physically effective neutral detergent fibre can be partially mitigated by the use of the artificial device (patent EP0609045A2). The experiment with dairy cows with average yield of 9335 kg milk, which had the pH measuring eCOW probe in their rumen, found the following difference between the control C group without RMS and the group RF with RMS: 0.14 pH for the control group as against 0.21 pH for the cows with the rumen cannula. Even such pH improvement in the rumen may play an important role for acidosis. The pH of the dairy cows with eCOW dropped under 5.8 in the control C group in 25.5 %, while that of the RF group in 13 %. The pH of the dairy cows with the rumen cannula dropped under 5.8 in the control C group in 14.8 %, while that of the RF group in 3 %.

#### Dedication: MZe NAZV QJ1510391

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## A New Approach Related to Digestible and Undigestible NDF Content of Different Forages

**<u>BACK</u>** 

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#### Summary

Highly digestible fiber will rapidly ferment in the rumen allowing for more dry matter intake (Kammes et al, 2012). Undigested NDF is the functional fiber fraction that influences physical effectiveness, gut fill, and digestion/passage dynamics of forages. Grasses, legumes, and grain-forages such as corn silage behave differently in the rumen (Kammes et al, 2012). The authors determined dNDF<sub>48</sub> content of the most important silages and hays based on routine farm samples between 2013-2015 in Hungary (n= 4859). The uNDF<sub>48</sub> was applied as an indicator for rumen-undegradable fibre content for classification of the different forages.

#### Introduction

Fiber digestibility and indigestibility are critical factors when assessing forage quality and formulating diets. Nutritionists have focused primarily on measures of fiber digestibility, but recently the focus has included indigestible fiber (Grant et al, 2012). Mertens (2013) used term "undigested NDF (uNDF)". The recommended method requires 240 hours of in vitro fermentation using a Tilley-Terry system with modifications described by Raffrenato and Van Amburgh (2010). Grasses, legumes, and grain-forages such as corn silage behave differently in the rumen (Kammes et al, 2012). In order to understand the different behaviour in the rumen, the authors determined dNDF<sub>48</sub> content of the most important silages and hays based on routine farm samples between 2013-2015 in Hungary (n= 4859). The uNDF<sub>48</sub> (calculated as difference of NDF- and dNDF<sub>48</sub> content) applied as an indicator for undegradable fibre content in order to classify the different forages (the actual dataset based on 48 hours *in vitro* incubation time).

#### **Materials and Methods**

Routine farm samples were dried (70 °C, 8 hours) according to method EN ISO 6496:1993. Spectra were determined according to the guidelines of NEN-EN-ISO 12099 (Q-Interline Quant FT-NIR analyser). Crude nutrient content, fiber fractions, protein- and organic matter digestibility, NDF degradability were determined by NIR method (BLGG AgroXpertus database, Wageningen). Wet chemical and *in vitro* reference methods can be provided by the authors (NEN-ISO 5983-2, EG guideline L54 2009/152, NEN-ISO 6492 EG guideline L54 2009/152; NEN-EN-ISO 6865 EG guideline L54 2009/152; EG guideline L54 2009/152; NEN-EN-ISO 15914; AOAC Official Method 2002.04; NEN-EN-ISO 13906;Tilley and Terry, 1963).

#### **Results and Discussion**

Nutrient content and digestibility of routine maize silage with low starch (<300g/kg DM - 2013) and high starch (>350 g/kg DM -2014), lucerne silage/haylage samples, ryegrass silage (*Lolium multiflorum*), rye silage in boot-heading stage period (*Secale cereale*), whole crop cereal silage (winter barley, winter wheat, winter triticale, spring oat) in two different phenological stages (in boot-heading period and in milky-dough stage period), legume-cereal blend silage (barley and pea, wheat and pea, triticale and pea, oat and pea in different phenological stages), lucerne hay and grass (meadow) hay samples (n= 4859, 2013-2015, Hungary) in Table 1-2.

**Table 1** Nutrient content and digestibility of routine maize silage (2013, 2014, 2015) and lucerne silage/haylagesamples (n= 2869, 2013-2015, Hungary)

				Maiz	e silage				cerne lage		ilted cerne		cerne ylage
Year of harvest		2	013	2	014	2	015	2013	3-2015	2013	3-2015	201	3-2015
Sample number		7	/24	5	526	e	517	1	44	2	470		388
		mean	st. dev.	mean	st. dev.	mean	st. dev.						
Dry matter	g/kg	328	58	357	52	352	56	264	28	352	28	489	73
Crude protein	g/kg DM	75	11	73	8	75	10	190	25	195	27	194	24
Crude fiber	g/kg DM	216	28	168	22	195	28	301	49	280	40	274	39
Sugar	g/kg DM	22	15	17	6	21	11	25	12	23	14	34	18
Starch	g/kg DM	257	72	360	55	299	72						
NDF	g/kg DM	444	55	356	42	411	56	411	58	395	56	400	54
ADF	g/kg DM	250	31	198	25	229	32	327	45	308	35	310	35
ADL	g/kg DM	18	3	17	2	18	3	61	11	58	10	59	9
OMd	%.	73	2	75	2	74	2	64	5	67	5	67	4
NDFd <sub>48</sub>	%	54	4	50	4	53	4	43	9	41	9	40	8
dNDF48	g/kg DM	242	43	180	31	220	44	176	48	164	44	162	37
uNDF <sub>48</sub>	g/kg DM	202	2	176	18	191	20	235	44	231	46	238	41

**Table 2** Nutrient content and digestibility of routine ryegrass-, whole crop cereal-, legume-cereal blend silage and hay samples (n= 1990, 2013-2015, Hungary)

		•	grass	Rye	silage	Who	ole crop	cereal	silage	Legume-cereal			erne ay	(mea	rass adow) ay
			age ım spp)		cale ·eale	in boot – heading stage		milky-dough stage		silage all stages		middle quality		middle quality	
Year of harves	t	2013	8-2015	2013	8-2015	2013	8-2015	2013	8-2015	2013-	2015	2013	-2015	2013	-2015
Sample numbe	r	2	89	4	69	5	10	1	00	192		2	44	1	86
		mea	st.	mea	st.	mea	st.	mea	st.	mean	st.	mea	st.	mea	st.
		n	dev.	n	dev.	n	dev.	n	dev.	mean	dev.	n	dev.	n	dev.
Dry matter	g/kg	335	91	291	69	292	69	358	80	296	124	884	57	899	70
Crude protein	g/kg DM	141	32	134	31	133	31	90	24	142	32	190	34	93	23
Crude fiber	g/kg DM	272	39	306	43	306	42	273	42	286	37	308	51	337	28
Sugar	g/kg DM	67	52	40	34	39	34	56	38	40	28	48	18	71	30
Starch	g/kg DM			19	4	49	89	116	88	64	47				
NDF	g/kg DM	500	62	568	70	567	70	515	63	521	65	488	55	660	47
ADF	g/kg DM	304	44	338	50	337	49	310	51	326	44	343	45	369	31
ADL	g/kg DM	26	7	27	8	27	8	33	7	39	10	68	10	48	8
OMd	%.	73	5	71	5	71	5	65	3	67	5	63	6	56	6
NDFd <sub>48</sub>	%	65	8	66	7	66	7	48	6	54	9	39	7	39	10
dNDF <sub>48</sub>	g/kg DM	324	37	371	36	369	38	251	54	283	60	191	35	258	58
uNDF48	g/kg DM	176	53	197	64	198	63	264	38	238	58	298	58	402	79

#### Conclusions

According to the nutrient content and digestibility data, four classes were identified to describe the dNDF-uNDF content variations for the above mentioned silages and hays (Table 3). These classifications will help to provide more accurate formulation of forages in rations to optimise the proportion of uNDF to dNDF and therefore obtain the best balance between effective fibre to maintain a healthy rumen and highly digestible fibre to maximise nutrient availability for animal production.

**Table 3** Classification of the different silages and hays according to the degradable  $NDF_{48}$  and non-degradable  $NDF_{48}$  content (n= 4859, 2013-2015, Hungary)

High dNDF (>300 g/kg DM) in combination with	Low dNDF (<200 g/kg DM) in combination with
low uNDF (<200 g/kg DM) concentration generally	low uNDF (<200 g/kg DM) concentration generally
<ul> <li>high quality ryegrass silage with NDFd<sub>48</sub> higher than 70%</li> <li>whole crop cereal silages (in boot –heading stage) with NDFd<sub>48</sub> higher than 65%</li> <li>legume-cereal silage (in boot –heading stage of the cereal) with NDFd<sub>48</sub> higher than 65%</li> </ul>	<ul> <li>maize silage (dough stage, starch content of minimum 35% DM)</li> <li>legume-cereal silage (milky stage of the cereal) with NDFd<sub>48</sub> 40-50%</li> </ul>
High dNDF (>300 g/kg DM) in combination with	Low dNDF (<200 g/kg DM) in combination with
high uNDF (>200 g/kg DM) concentration generally	high uNDF (>200 g/kg DM) concentration generally
<ul> <li>high quality grass (meadow) hay with NDFd<sub>48</sub></li> <li>higher than 55%</li> </ul>	<ul><li>lucerne silage/haylage</li><li>lucerne hay</li></ul>

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## Mineral Content (Ca, P, K, Na, Mg, S, Mn, Zn, Cu) of the Most Important Forages in Hungary (2013-2015)

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BACK

#### Summary

Forage source can influence the site and extent of absorption, fecal output, and apparent digestibilities of macrominerals (Khorasani et al, 1997). The authors determined Ca, P, K, Na, Mg, S, Mn, Zn, Cu content of the most important silages (maize silage, lucerne silage and haylage, high sugar grass silage *-Lolium multiflorum*, rye silage (in boot-heading period), whole crop cereal silages (harvested in boot-heading period or in milky-dough stage), mixed silages (mainly cereal-pea based blends), lucerne hay and grass (meadow) hay based on routine farm samples (2014-2015) in Hungary. ICP-OES method was used for the determination.

#### Introduction

The authors determined Ca, P, K, Na, Mg, S, Mn, Zn, Cu content of the most important silages (maize silage, lucerne silage and haylage, high sugar grass silage (*Lolium multiflorum*), rye silage (harvested before heading), whole crop cereal silages (harvested before heading and in milky stage), mixed silages (mainly cereal based blends), lucerne hay and grass hay based on routine farm samples (2014-2015) in Hungary. ICP-OES method was applied for the determination.

#### **Materials and Methods**

Routine farm samples were dried (70 °C, 8 hours) according to method EN ISO 6496:1993. Inductively coupled plasma-optical emission spectrometry (ICP-OES) method was used to determine the different minerals from routine farm silage, haylage and hay samples during 2014-2015.

#### **Results and Discussion**

Mineral content (Ca, P, K, Na, Mg, S, Mn, Zn, Cu, Fe) of routine maize silage, lucerne silage/haylage samples, ryegrass silage (*Lolium multiflorum*), rye silage in boot-heading stage period (*Secale cereale*), whole crop cereal silage (winter barley, winter wheat, winter triticale, spring oat) in two different phenological stages (in boot-heading period and in milky-dough stage period), legume-cereal blend silage (barley and pea, wheat and pea, triticale and pea, oat and pea in different phenological stages), lucerne hay and grass (meadow) hay samples (n= 4859, 2013-2015, Hungary) in Table 1-2.

**Table 1** Mineral content of routine maize silage (2013, 2014, 2015) and lucerne silage/haylage samples (n= 2869, 2013-2015, Hungary)

				Maiz	e silage				cerne age		ilted erne		cerne vlage
Year	of harvest	2	013	2	014	2	015	2014	-2015	2014-2015		2014-2015 2014	
Samp	ole number	,	76	1	35	1	33	1	13		37		33
		mean	st. dev.	mean	st. dev.	mean	st. dev.						
Ca	g/kg DM	2,8	0,7	2,4	0,8	3,0	1,9	19	3	17	3	17	4
Р	g/kg DM	1,9	0,6	2,0	0,5	2,0	0,6	2,9	0,5	2,8	0,6	2,8	0,6
Ca/P		1,6	0,5	1,3	0,3	1,5	0,5	6,7	1,4	6,7	2,1	6,3	2,3
Κ	g/kg DM	11	4	9,2	3,0	12	3	25	9	25	7	23	7
Na	g/kg DM	0,2	0,2	0,1	0,3	0,3	0,9	0,6	0,4	0,8	0,5	0,8	0,9
Mg	g/kg DM	2,4	0,8	1,9	0,6	2,3	0,8	3,1	0,7	3,5	0,9	3,2	0,9
S	g/kg DM	1,1	0,3	1,0	0,2	1,2	0,3	2,4	0,4	2,6	0,6	2,5	0,6
Mn	mg/kg DM	41	7	33	13	28	12	70	10	63	23	<b>98</b>	165
Zn	mg/kg DM	29	14	33	48	31	13	29	6	30	8	31	11
Cu	mg/kg DM	4,8	0,7	5,1	1,4	4,7	1,4	10	2	10	2	10	2

**Table 2** Mineral content of routine ryegrass-, whole crop cereal-, legume-cereal blend silage and hay samples (n= 247, 2014-2015, Hungary)

		Ryg	rass	Rye s	silage	Who	le crop	cereal s	ilage	Legu cereal		Lucer	ne hay	Gras	s hay
		sila (Loliur		(Sec cere	cale eale)	boo heao sta	ding	milky- sta	0	all st	ages	mid qua			ldle llity
Year	of harvest	2014	-2015	2014	-2015	2014	-2015	2014	2015	2014	2015	2014-	2015	2014	-2015
Samp	le number	4	0	7	7	1	3	4	5	3	7	4	4	3	1
		mean	st. dev.	mean	st. dev.	mean	st. dev.	mean	st. dev.	mean	st. dev.	mean	st. dev.	mean	st. dev.
Ca	g/kg DM	5,2	1,1	4,2	2,0	4,5	2,1	3,6	1,5	5,4	1,9	14,2	3,0	4,9	2,0
Р	g/kg DM	2,8	0,7	3,0	0,8	2,9	0,8	2,3	0,4	2,6	0,7	2,6	0,6	2,0	0,8
Ca/P		2,0	0,6	1,5	0,8	1,6	0,9	1,6	0,7	2,1	0,7	5,8	2,1	2,8	1,5
Κ	g/kg DM	32	8	31	8	29	9	18	8	25	9	21	6	20	7
Na	g/kg DM	1,1	1,0	0,2	0,4	0,3	0,3	0,8	0,7	1,0	1,0	0,8	0,5	0,2	0,3
Mg	g/kg DM	1,7	0,3	1,7	0,8	1,8	0,7	1,9	0,5	2,1	0,8	3,1	0,8	1,8	0,6
S	g/kg DM	2,0	0,4	1,7	0,4	1,9	0,6	1,8	0,8	1,8	0,6	2,3	0,7	1,8	0,7
Mn	mg/kg DM	82	45	63	20	63	24			73	48	38	15	82	62
Zn	mg/kg DM	32	13	32	10	40	12			31	18	26	8	22	8
Cu	mg/kg DM	7,0	0,7	8,0	2,2	8,1	3,2			7,5	3,7	9,8	2,0	4,3	1,5

#### Conclusions

Mineral concentrations in forages can be highly variable with geographical, climatic and seasonal variation (Suttle, 2010) and so a knowledge of the mineral content of forages is important. This study showed maize silages were low in each mineral. There were no significant differences among the years. Lucerne silage contained a higher concentration of all minerals studied than the cereal silages, except Na. Typical high K-forage were the lucerne silages/haylages, the ryegrass silage, the rye silage, the whole crop silage harvested in boot-heading period and the legume-cereal silages. Therefore the above mentioned silages and haylages have to be limited in close-up TMR (lower than 1.5%DM K-level is required in the diet). Otherwise, lucerne hay contained high Mg-level, what may have positive effect in prevention of pospartum tetani (Charlton and Armstrong, 1989) or hypocalcemia (Schonewille *al.*, 1997) (higher than 0.35%DM level of Mg is recommended in close up diet). The whole crop silage harvested in boot-heading period contained more K, than cereal silage cut in milky-dough stage. High Mn-level was found in ryegrass silage, rye silage, legume-cereal silages and grass hay.

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### Corn Silage Processing Score in Hungary Between 2013-2015

**BACK** 

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#### Summary

The Corn Silage Processing Score (CSPS) was developed (USDA Forage Research Center) as a tool to define adequacy of kernel processing by forage harvesters. The CSPS is the percentage of starch passing through the coarse 4.75 mm screen (Ferreira and Mertens, 2005). Samples from different areas of Hungary collected by farmers during 2013 (n = 147), 2014 (n = 181) and 2015 (n = 243) were assessed. According to our results, 7% (2013), 13% (2014) and 13% (2015) of the analysed corn silages were in the optimal quality range. Inadequate corn processing can cause serious losses due to lower real starch digestibility than routine analysis would suggest. It is recommended to introduce corn silage processing quality control on dairy farms during the corn harvesting period, to improve the management and technical background in order to reduce the financial losses caused by inadequate CSPS values.

#### Introduction

Processing of corn silage may improve dry matter intake, starch digestion, and lactation performance (Bal et al., 2000 a,b). Processing increased (p<0.01) ruminal starch disappearance for both immature (844 vs. 664 g/kg) and mature (790 vs. 525 g/kg) whole-plant corn silage (Bal et al, 2000b). Dry matter intake (25.9 vs. 25.3 kg/d), milk yield (46.0 vs. 44.8 kg/ d) and milkfat yield (1.42 vs. 1.35 kg/d) were higher for the processed corn silage treatments (at a 1 mm roll clearance) compared with the control corn silage (Bal et al., 2000a). Kozakai et al. (2007) have found more rapid and greater colonization of the processed compared to unprocessed silage, by the rumen bacteria which facilitates ruminal digestion and fermentation. CSPS-data have been available in the USA since 2006 (CVAS laboratories). There has been a significant improvement in 2012 compared to the previous period. The CSPS analytical technique helped to identify the inadequate corn processing resulting in a positive effect on harvest technology. Authors investigated Corn Silage Processing Score (CSPS) of farm corn silage samples in Hungary between 2013-2015.

#### **Materials and Methods**

The CSPS is the percentage of starch passing through the coarse 4.75 mm screen (Ferreira and Mertens, 2005). Samples from different areas of Hungary collected by farmers during 2013 (n = 147), 2014 (n = 181) and 2015 (n = 243) were assessed. Samples were dried (70 °C, 8 hours) according to method EN ISO 6496:1993. Starch content was determined by Near-Infra Red Spectroscopy. Spectra were determined according to the guidelines of NEN-EN-ISO 12099 (Q-Interline Quant FT-NIR analyser). Reference starch method: NEN-EN-ISO 15914. Corn silage processing score (CSPS) was determined on dried samples with Ro-Tap Sieve shaker according to Ferreira and Mertens (2005). Modified starch digestibility, modified digestible starch and modified net energy content according to CSPS value were calculated based on Schwab et al. (2003).

#### **Results and Discussion**

CSPS results and distribution of the different fractions are shown in Table 1. Significant improvement was found in corn processing in 2015 compared to 2014. The ratio of inadequately processed corn silages was rather high (2013: 28%; 2014: 23%; 2015: 13%).), but decreasing tendency was found. Although, 65%, 61% and 74% of the corn silage samples were acceptable or adequate from a processing point of view in 2013, 2014 and 2015, respectively. According to our results, 7% (2013), 13% (2014) and 13% (2015) of the analysed corn silages were in the optimal quality range. The minimum were 31% (2013), 21% (2014) and 28% (2015), while the maximum values were 79% (2013), 75% (2014) and 81% (2015).

Detailed results of CSPS-corrected starch digestibility and modified net energy content based on the actual measured CSPS values are shown in Table 2. There were significant differences in corn silage starch content across the different years: there was lower starch concentration in 2013 and 2015 compared to 2014 (due to low precipitation volume, causing a drought-effect). CSPS-corrected starch digestibility was found to be higher in 2015 compare to 2013 and 2014.

Estimated starch, milk and corn-field equivalent losses are indicated in Table 3. The CSPS mean values of corn silages in 2013 were acceptable, but not optimal causing losses of 41 g/kg DM starch and 0.29 MJ/kg DM net energy compared to the original laboratory results (without CSPC-correction) due to undigested starch in inadequately processed corn silages (seed size: >4.75 mm). Starch-, energy-, estimated milk- and equivalent corn-field losses were significantly higher in 2014 compared to 2013 and 2015 due to the higher original starch content, as the effect of the year. Estimated milk losses were found to be considerable and significantly different (2013: 0.7 kg/cow/day; 2014: 1.3 kg/cow/day; 2015: 0.9 kg/cow/day, p $\leq$ 0.05), according to the CSPS and actual starch content of the corn silage. The losses (based on undigestible starch content) were equivalent to 14 ha, 19 ha and 12 ha corn grain, as an average for 2013, 2014, 2015, respectively (for 500 dairy cows during a 365 days period).

 Table 1
 Distribution of Corn Silage Processing Score mean values of corn silage (2013-2015, Hungary)

		Harvest 2013	Harvest 2014	Harvest 2015
Mean		55±11a	57±12a	61±9b
Sample number		147	181	243
Distribution of CSPS	fractions (% of to	tal sample numbe	er)	
<50	inadequate	28	26	13
50-60	acceptable	36	27	28
60-70	adequate	29	34	46
>70	optimal	7	13	13

<sup>*ac*</sup> different letters show significant differences  $p \le 0.05$ 

 Table 2
 CSPS-corrected starch digestibility, digestible starch and net energy content of corn silages based on actual measured Corn Silage Processing Score (2013-2015, Hungary)

	CSPS	Original starch content	Modified starch digestibility	Modified digestible starch	Original NEl	Modified NEl (based on digestible
	%	g/kg DM	%	g/kg DM	MJ/kg DM	MJ/kg DM
Harvest 2013 (n= 147)	55±11a	286±55a	86±9a	244±48a	6.32±0.23a	6.03±0.27a
Harvest 2014 (n= 181)	57±12a	369±51b	84±10a	307±47b	6.58±0.17b	6.01±0.35a
Harvest 2015 (n= 243)	61±9b	301±60c	87±8b	261±50c	6.35±0.26a	5.98±0.24a

<sup>*abc*</sup> different letters show significant differences  $p \le 0.05$ 

 Table 3
 Estimated starch-, milk and corn-field equivalent losses based on actual measured Corn Silage Processing

 Score (2013-2015, Hungary)

	Harvest 2013	Harvest	Harvest
Starch losses (g/kg DM)	41±28a	62±42b	40±26a
NEl losses (MJ/kg DM)	0.29±0.26a	0.57±0.39b	0.37±0.25c
Milk losses (kg milk/day/cow, 7kg DMI corn silage, 3 MJ NEl requirement/kg milk)	0.7±0.6a	1.3±0.9b	0.9±0.6c
Corn-field equivalent losses (ha corn seed - starch equivalent, for 365 days, 500 dairy cows, 7 kg DMI silage, 700 g/kg DM starch in	14±8.4a	19±13b	12±8.0a
$^{abc}$ different letters show significant differences $n < 0.05$			

abc different letters show significant differences  $p \le 0.05$ 

#### Conclusions

The wide range of farm silage CSPS (minimum value of 21% and maximum value of 81%) in Hungary between 2013-2015 shows the challenge of corn processing. Inadequate corn processing can cause serious losses due to the lower real starch digestibility (starch losses based on actual CSPS-values were found: 14 ha, 19 ha and 12 ha corn grain for 500 dairy cows during a 365 days period, as an average for 2013, 2014, 2015, respectively). It is recommended to introduce *corn silage processing quality control* on dairy farms during the corn harvest period, to improve the management and technical background in order to reduce the financial losses caused by inadequate CSPS values.

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### Effect of Storage Length on the Maize Starch Degradability

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#### Introduction

Maize starch effective degradability ( $ED_{starch}$ ) in the rumen influences the amount of ruminal organic matter truly digested.  $ED_{starch}$  is now included in the new French ration system INRA 'Systali' (Sauvant and Nozière, 2013). From a large *in sacco* database (n=168), Peyrat *et al.* (2016) proposed a formula based on forage DM and starch content. This equation was calibrated for a storage length of 2 months in silo (Peyrat, personal communication) due to a lack of variability in the database to include a length of storage factor despite having been demonstrated significant in several studies (Newbold *et al.*, 2006, Ward and De Ondarza, 2008). On farms, maize silage is fed after a few weeks to several months of storage; taking into account the length of storage effect on  $ED_{starch}$  is therefore necessary. This study is based on i) data collected in an *in sacco* degradability trial (Férard *et al.*, 2014) and ii) data from bibliography to propose a corrective coefficient of maize whole plant  $ED_{starch}$  depending on the length of storage of the silage.

#### **Material and Methods**

The *in sacco* study lead on ARVALIS Institut du vegetal La Jaillière experimental farm (FR-44370) investigates 5 whole plant maize harvested in 2013 from 287 to 403 g DM/kg. Samples were ensiled in lab jars (2l capacity) during 12, 40 or 68 days. Harvested forages were characterized by field measures (DM content, DM yield, proportion of cob) and by chemical composition before ensiling. For each storage length, fermentation parameters were analysed (pH, NH3-N, N solubility, volatile fatty acids, alcohols). ED<sub>starch</sub> was calculated by the step by step method (4 times of rumen incubation in fistulated cows) assuming an outflow rate of 6%.h<sup>-1</sup>.

Maize ED<sub>starch</sub> at 300 days of ensiling were predicted using the equation established on the data of Der Bedrosian *et al.* (2012) study: ED<sub>starch 300d</sub> = ED<sub>starch 68d</sub> / (-0.0075\*DM(%)+1.2346).

With the data for  $ED_{starch}$  at 68 days and 300 days of ensiling, slopes (SloED<sub>starch</sub> <sub>68-300d</sub>) of the relation  $ED_{starch} = f(length of storage)$  were studied according to DM and starch contents of the corresponding forages before ensiling.

#### **Results and Discussion**

All silages were very well preserved according to Dulphy and Demarquilly (1981). For the 2 silages at DM < 350 g/kg, *in sacco* starch degradability increased slightly (+0.6 percentage unit between 12 and 68d) whereas for the 3 silages with DM  $\geq$ 350 g/kg, ED<sub>starch</sub> has risen sharply from 0.782 at 12 days to reach 0.855 at 68 days of ensiling (Table 1).

The calculated ED<sub>starch</sub> slopes between 68 and 300 days were strongly correlated to DM at harvest (R<sup>2</sup>=0.98) (Figure 1). SloED<sub>starch</sub> 68-300d were 4 times lower for the 2 silages at DM < 350 g/kg (on average 318 g DM/kg) than for the 3 silages at DM > 355 g/kg (on average 390 g DM/kg). Considering the evolution after 2 months of ensiling, the increase in ED<sub>starch</sub> per day has been estimated at 0.0022 percentage unit per additional 10 g/kg of whole plant DM beyond 291 g/kgDM (e.g: 0.024 percentage unit per day of ED<sub>starch</sub> for a silage harvested at 400 g/kgDM): this is the corrective coefficient (ccED<sub>starch</sub>). These results are very consistent with the prediction of ruminal starch degradability increase over months of storage (Junges *et al.*, 2013) estimated at 0.01 (for 48h incubation) and 0.03 (for 24h incubation) percentage unit per day for a maize silage harvested at 400 g/kg of DM.

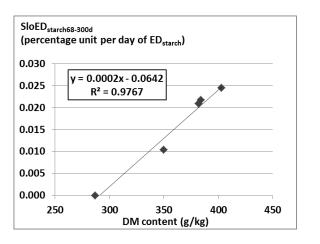
	Whole plant	Kernel DM	Starch	ED <sub>starch 12d</sub>	ED <sub>starch 40d</sub>	ED <sub>starch 68d</sub>	ED <sub>starch 300d</sub>	SloED <sub>starch</sub>
	DM content	content	content	measured	measured	measured	predicted	68-300d
	(g/kg)	(g/kg)	(g/kg)	(%)	(%)	(%)	(%)	(unit.day <sup>-1</sup> )
DM<355 (g/kg) N=2	318	481	239	88.9	89.6	89.6	90.8	0.0052
DM≥355 (g/kg) N=3	390	635	362	78.2	80.8	85.5	90.7	0.0224

 Table 1
 Characterization and starch degradability of the 5 maize silages studied

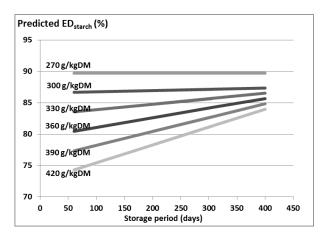
When the coefficient ccED<sub>starch</sub> is applied on the predicted maize ED <sub>starch</sub> *in sacco* "Systali" (Peyrat *et al.*, 2016), reference curves can be obtained depending on the forage DM at harvest (Figure 2).

As a consequence of ED<sub>starch</sub> increase, the lengthening of maize silage ensiling duration from 2 to 8 months leads to an increase of 4 g/kgDM and 20 g/kgDM in the amount of ruminal starch digested respectively for a silage harvested at 318 g/kg and 390 g/kg of DM.

**Figure 1** Daily variation of ED<sub>starch</sub> between 38 days and 300 days of ensiling for the 5 silages studied



**Figure 2** Evolution of maize predicted ED starch (Peyrat et al., 2016) depending on the DM content at harvest and corrected by the silage storage length effect (1)



This *in sacco* trial confirmed that maize silage  $ED_{starch}$  increases during the first weeks of storage (Der Bedrosian *et al.*, 2012, Ferraretto *et al.*, 2015). This increase is especially important as the DM content at harvest is high (late harvest stage) as observed by Der Bedrosian *et al.*, (2012) and Windle *et al.* (2014).

This study results in a correction factor that can be applied to the  $ED_{starch}$  "SYSTALI" (Peyrat *et al.*, 2016) to refine ruminant ration formulation with:

(1)  $ED_{starch corrected}$  (%) =  $ED_{starch SYSTALI}$  (%) + 0.00022 \* [DM (g/kg) - 291] \* [storage length (days) - 60]

#### Conclusion

The digestive starch dynamic influences the amount of ruminal organic matter truly digested. Thus, the diet balance is also under variation over months due to the duration of ensiling on maize starch degradability.

To improve ruminant diet adjustment, this study proposes a first approach to take into account the ensiling time effect. It provides an equation to take into consideration a well-known phenomenon observed in commercial farms by farmers and technicians even though very few references can be directly practicable into ruminant rationing. Ongoing studies involving more hybrids types will strengthen knowledge about length of storage effect on starch degradability.

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## Effect of Crimped Maize Grain Ensiled with High Moisture Grains of Transgenic Bt Maize in Fattening Bulls

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#### Abstract

The maize event MON 810 carries the novel gene cry1Ab from a naturally soil bacterium, Bacillus thuringiensis (Bt). Thus the transgenic Bt-maize produces the insecticidal Cry1Ab protein which provides resistance against the European corn borer. However, the increasing use of genetically modified plants in the production of animal feed has raised concerns about their safety. A feeding trial was carried out with 40 Holstein breed bulls with an initial live weight 298 kg which were randomly distributed into 4 groups with 10 bulls in each, housed in boxes. The feed consisted of maize silage, lucerne silage, meadow hay, wheat, rape extr. meal, minerals and crimped maize grain in two feeding TMR with content of isogenic maize and transgenic maize (event MON 810) intended for bulls. Bulls were fed TMR diet for ad libitum intake. Water was provided ad libitum. The TMR contained 139.84 g.kg<sup>-1</sup> crude protein, 6.45 MJ.kg<sup>-1</sup> NEV, 155.3 g.kg<sup>-1</sup> crude fibre, 264.47 g.kg<sup>-1</sup> starch, fat 28.8 g.kg<sup>-1</sup>, Ca 9.21 g.kg<sup>-1</sup>, P 3.67 g.kg<sup>-1</sup>, Na 2.06 g.kg<sup>-1</sup>, Mg 2.18 g.kg<sup>-1</sup> and K 10.735 g.kg<sup>-1</sup> in dry matter. The feeding trial was conducted with 2 x 20 bulls from average live weight 298 kg to the average slaughtering weight 620 kg in control and 622 kg in experimental group. The live weight gain was 1.248 kg.d<sup>-1</sup> and 1.255 kg.d<sup>-1</sup>. The experiment lasted for 258 days. Data were processed by analysis of variance. The significance of differences was evaluated by the t-test. In fattening experiments there were studied live weight growth of bulls and consumption of feed mixtures per unit of live weight growth. Obtained results demonstrate minimal, statistically non-significant differences of individual parameters in tested groups. Bulls were slaughtered at the age of 586 days after achieving the live weight 620.5 kg. Feeding of TMR with proportion Bt transgenic and isogenic control maize to Holstein breed bulls did not influence zootechnical parameters, as well as it has no negative effect on feed conversion, growth performance, meat quality and health status.

Key words: high moisture grain; fermentation process; transgenic maize MON 810; Holstein bulls

#### Introduction

Transgenic crops have been grown commercially for about 18 years, with a continuous increase of the cultivated area, and now they represent a significant proportion of the total world production for the major GM crops: soybean, maize, rapeseed and cotton. These crops are used directly or preserved in animal nutrition or as by-products from the processing industry such as distillers grain, extracted oil meal, sugar beet pulp, *etc.* Therefore, nutritional and safety assessments of feeds from GMP are one of the key questions. Most animal studies were conducted for the nutritional assessment of feeds. Flachowsky et al. (2005) and Chrenková et al. (2007) summarized the effects on the nutrients digestibility, feed intake, health and performance of animal as well as effects on the quality of food of animal origin. In the European context, Slovakia ranks among other 7 EU countries which have practical experience in Bt maize cultivation (Křistková, 2009; James, 2011). Maize MON 810 contains the Cry1Ab protein protecting the crop against certain lepidopteran insect pests, including the European Corn Borer (*Ostrinia nubilalis*) and pink borers (*Sesamia spp.*).

Crimped ensilage maize of rolled, moist grain is a suitable alternative to combine harvesting for grain intended to be used on-farm and when moist weather prevails during harvest. Farmers are recommended to apply additives during crimping (Pauly, 2015). Benefits of storage of high moisture grains in Nordic conditions have been already proven some decades ago (Palva *et al.* 2005; Jaakkola *et al.*, 2005). No drying costs, less dependency on weather and an extended harvesting season are the main arguments that keep the method increasing in popularity. The method has become more common on farms with large herd sizes and total mixed ration (TMR) feeding.

The aim of this work was to verify substantial equivalence of maize MON 810 in fattening Holstein bulls from the investigation of fattening ability; carcass and consumer meat quality were obtained at the average age of 586 days.

#### **Material and Methods**

Experiment consisted of two variants (without preservative agents, control crimped isogenic maize grain (DKC 5143) and experimental variant transgenic maize MON 810. We preserved both variants into plastic sacks (2 meter diameter, 10 - 50 meter length) without any additives, volume of which were hermetically maintained. After five months of fermentation were the samples of silages determined for nutrient experiment opened and from average samples we evaluated parameters of nutritive value and fermentation process according to the AOAC (1995). Organic acids levels were determined by gas chromatography, alcohol by the microdiffusion method according to Conway (1962). Material for experiment was obtained from farm Mladějovice, Šternberk in the Czech Republic.

A feeding trial was carried out with 40 Holstein bulls, which were randomly distributed into 4 groups, with 10 bulls in each, housed in boxes. Bulls were fed TMR diet for *ad libitum* intake.

The diets were fed twice daily at 6.00 and 16.00 h in two equal doses. Water was provided *ad libitum*. The experiment lasted for 258 days. Detail dissection of right half carcass was done 24 hours after slaughtering to obtain weight and proportion of basic tissues (muscle, fat and bones). Weight of valuable cuts was calculated as a sum of weight of round (boneless round without back shank), shoulder (boneless shoulder without front shank), back (boneless *musculus longissimus thoracis* between 9<sup>th</sup> *thoracic vertebra* and 10<sup>th</sup> *thoracic vertebra*) and tender loin. The samples (approximately 500 g) were taken from MLTL (*musculus longissimus thoracis et lumborum*) and for the first time they were analysed as a fresh meat for physic-chemical meat quality after 48 hours of chilling.

Table 1	Content of nutrients and mycotoxins in crimped isogenic maize grain and crimped transgenic maize grain
(g.kg <sup>-1</sup> orig	zinal matter)

Parameter	Crimped isogenic maize grain	Crimped transgenic maize grain MON
	(DKC5143)	810
Dry matter	642.2±0.3	616.6±0.1
Crude protein	68.4±1.7	65.7±1.5
Fat	33.4±0.5	25.3±1.1
Crude fibre	9.6±0.1	16.6±0.3
Starch	438.1±0.3	435.2±0.1
ADF	17.8±0.1	23.6±1.1
NDF	87.3±0.1	75.7±0.6
ME (MJ.kg <sup>-1</sup> dry matter)	14.48	14.43
NEF (MJ.kg <sup>-1</sup> dry matter)	6.45	6.45
Fumosidines (FUM) *	1250	trace
Resul	lts of fermentation process of high moist	ture corn silage
pH	4.54	4.41
Acetic acid	0.89	1.01
Propionic acid	0.62	0.77
Butyric acid	0.03	0.08
Lactic acid	3.48	3.05
Total amount of acid	5.17	5.15
Alcohol	0.05	0.05

NEF = net energy of fattening, \*Total were of quantification in  $\mu$ g.kg<sup>-1</sup>

**Table 2**Composition of the TMR diet

Ingredients (kg.d <sup>-1</sup> )	Daily feed intake	Ingredients (kg.d <sup>-1</sup> )	Daily feed intake
Maize silage	10.5	Rape meal	0.5
Lucerne silage	5.0	Vitamin- mineral mix*	0.2
Meadow hay	1.0	Crimped maize grain	2.8
Wheat	0.6		

\*The supplement vitamin- mineral mix provided (per kg of additive): calcium carbonate, wheat, monocalcium phosphate, sodium chloride, zinc chelate and amino acid n-hydrate, sugar beet molasses, vit. A, D3, E, copper sulphate pentahydrate (CuSO<sub>4</sub>.5H<sub>2</sub>O), dioxide magnesia

**Table 3**Nutritive value of TMR the diets

Parameters	Control group	Experimental	Parameters	Control group	Experimental	
(g.kg <sup>-1</sup> DM)		group			group	
Crude protein	139.8	138.6	Ca:P	Ca:P 2.51		
NEF (MJ.kg <sup>-1</sup> )	6.4	6.4	K:Na	5.21	5.57	
Crude fibre	155.3	162.4	Parameters (mg.kg <sup>-1</sup> )			
Starch	264.5	251.4	Zn	68.6	87.6	
Fat	28.8	21.6	Cu	20.4	21.7	
Ca	9.2	8.5	Ι	1.0	1.1	
Р	3.7	3.2	Se	0.4	0.5	
Na	2.1	2.0	Parameters IU.kg <sup>-1</sup>			
Mg	2.2	2.2	Vit. A	3 118	2 033	
K	10.7	11.0	Vit. D	926	1 017	

Chemical parameters of meat (proteins, fat and total water content) were analysed afterwards, when no more changes in chemical composition of meat are in progress. The devices Nicolet 6700 Spectrometer or Infratec 1265 with the application module for fat content assessment 1 - 10% were used. The pH meter Toledo with combined stab electrode was used to measure pH value. Water holding capacity was analysed by the Gramm Hama method. Meat colour (values L, a and b) was measured by the MiniScan XE plus.

The results were quoted as mean  $\pm$  standard deviation (SD); statistical evaluation of the results was performed by the one-way ANOVA and Tukey test.

#### **Results and Discussion**

The diet consisted of maize silage, lucerne silage, meadow hay, wheat, rape meal, minerals and crimped isogenic maize grain or transgenic maize (Table 2). Water was provided *ad libitum*. The nutrient compositions of diets revealed no major differences. The TMR contained 138.60 - 139.84 g.kg<sup>-1</sup> crude protein, 155.3 - 162.4 g.kg<sup>-1</sup> crude fibre, 251.36 - 264.47 g.kg<sup>-1</sup> starch, fat 21.62 - 28.8 g.kg<sup>-1</sup>, Ca 8.552 - 9.21 g.kg<sup>-1</sup>, P 3.242 - 3.67 g.kg<sup>-1</sup> and energy value NEF (net energy of fattening) its 6.43 - 6.45 MJ in kg dry matter (is presented in Table 3; standard of analysis according to AOAC,1995).

Increased content of mycotoxins was observed in isogenic maize (1250 µg.kg<sup>-1</sup>). All samples were contaminated by toxins under the EU limit (Table 1). By the valuation fermentative of the process at both of them silage from moist maize grain are found out that he course was he at both of them silage similar. Silage from grain GM maize had lower pH and by over little higher contents of acetic acid and propionic acid. The content of lactic acids was lower than at silage from grain izogenic maize.

The feeding trial was conducted with 2 x 20 growing bulls, Holstein breed, from average live weight 298 kg to the average slaughtering weight 620 kg in control and 622 kg in experimental group (Table 4). The live weight gain was 1.248 kg.d<sup>-1</sup> and 1.255 kg.d<sup>-1</sup>. Several existing studies, which compare the substantial equivalence between conventional and transgenic plants, applied usually to the production of fodder mixture for farm animals (Flachowsky et al., 2005; Sampietro et al, 2011; EFSA, 2008). Metabolism and performance data revealed no significant differences between the bulls that received the TMR with conventional, non-modified maize, and those that received the TMR with modified maize diets.

Table 4	Performances of b	ulls in response to	dietary supple	ementation of isog	genic maize and	l transgenic maize

Parameters (n=20)	Control group	Experimental group		
Initial weight (kg)	298	298		
Final weight (kg)	620	622		
Daily weight gain (kg.day <sup>-1</sup> )	1.25±0.2	1.26±0.1		

P >0.05; Means ±SD

The effect of addition of crimped isogenic maize grain or crimped transgenic maize grain to TMR is shown in figure 1. Average daily gains during the experiment in the control group were 1.25 kg.day<sup>-1</sup> and the values ranged from 0.79 kg.day<sup>-1</sup> to 1.60 kg.day<sup>-1</sup>. Average daily gains in the experimental group were at 1.26 kg.day<sup>-1</sup>, the range we have seen from 1.14 kg.day<sup>-1</sup> to 1.39 kg.day<sup>-1</sup>. In this group the variability was not significant. The health conditions are evaluated subjectively. Either in experimental or control group were not problems with the hygienic conditions of the animals, the weight gains were on adequate levels. No significant differences were found among experimental groups in feed intake and body weight in the fattening experiment (Table 4). It had no adverse effects on feed conversion, growth performance and health status. The transgenic maize in TMR has no deleterious or unintended effects.

Table 5Physico-chemical characteristics of meat Holstein bulls (Musculus longissimus thoracis et lumborum -MLTL) 48h post mortem

Parameters	Unit	Control group			Experimental group		
		Minimum	Maximum	SD	Minimum	Maximum	SD
Total water content	g.100g <sup>-1</sup>	74.7	75.1	0.2	75.0	75.7	0.15
Content of proteins	g.100g	21.7	21.6	0.05	21.2	21.5	0.15
Content of fat	g.100g	2.2	2.6	0.2	1.5	1.9	0.2
Energetic value	kJ.100g <sup>-1</sup>	416.64	461.44	1.57	416.64	442.1	12.73
pH 24h		5.76	5.79	0.01	5.83	5.81	0.01
Colour L	%	23.62	24.37	0.37	25.48	25.44	0.02
Water holding capacity	g.100g <sup>-1</sup>	33.07	34.12	0.52	36.56	35.74	0.41

An important parameter of meat quality is the end pH value, which affects water binding capacity, color of the meat and its tenderness, and thus plays a key role in sustaining meat quality during storage and the length of shelf life. As presented in Table 5, no differences between the pH values of the control and experimental groups were determined. Feeding GM maize had no influence on the other characteristics of meat quality.

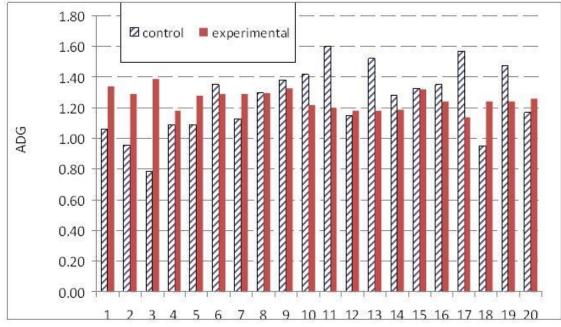


Figure 1 Comparison of daily weight gain in the fattening experiment; bulls (1-20) were fed TMR with content of isogenic maize (control group) or transgenic maize (experimental group).

#### Conclusion

Feeding TMR with GM maize MON 810 and isogenic maize to bulls did not influence biochemical and zootechnical parameters, as well as it had no negative effect on health status and growth performance of animals. No significant differences were found between the individual nutrients which corresponded with the results of live weight in slaughter bulls. There was no effect of diet on pH value, chemical composition of meat and its colour.

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# The Level of Rumen Fermentation in Heifers During Transition from Winter to Pasture Feeding

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#### Introduction

Grazing promotes the development of a massive and efficient digestive tract that is able to process and utilize nutrients from a large ration of voluminous feeds. Ruminal fermentation metabolites, VFA and microbial proteins present the basic nutrients supplied by roughage. In this transitory grazing period it is essential to guarantee the physiological processes of rumen fermentation (Kay, 1993). At the time when animals are turned out from the stable (winter feeding) to graze, the young grassland contains increased amounts of nitrogen and decreased amounts of dry matter and fibre, thus affecting the level of rumen fermentation and metabolism in the grazing animals. As claimed by Jambor (1986), transition to pasture not only wasted valuable proteins but often lead to dietetic and metabolic disorders: the altered nutrient intake lead to changes in ruminal biochemism, decreased blood glucose and increased urea levels. Concentrate supplements reduced molar proportion of acetate and increased the molar proportion of propionate in some studies (Bargo et al., 2002). Two studies evaluated the effect of forage supplements such as corn silage (Elizalde et al., 1992) or hay (Reis and Combs, 2000) on ruminal fermentation of dairy cows on pasture. This work focused on the changes of rumen fermentation in heifers in the period of transition from the winter feeding ration to pasture and in the subsequent grazing cycles.

#### **Material and Methods**

In this experiment 6 blackspotted heifers aged 6 months with a mean body weight of 220 kg were included. Prior to turn-out to pasture, the winter feeding ration consisted of 6 kg maize silage, 4 kg green rye cuttings, 1 kg pasture hay and 1 kg barley straw. After the animals had been turned out to pasture, the basic part of their ration consisted of grass crops, the nutritive value of which during the single cycles of grazing is given in Tab. 1. In the first week of pasture, they were given additional 2 kg of maize silage and during the first month, 1 kg of grass hay per animal and day. Samples of rumen contents were taken through a pharyngeal tube. The first sampling from animals was carried out one week prior to driving the animals to pasture. On pasture, rumen contents were sampled at weekly intervals from Week 1 to Week 4 (May, June - Cycle I), then in Week 8 and 12 (July, August - Cycle II) and in Week 16 (September - Cycle III). Volatile fatty acids in the rumen fluid were determined by gas chromatography using a Hewlett Packard (USA) gas chromatograph with a column containing 10% SP 1200 + 1% H<sub>3</sub>PO<sub>4</sub> on Chromosorb W. The energetic yield of VFA production was calculated in percentage of gross energy according to Orskov et al. (1968). Strictly anaerobic bacteria were determined on selective media in Black's bottles in a protective CO<sub>2</sub> atmosphere. RGCA medium supplemented with microcrystalline cellulose (6g.1<sup>-1</sup>, treated with phosphoric acid) was used to isolate cellulolytic bacteria. MLH medium (Mackie and Heath 1979) was employed to state the total numbers of lactate-utilizing bacteria. For the isolation of lactobacilli Rogoza Agar (Oxoid) was used while streptococci were isolated on Base Agar No. 4 supplemented with 1% starch. Statistical analyses were carried out using Student's t-test. The results are mean values  $\pm$  SD.

	Cycle of grazing				
	I.	II.	III.		
Dry matter	199	229	259		
Organic matter	180	208	230		
Crude fibre	51	73	94		
N-free extract	37	34	35		

**Table 1** Nutritive value of the grassland in the single cycles of grazing (g.kg<sup>-1</sup>)

#### **Results and Discussion**

After turn-out to pasture, total VFA (volatile fatty acid) concentrations in the rumen contents of heifers significantly decreased from 107.7 to 88.7 mmol.1<sup>-1</sup> (P<0.01; Tab. 2). Subsequently total VFA concentrations increased and reached their highest value (117.0 mmol.1<sup>-1</sup>) in the 8th week of grazing. The proportion of the molar acetic acid concentration in the rumen contents increased insignificantly from 67.1 to 70.1 mol% whereas that of propionic acid decreased insignificantly from 18.4 to 16.8 mol%. In the molar proportion of butyric acid no significant differences could be stated. In the following weeks the molar proportions of acetic acid were rather balanced and ranged from 66.2 to 68.0 mol%; the molar proportion of propionic acid insignificantly increased in the 4th week (18.7 mol%). The molar proportions of butyric acid on pasture insignificantly increased in week 3 (15.2 mol%) and appeared to be balanced, acquiring later values between 14.0 and 14.8 mol%. The energetic yield of VFA production in the rumen of young heifers decreased insignificantly from 73.6 to 72.7%, the acetate-to-propionate ratio revealed an insignificant increase from 3.66 to 4.18 (Tab. 2). The change of feeding after turn-out to pasture leads to an increase in the content of nitrogenous substances as well as a decrease of dry matter and crude fibre, thus affecting both supplementation and

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ruminal digestion (Bargo et al., 2003). In this experiment, the transition from winter feeding to grazing lead to a significant, one-week-lasting decrease of VFA concentrations in the rumen content of the heifers. Subsequently an increase of the total VFA levels occured during weeks 3 and 4 that approached the levels observed prior to driving the animals to pasture. On the basis of the total VFA dynamics it can be stated that the adaptation of the rumen ecosystem lasted 3-4 weeks. Supplementation with maize silage during the first week of grazing contributed to shortening of the adaptation period. Sayers (1999) found that supplementation with fiber-based concentrates increased the molar proportion of acetate and butyrate, and decreased the molar proportion of propionate. Khalili and Sairanen (2000) reported no changes in the molar proportion of any of the three major VFA. In this experiment, the transition to grazing was accopanied by an insignificant increase of the molar acetic acid concentrations and a decrease of propionic acid, whereas the proportion of butyric acid only decreased in the 1st week of grazing. The above molar proportions of VFA are reflected in the acetate:propionate ratio that increase after turn-out to pasture as well as in the energetic yield of VFA production that revealed a decrease during the transitory period. For several types of diets Orskov (1975) determined an energetic yield of VFA production between 73 and 87%. In this relation, the energetic yield of VFA production in our experiment fluctuated at the lower border of the abovementioned range.

		Stable Sampling on pasture at weekly intervals							
			1	2	3	4	8	12	16
Total VFA	Х	107.7	88.7*	96.5	103.2	114.8*	117.0	113.6	109.2
$(\text{mmol.l}^{-1})$	SD	5.2	4.1	4.6	3.7	5.8	7.3	6.4	4.4
Acetic acid	Х	67.1	70.1	68.0	66.8	66.2	67.1	66.9	67.2
(mol %)	SD	2.6	3.8	3.2	2.8	1.9	2.6	3.0	2.8
Propionic acid	Х	18.4	16.8	17.4	17.1	18.7	17.4	18.1	17.9
(mol %)	SD	1.6	2.7	2.5	2.6	2.0	2.0	2.1	1.8
Butyric acid	Х	13.0	11.9	13.6	15.2	14.3	14.8	14.0	14.2
(mol %)	SD	1.1	2.1	1.7	1.8	1.5	0.9	1.3	1.2
Acetate : propionate	Х	3.66	4.18	3.91	3.9	3.53	3.84	3.69	3.76
	SD	0.43	0.57	0.63	0.6	0.4	0.55	0.48	0.52
Energetic yield of VFA	Х	73.6	72.7	73.3	73.4	73.9	73.5	73.7	73.6
(E %)	SD	0.67	0.82	0.76	0.89	0.93	1.05	0.69	0.74

**Table 2** Rumen fermentation in the rumen content of heifers

\* p < 0.01 significance in comparison with the preceding value, (n = 6)

Cellulolytic bacteria counts in the rumen contents significantly decreased from 8.08 to 7.61 log  $10 \text{ ml}^{-1}$  (P<0.01) and then a significant increase to 8.39 log $10 \text{ ml}^{-1}$  was observed again in the 3rd week of grazing (P<0.05). During pasture, a significant increase in the counts of lactate-utilizing bacteria was recorded. As to the numbers of lactobacilli, a significant decrease (P<0.05) with a subsequent insignificant increase during grazing could be seen. Throughout the grazing season, the counts of streptococci reached values that surpassed those recorded during the winter feeding period (Tab. 3). Our data concerning cellulolytic bacteria in the rumen contents of heifers coincide with those published by Leedle and Butine (1987) and Dehority et al. (1989) who reported the numbers of these bacteria in animals held on rations containing high proportions of bulk feeds. In comparison with the winter feeding, increased numbers of lactate-utilizing bacteria and streptococci as well as decreased numbers of lactobacilli were observed throughout the grazing period.

Table 3	Total numbers of bacteria in the rumen contents of heifers (log 10.ml <sup>-1</sup> )
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Group of bacteria	Stable	Stable Sampling on pasture at weekly intervals					
			1	2	3	4	8
Cellulolytic	Х	8.08	7.61**	7.92	8.39*	8.43	8.38
bacteria	SD	0.07	0.21	0.31	0.19	0.22	0.2
Lactate ulitizing	Х	7.54	8.95**	8.86	9.12	9.69*	9.58
bacteria	SD	0.31	0.12	0.27	0.12	0.38	0.36
Lactobacilli	Х	4.05	3.03*	3.48	3.76	3.52	3.8
	SD	0.15	0.25	0.1	0.11	0.19	0.04
Streptococci	Х	5.04	5.16	5.43	6.14*	5.93	5.5
	SD	0.22	0.16	0.21	0.19	0.31	0.19

\* p < 0.05, \*\* p < 0.01 significance in comparison with the preceding value, (n = 6)

#### Conclusion

The transition from winter feeding to grazing had an adverse affect upon rumen fermentation which required a certain adaptation phase in order to normalize. In our experiment this adaptation phase took 3 weeks.

References are available from the author.

## Session 4 – Technologies in Production of Silages and Energy Production in Biogas Plants

### **Oral presentations**

Silage safety practices that save lives Bolsen, R. E., Charley, R., Wilkinson, J. M., Bolsen, K. K.

Long-term storage of brewers' grains with no additives in the bag Loučka, R., Tyrolová, Y., Homolka P., Jančík, F., Kubelková, P., Výborná, A.

Ash concentrations in silages can be used to assess avoidable top layer losses due to aerobic spoilage in silos on commercial farms Chamberlain, A. T., Wilkinson, J. M., Bolsen, K. K.

### **Poster presentations**

Labour requirement for barn dried hay production and feeding on dairy farms Mačuhová, J., Jakschitz-Wild, S., Haidn, B., Thurner, S.

Influence of soil conservation technologies of maize seeding on yield and forage <u>quality</u> Nerušil, P., Menšík, L., Kincl, D., Srbek, J.

Increasing of effective fiber at TMR by new technology (Shredlage) for harvest of maize silage Jambor , V., Vosynková, B., Illek, J., Kudrna, V.

#### Silage Safety Practices that Save Lives

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#### Introduction

Few farming operations invite as many different opportunities for injury or fatality as a silage program. One of these is an avalanche or collapsing silage. It only takes a fraction of a second for part of an exposed silage feed-out face to break off silently and fall, without prior warning. The result can be deadly for anyone beneath. Many bunker silos and drive-over piles are too large to be safe for the persons filling and feeding out. Bigger livestock units have resulted in greater amounts of silage being stored in outdated bunkers and piles. Unless new silos are built, the footprint of drive-over piles enlarged, or packing density increased significantly, there is nowhere for additional silage to be stored but in bunkers or piles of ever-increasing height. Thus, the risk of an avalanche tragedy is increased. It is not uncommon for cattle feedlots, large dairy units and anaerobic digestion plants to have bunkers and piles with silage feed-out faces that are 5.5 to 7.5 m high. Common sense tells us that a 6 to 7 m tall silage face is far more dangerous than one that is only 3 to 3.5 m high.

Unfortunately, the silage industry has a long way to go to eliminate serious injuries and fatalities from silage avalanches. There have been several fatalities in the United States the past few years, including an 11-year old boy in New Hampshire, a 30-year old truck driver in Idaho, and a 63-year old employee in Pennsylvania (Bolsen and Bolsen 2013; Bolsen and Bolsen 2014). Although rarely reported, the authors have heard many stories about someone having a near miss or suffering a serious injury in a silage avalanche.

In this paper silage avalanche and fall from dangerous height tragedies are documented and guidelines for reducing the risk of injury and death are outlined.

#### **Materials and Methods**

Documentary evidence concerning silage avalanches and falls from dangerous heights is presented as four detailed case studies. Guidelines for improving the safety of bunkers and piles, based on the case studies, are presented with the objective of reducing the risk of injury or death in a silage program.

#### **Results and Discussion**

#### Case studies

A New Mexico online news organization reported the following fatal accident (Tucker 2014). On January 13, 2014, Jason Leadingham was working in a bunker silo when a massive amount (10 to 15 tonnes) of maize silage collapsed on him. Pirtle Farms LP of Roswell, New Mexico who employed Jason as a truck driver owned the silo. Jason's body was not recovered from the silage until about 2.5 hours after the avalanche. The cause of death was determined to be mechanical asphyxiation. A silage sample bag was near Jason's left hip. He was clutching silage in his hands and had silage in his mouth, which suggest that Jason struggled to survive in the final moments of his life.

A Nebraska newspaper reported that a 53-year old man died on October 21, 2013 in a silage accident (Bolsen and Bolsen, 2014). Stanton County Sheriff Mike Unger said Matthew Winkelbauer died after he was buried by a large silage pile that fell in an open silage pit at Four-Quarters Feedlot east of Norfolk. Winkelbauer, who was the owner and operator of Four-Quarters, was pronounced dead at the scene. A co-worker was seriously injured in the accident. The victim was standing in front of the feed-out face, which was about 4 m high, and the avalanche pushed the falling silage more than 8 m from the feed-out face.

William John Davidson who was 52 years old died at Poldean Farm, Moffat, Scotland at about 2:38 pm on January 26, 2013 (<u>https://www.scotcourts.gov.uk/search-judgments/judgment?id=619a8ea6-8980-69d2-b500-ff0000d74aa7</u>). The cause of William's death was a severe head injury sustained in a fall from a height of 4.9 m after he lost his footing while working on top of a silage pile. A reasonable precaution whereby his death might have been avoided would have been that of keeping a safe distance from the edge of the silage pile. A defect in the system of working, which contributed to the death, was the absence of any measure whereby the need to work close to the edge of the silage pile was avoided.

Professor Ali Assadi-Alimouti, University of Tehran, Iran (personal communication 2013) described the serious injuries he received in a silage avalanche: "It was March 15, 2010 and I went to see a large dairy farm client. Two of the herdsmen and I went to the large bunker silo (8,000 tonnes capacity). The height of the feed-out face was about 6 meters. After visual appraisal of the silage, we were walking out of the bunker and a large silage avalanche fell on us. Observers testified later that it was around 10 tonnes of silage. One of the herdsmen remained outside of the silage from his head, and thank God, he could call to others to save us. The worst injuries happened to me, including multiple fractures to my tibia and femur, and I was in a coma for 30 hours in a hospital. The other herdsman suffered a broken leg and had respiratory problems due to inadequate oxygen for 10 minutes. I was the last one rescued, being trapped under the silage for about 20 minutes. It is by the grace of God that I am alive. God gave me another chance for life."

#### Guidelines for improved safety procedures

Guidelines to decrease the risk of a serious accident or fatality caused by a silage avalanche or a fall from a dangerous height have been formulated (Bolsen et al., 2015). The important principles are i) avoid excess height when filling bunker silos and building drive-over piles and ii) avoid working close to the unstable exposed feed-out face.

#### Specifically:

1) Bunker silos and drive-over piles should not be filled higher than the unloading equipment can reach safely, and, typically, an unloader can reach a height of 3 to 3.5 meters;

2) Do not exceed the height of the safety rail on the bunker walls;

3) Never allow people to approach the feed-out face;

4) Never stand closer to the feed-out face than three times its height;

5) Suffocation is a primary concern and a likely cause of death in many silage avalanches, so follow the '*buddy rule*' and never work alone in a bunker or pile;

6) Never drive the unloader parallel to and in close proximity of the feed-out face in an over-filled bunker or pile;

7) Take silage samples from an unloader bucket after it is moved to a safe distance from the feed-out face;

8) Use caution when removing plastic or oxygen-barrier film, tires, tire sidewalls or gravel bags;

9) Do not work or stand close to the top edge of the feed-out face;

10) Do not "pitch" surface spoilage. It is simply too dangerous to remove spoilage from the top of many bunkers and piles;

11) Never ride in a front-end loader bucket;

12) Never park vehicles or equipment near the feed-out face;

13) Post warning signs around the perimeter of bunkers and piles saying, 'Danger! Silage Face Might Collapse'; and

14) Avoid being complacent and never think that an avalanche or a fall cannot happen to you.

#### Conclusions

A silage avalanche can occur anywhere, anytime, in any bunker or pile, without warning and in any ensiled forage. We cannot stop avalanches from happening, and they are impossible to predict, but we can prevent people from being under them. A fall from a dangerous height does not have to happen; it is avoidable.

Every farm, feedlot, dairy and digester plant should have safety policies and procedures for their silage program, and they should schedule regular meetings with all their employees to discuss safety.

If a silage program is not safe, nothing else about it really matters.

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#### Long-term Storage of Brewers' Grains with no Additives in the Bag

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#### Introduction

Wet Brewers' Grains = WBG has been commonly stored with the help of a system of plastic bags filled with WBG directly from the body of a truck. As, due to technological reasons, a preservative cannot be applied when filling the bag directly from the body of a truck, it is recommended in practice to feed the WBG to livestock within two weeks from storage. But such period is relatively short for some farmers with lower numbers of livestock.

The work was aimed at finding out whether the time of storage of the brewers' grains could be extended by several weeks without significantly reducing its quality. That could bring considerable savings to the farmers, particularly if they did not need to apply preservatives to the brewers' grains or to combine them with another material with higher amount of dry matter (e.g. with malt sprouts). The combination of brewers' grains with another material causes technological problems and the addition of preservatives, both biological and chemical, does not guarantee a good result. When mixing both materials, they may get contaminated by undesirable micro-organisms (Orosz and Davies, 2015).

#### **Materials and Methods**

The experiment took place from 9.6. to 29.9.2015. It was performed in The Special-Purpose Farm of the Research Institute of Animal Science of Prague. Immediately after storing the brewers' grains with an average content of dry matter of 19.8 %, the Thermochron temperature sensors were inserted into the bag at three places: one sensor stayed on the bag surface, the second was under the canvas in a depth of 5 cm, the third in a depth of 20 cm, and the fourth in a depth of 35 cm. The hole left from the insertion of the sensors was immediately closed up with a special adhesive tape. The sensors (as big as a watch battery) remained at the same place all over the time of storage of the brewers' grains. The temperature was measured each 2 hours with precision of 0.065°C. After the end of the experiment, the sensors were removed and the temperatures were read with the help of a special reader.

Samples of brewers' grains were taken from three different places of the bag from a depth of about 30 cm were taken each 2 weeks (8 samplings in total). The holes in the canvas were closed up with a special adhesive tape immediately after the sample taking. The following nutritional values were determined in the samples: dry matter, protein, fibre, fat, ashes, reducing sugars (WSC), neutrally detergent fibre (NDF), acido-detergent fibre (ADF) and indicators of fermentation quality: pH, acidity of water extract, lactic acid, volatile fatty acids (acetic acid, propionic acid, butyric acid, valeric acid) and ammoniac nitrogen (NH3-N). All samples were analysed by common chemical procedures according to ČSN, or AOAC (1995), respectively.

After opening the bag, aerobic stability of the silage samples was established according to Ranjit and Kung (2000), in the hours when the silage temperature grew by 3°C above the average laboratory ambient temperature of 20°C. The measurement took 7 days; after that, the samples were analysed with respect to the same indicators as those used for the fermentation quality. The results were evaluated by statistical methods under use of the STATISTICA 10 program (ANOVA at repeated measurements, Tukey's HSD test, correlation).

#### **Results and Discussion**

It can be stated based on the continuous measurement of temperatures inside and outside the bag that after storing the brewers' grains into the bag (9.6.2015), they had a temperature of 60°C. In the subsequent 16 days, the temperature quickly dropped. In a depth of 35 cm under the canvas, the temperature decreased to the final 22°C in 390 hours. Further measurement of temperatures (until 21.9.2015) shows that the brewers' grains have good insulation properties, as the temperatures were almost independent of the outside temperature in a depth of 20 cm already.

Between the first and second sampling (after 14 days), only the decrease of reducing sugars (from 2.68 to 0.25 % in dry matter) was significant (P<0.05); it was most probably caused by yeast. As against the first sampling, the content of nitrogen substances increased significantly (from 22.5 to 29.9 % in dry matter) within 4 weeks, but after that, the increase stopped. A turn in the quality deterioration of the indicators of the nutritional and the energy value of the brewers' grains was registered after two months of storage.

The increasing dry matter (as well as the increasing time of storage) correlated with the increasing content of protein and with the decreasing content of ashes, sugars and hemicellulose in the dry matter. The content of hemicellulose correlated significantly (P<0,05) with NDF. The content of ashes correlated negatively with the content of fibre and ADF.

The worsening fermentation quality was registered on 12.8. While on 21.7., the content of lactic acid was 0.72 %, on 12.8. it was 0.24 %. The content of acetic acid had an opposite development: while on 21.7., it was 0.19 %, on 12.8. it was 0.42 %.

BACK

After opening the silo, the brewers' grains started quickly spoiling, the temperature increased by  $3^{\circ}$ C within 26 hours, then it started decreasing and after 24 hours it equalized the laboratory temperature. The aerobic stability was assessed also by the fermentation indicators. Within 7 days of measurement, the pH values (P<0.05) changed from 3.7 to 5.1, the values of lactic acid from 0.8 to 0.2 %, and those of acetic acid from 0.34 to 0.12 %. We state for comparison that at the beginning of the experiment, i.e. after the storage of fresh brewers' grains in the bag on 9.6., the pH values were 3.75, those of lactic acid 1.4 % and those of acetic acid, 0.2 %. The differences between the initial and final quality at storage in the bag were minimal, which changed substantially immediately after the air was allowed access to the silage. The opening of the bag, i.e. the contact with air had a crucial impact on the quality of the brewers' grains stored - the time of storage had by far lower impact. That confirmed the results of Orosz and Davies (2015) suggesting that if brewers' grains are stored in the short term and without preservatives, they must be fed maximally within 14 days.

Date	pН	Lactic acid (% FM)	Acetic acid (% FM)	NH <sub>3</sub> -N (mg N/100 g)
9.6.	3.76	1.44 <sup>d</sup>	0.17 <sup>a</sup>	8.30 <sup>a</sup>
23.6.	3.73	1.36 <sup>cd</sup>	0.13 <sup>a</sup>	8.30ª
7.7.	3.73	$0.8^{4bc}$	$0.17^{a}$	$8.57^{ab}$
21.7.	3.76	$0.72^{ab}$	0.19 <sup>a</sup>	8.43 <sup>ab</sup>
12.8.	3.72	$0.24^{a}$	0.42 <sup>b</sup>	$7.80^{a}$
24.8.	3.77	$0.40^{ab}$	0.42 <sup>b</sup>	9.60 <sup>b</sup>
8.9.	3.76	0.53 <sup>ab</sup>	0.43 <sup>b</sup>	9.60 <sup>b</sup>
SD	0.03	0.43	0.13	0.75
SEM	0.01	0.09	0.03	0.15

 Table 1
 Changes in indicators of fermentation during storage of brewers' grains

Different letter superscripts within a column indicate statistical differences,  $\alpha = 0.05$ 

Table 2	Aerobic stability of brewers' grains	
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Data	, nII	Lastic soid (0/ EM)	A patie paid (0/ EM)	NUL N $(m \propto N/100 \propto)$
Date	рН	Lactic acid (% FM)	Acetic acid (% FM)	NH <sub>3</sub> -N (mg N/100 g)
21.9.	3.75 <sup>a</sup>	0.81 <sup>b</sup>	0.34 <sup>b</sup>	8.03 <sup>b</sup>
29.9.	5.13 <sup>b</sup>	0.21ª	0.12 <sup>a</sup>	6.28 <sup>a</sup>
D'00 11 11	• , •,•	1 1 1 1 1 1 1 1 1	1:00 0.05	

Different letter superscripts within a column indicate statistical differences,  $\alpha = 0.05$ 

The review of Orosz and Davies (2015) lists a lot of different publications with results of experiments using biological or chemical preservatives. But they cannot be used for comparison with our results, as they describe an older technology of storage of brewers' grains in silage troughs or long plastic sleeves filled under use of pressing machines. The technology used in our experiment is a new one. The bringing of fresh brewers' grains directly from the brewery to the farm and filling it into a bag fixed to the truck body guarantees that the grains are still warm and that no undesirable micro-organisms or air penetrate into the bag.

#### Conclusion

If the brewers' grains are stored in anaerobic conditions, they spoil minimally. They can be stored in a closed plastic bag up to 6 weeks even without preservatives. The indicators of fermentation quality worsened (the acidity of the water extract and the lactic acid decreased, the acetic acid increased) significantly (P < 0.05) only after 6 weeks. As compared to the current practice, the storage period can be extended 3 times (from the recommended 2 weeks to 6 weeks). When the bag was opened, the brewers' grains started quickly spoiling; their aerobic stability was established at 26 hours; all indicators of fermentation quality significantly worsened (P < 0.05) during 7 days of measurement. It must be therefore quickly fed to the animals.

#### Dedication: MZe NAZV QJ1510391

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## Ash Concentrations in Silages can be used to Assess Avoidable Top Layer Losses Due to Aerobic Spoilage in Silos on Commercial Farms

BACK

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#### Introduction

Dry matter (DM) losses in stored silage can be considerable and one of the biggest components is loss in the top layer, which can account for 30 to 150 g/kg of original crop DM ensiled in covered silos (McGechan, 1990). Theoretically, loss due to aerobic spoilage may be eliminated if oxygen is excluded from the top layer by covering the top surface of the silo with an oxygen barrier film (Wilkinson and Fenlon, 2014). Thus, aerobic top layer loss may be considered to be avoidable by best-practice silo management.

Few farmers weigh forage as it is ensiled and most do not weigh all that is removed, making it difficult to quantify storage losses in commercial silos. Previous research has used differences in silage ash concentration at different depths below the top surface of the silo to assess organic matter (OM) losses during the storage period (Bolsen et al., 1993; Bolsen, 1997). Covering silos with plastic film has a large influence on upper layer losses and Bolsen (1997) recorded average OM losses in the top 500 mm on 127 commercial farm horizontal silos of 203 g/kg in covered and 470 g/kg in uncovered silos. The aim of the research reported here was to assess avoidable dry matter losses in the top layer of silos on commercial dairy farms and to present the results in financial terms for use by farmers and advisors.

#### **Material and Methods**

Silage was sampled from 12 walled bunker silos in England in the winter of 2015/16. All crops had been ensiled for at least six months at the time of sampling. Three different types of forage (grass, maize and whole crop wheat silage) were stored in the silos and a wide range of covering and sealing protocols were used. Pairs of samples, each of approximately 500 g, were taken across the exposed feed-out face as a column from the top 200 mm and also from the central core, defined as being 1.5 m below the top layer sample and at least 1.5 m from the edges of the silo. 31 pairs of samples were collected. Each entire sample was dried in a forced air oven for 48 hours as it was not possible to reliably sub-sample silages with variable amounts of visible spoilage. The dried material was ground to pass through a 1.0 mm screen and divided into three parts (each 40 to 60 g), which were ashed at a maximum temperature of 600°C for 14 hours (Sluiter, et al., 2005). The difference in ash concentration between core and top layer within a pair of samples was tested for significance using a two-sample t-test (n=3). For 18 out of 31 pairs the difference was significant (p<0.05). The ratio of ash in the top layer sample to the ash concentration of the core was used to calculate the avoidable loss (g/kg DM) in the top layer. Muck and Holmes (2000) recorded an average core FW density of 637 kg/m<sup>3</sup> for 168 commercial bunker silos filled with forage maize and lucerne and FW density in the top layer was assumed to be 490 kg FW/m<sup>3</sup>, or 0.77 of core density (D'Amours and Savoie, 2005), to calculate avoidable loss of fresh weight (FW) and DM per square meter of top surface. The financial value of avoidable loss per square metre of silo top surface was calculated assuming a value of €120/tonne silage DM.

#### **Results and Discussion**

In 13 pairs of samples (42% of all samples) there was no significant difference in ash content and there was minimal visible spoilage in these silos. Summary statistics for the 18 pairs of samples (58%) with significant differences in ash between the core and top 200 mm of the silos are in Table 1. Median avoidable losses from the top 200 mm per square metre of top surface were 56 kg FW (range 8 to 276 kg) and 19 kg DM (range 2 to 77 kg, Table 1). Ashbell and Weinberg (1992) compared different silo coverings and took 'top-surface' samples varying in depth from 150 to 500 mm; using the above methodology losses ranging from 5 to 360 kg FW were derived from their results.

Avoidable top layer losses translate into financial losses ranging from  $\notin 0.26$  to  $\notin 9.26/m^2$  of top surface (median =  $\notin 2.22$ ). Actual losses are often greater than this as much of the visible silage remaining in top 200 mm maybe judged to be unfit for feeding to livestock and discarded. The technique described here is sufficiently powerful to detect top layer losses as small as 8 kg FW/m<sup>2</sup>.

 Table 1
 Summary statistics for core DM, ash concentrations in the core and the in the top 200 mm, estimated avoidable losses due to aerobic spoilage (FW and DM) and calculated financial losses for 18 paired samples from commercial bunker silos.

	DM of core sample (g/kg)	Ash in core (g/kg DM)	Ash in top 200 mm (g/kg DM)	Avoidable loss in top 200 mm per m <sup>2</sup> of top surface		Financial loss /m² of top surface (€)
				kg FW	kg DM	
Median	290.0	32.3	60.7	56.3	18.5	€ 2.22
IQR <sup>a</sup>	27.5	51.6	88.0	68.6	21.7	€ 2.60
Minimum	270	25	34	8	2	€ 0.26
Maximum	520	93	200	276	77	€ 9.26

<sup>a</sup>Inter-quartile range

#### Conclusions

The results presented in this paper indicate that measuring the ash content of paired silage samples can be used to quantify avoidable losses in the top layer of a bunker silo due to aerobic spoilage. More than half (58%) of samples showed significant losses in the top 200 mm of the silo. Whilst avoidable losses can be considerable (up to 276 kg FW in top 200 mm per square metre of top surface) the technique described here is sufficiently powerful to detect losses down to 8 kg FW/m<sup>2</sup>. Expressing the losses per square metre of silo surface allows comparisons between silos and between different covering protocols. Converting losses into financial terms allows farmers and their advisors to assess losses relative to the costs of possible preventive measures.

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#### Labour Requirement for Barn Dried Hay Production and Feeding on Dairy Farms

**BACK** 

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#### Abstract

The interest of dairy farmers in barn hay drying systems and producing of so-called "hay milk" is growing in the last years in Germany, especially near to the Alps. However, only few data are available regarding the labour requirement for this production system by recently used technique. The aim of this study is to evaluate the labour input for the whole process of barn dried loose hay production (from cutting until storing of the dried hay) as well as for its feeding (and other with feeding connected tasks) on Bavarian dairy cow farms. Up to ten dairy farms with dried hay production will be included in the analysis. The recordings will be performed during several representative periods. Moreover, computer based calculation models should be developed to estimate the labour required associated with hay barn drying production and its feeding. Therefore, selected tasks will be timed on the level of single working elements to determine the standard times. Thereafter, calculation models will be created to estimate the labour requirement for the selected tasks. The standard times together with influence variables will be the basis for these models. At the moment, the study is at the beginning and recordings will just start.

#### Introduction

Hay is the oldest way to conserve forage. However, recently the feeding of silage has prevailed in most dairy farms. The feeding of dairy cows only with hay in order to produce so called "hay milk" has become an interesting alternative to silage feeding and production of "silage milk" in Austria already for several years (Lehnert, 2012) and recently also in Germany (Huber et al., 2015). Nevertheless, the weather conditions are frequently unsuitable to produce hay of high quality by field drying. Therefore, the interest of farmers in barn hay drying techniques increases. For barn hay drying, it is enough to reduce the hay moisture content to 40 % during the field drying process (Wirleitner et al., 2014). Thereby, the time needed for hay drying on the field and the number of turnings can be reduced, and therefore the loss of leaves due to material disintegration decreases and hay quality increases. Due to many influence factors, values of labour requirement vary widely for production of grass silage, field and barn dried hay (Eichhorn, 1999; Schick and Stark, 2002; Ammann, 2007; Diverse authors, 2011). However, in all studies, the time requirement for production of grass silage in bunker silos was lower than for field dried hay; and further, time requirement for field dried hay was lower than for barn dried hay. The potential for lower labour requirement will be seen by labour requirement for barn dried hay feeding in comparison to silage feeding (Neuhofer, 2010; Eilers et al. 2013).

For barn drying systems, the hay can be harvest loose or baled. The loose drying is performed in so-called drying boxes. These boxes are mostly filled up with a hay crane which can be frequently used also for feeding. The production of loose barn dried hay is the system of interest to be studied in this investigation. The aim of the study is to evaluate the labour input for the whole process of barn dried loose hay production (from mowing until the dry hay is stored) as well for its feeding by usually applied techniques on Bavarian dairy cow farms.

#### **Material and Methods**

#### Evaluation of labour input on farms using a work diary

The recording of labour input for barn dried hay production and feeding will be performed using working dairies. During representative periods, all farm operators included in these processes have to record the time needed to perform the tasks of interest daily. For the hay production and harvesting, the labour input will be evaluated during several cuts within one year. The recording of labour input for feeding and other tasks connected with feeding will be performed during two periods of 14 days. Due to the fact that the cows are grazing during summer and feed in the barn during winter, the recording of labour input for feeding will be performed during these two periods (winter and summer). Up to ten dairy farms will be included into the study.

Timing of selected tasks and creation of calculation models for estimation of labour requirement

Computer based calculation models (MS Excel format) will be developed to estimate the labour requirement associated with hay production, harvesting, and feeding. Therefore, for chosen techniques, selected tasks will be timed on the level of single working elements to determine the standard times for these elements. During each recording, influence variables and procurement quantities (volumes, distances etc.) necessary for determination of standard times will be recorded. The standard times together with influence variables will be included in created calculation models to estimate the labour requirement for the selected tasks.

#### **Results and discussion**

Recently we are looking for the farmers with the willingness to partake in the study. Around 30 farmers were contacted and asked for participation. We expect that first measurements will be started in middle of August. The work dairies needed for the recording of labour input were created. In Figure 1, the part of a work diary for recording the labour input for feeding and with feeding connected tasks is shown.

Figure 1	Part of work diar	v for recording of labour i	put for feeding and with feedin	g connected tasks.

		Tasks		Monday	Tuesday	Wednesday
		Orders, booking, bills - feed stuff	1	1	1	1
	~ C	Orders, booking, bills -spare parts, materials	2	2	2	2
tion	atio	Calculation of feed ration	3	3	3	3
nisa	agen	Meetings (feed consultants, between labours)	4	4	4	4
Organisation,	management, administration	Other PC work connected with feeding	5	5	5	5
0	⊏ ຫ	Futher education, seminars, visits	6	6	6	6
		Miscellaneous	7	7	7	7
		Feed preparation, restoring, concentrat mixing	8	8	8	3
		Forage or TMR feeding	9	9	9	ç
		Concentrat feeding	10	10	10	10
		Green fodder feeding (harvesting, transport, feeding)	11	11	11	11
	ß	Pushing up feed	12	12	12	12
	Feeding	Cleaning of feed table, removal of feed remains	13	13	13	13
	Fe	Pushing of feed remains to calves/young cattle	14	14	14	14
	Feed stuff delivery and storage	Feed stuff delivery and storage	15	15	15	15
6		Silo (uncover, cover, removal of remains)	16	16	16	16
cows		Sampling of feed	17	17	17	17
õ		Miscellaneous	18	18	18	18
		Animal traffic: pasture - milking and back	19	19	19	19
		Animal traffic: barn - pasture, between pastures	20	20	20	20
		Animal control	21	21	21	21
	sing	Supply of water	22	22	22	22
	Grassing	Supplementary feeding at pasture	23	23	23	23
	Ŭ	Preparing and repairing of fences, preparing of pasture	e <sup>24</sup>	24	24	24
		Pasture maintenance	25	25	25	25
		Miscellaneous	26	26	26	26
	ore	Feeding of milk or replacement, watter supply	27	27	27	27
	calves before weaning	Roughage feeding	28	28	28	28
	ves veai	Concentrat feeding	29	29	29	29
	ca	Miscellaneous	30	30	30	30

#### Acknowledgement

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## Influence of Soil Conservation Technologies of Maize Seeding on Yield and Forage Quality

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#### Introduction

Maize (*Zea mays*) is still considered a significant fodder, foodstuff and industrial plant both in the Czech Republic and worldwide (Zimolka 2008; Yu et al. 2016) which is grown on arable land. Although global agricultural output has improved dramatically over the past 50 years, future demand for maize as food, feed, and bioenergy resource will increase tremendously due to the growing population (Tilman et al. 2002; Godfray et al. 2010). The total area of crop acreage of maize in the CR in 2015 was 324 928 ha (Czech Statistical Office 2015). Although maize for green and silage covers the same area as in 2000, the area of maize grown as bioenergy resource has increased at the expense of fodder. The disadvantage of maize production is its proneness to soil erosion especially on sloping lands (Basic et al. 2004). With respect to seeding procedure maize belongs among crops which are energy accumulators and it decreases soil fertility due to loss of organic matter in soil, soil structure deterioration, and reduction of soil biological activity, etc. (Hanegraaf et al. 2009; Brown et al. 2014). It is an urgent task for agricultural scientists and farmers to find the best agricultural practice and sustain maize productivity while improving environmental quality (Yu et al. 2016). This paper examines the effects of soil conservation technologies maize seeding of on yield and forage quality (conservation technologies of seeding into rye (*Secale cereale*) stubble, i.e. to minimize tillage).

#### Material and methods

Site: In 2015 at the Velke Opatovice site (average annual temperature 7.4 °C, annual long-term rainfall average 545 mm, altitude 360-380 m) on a mildly sloping plot with southern orientation, a pilot experiment was established to test soil conservation technologies (SCT) of maize seeding on arable land. The previous crop (rye) was harvested in May 2015 for green matter in the early growth phase of earing. Maize was seeded in mid-May into a) rye stubble (treated with a system herbicide with glyphosate at the rate of 6  $l.ha^{-1}$ ; b) minimum-tilled rye stubble (stubble tillage + preseeding preparation). A very early maize hybrid (FAO 220) suitable for biogas silage was used. The seed rate was 95 thousand germinating seeds per hectare (wide row - spacing of 75 cm), 115 thousand germinating seeds (narrow row spacing 37.5 cm) respectively. The experiment treatments: (a) SCT seeding into rye stubble with: 1) Monosem; 2) *Kinze*; 3) *PS*-2; 4) *PS*-2 + *Kuhn Striger*; 5) *Kuhn* + *Kuhn Striger*; (b) SCT seeding into minimum-tilled rye stubble with: 1) Monosem; 2) Kinze; 3) Kuhn. Treatments' description: 1) Monosem model NG 8R Plus), an 8-row planter with a down pressure seeding system and seed discs for precision seeding, row spacing 75 cm; 2) Kinze (model 3500 -Interplant), adjustable (8/15 rows) disc planter with pneumatic down pressure seeding unit, row spacing 75 cm, resp. 37.5 cm; 3) PS-2, a prototype of seeding drill – 2-row active rotary cutters, row spacing 75 cm, working width of rotor 150 mm, adjustable cutting depth 50-150 mm, seeding shoe with a mechanical seeding unit for precision seeding; 4) PS-2 in combination with soil preparation by deep cultivation of rye stubble with 8-row Kuhn Striger machine (subsoilers with soil looseners, soli cultivation 25 cm deep in strips 15-25 cm wide and strip spacing 75 cm; 5) Seeding drill Kuhn (model Maxima 2 RT) in combination with deep cultivation with Kuhn Striger machine (precision 8-row seeding drill, down pressure seeding units, row spacing 75 cm). Fertilization: organic fertilizer digestate, rate 30 m<sup>3</sup>.ha<sup>-</sup> <sup>1</sup>; mineral fertilizer urea (90 kg.ha<sup>-1</sup> N) and Eurofertil Top 45 NPS (6 kg.ha<sup>-1</sup> N and 44 kg.ha<sup>-1</sup> P). Harvest: the end of September, self-propelled forage harvester with an 8-row adapter. Sampling: harvested maize samples were evaluated for yield parameters and forage quality parameters were determined after mechanical processing (cutting and homogenisation with a stationary cutter) using AgriNIR<sup>TM</sup>, a portable analyser of voluminous feedstuff.

#### **Results and Discussion**

Plant growth and development of maize were significantly affected during the growing season by rainfall deficiency and high air temperatures with frequent occurrence of tropical days when average daily temperature did not fall under 20 °C. The rainfall deficit during the growing season was 83.2 mm at the Velke Opatovice site and its uneven distribution throughout the summer and prior to the harvest had a negative impact on the yield of maize from the observed experimental plots. The highest dry matter production from SCT treatments was detected in a prototype planter PS-2 treatment (8.05 t.ha<sup>-1</sup>) and a planter Kinze 3500 treatment (8.04 t.ha<sup>-1</sup>) - *Tab. 1*. Deep cultivation of rye stubble with Kuhn Striger machine in combination with planters PS-2 or Kuhn did not demonstrate a positive effect of soil conservation technologies on plant development and yield increase. From the treatments with minimized tillage the highest dry matter production was reached in Kinze treatment (9.04 t.ha<sup>-1</sup>), which is 31 % higher yield than in PS-2 treatment with deep rye stubble cultivation. This also demonstrated positive impact of the increased seed rate (115 thousand germinating seeds) in combination with seeding in row spacing 0.375 m. Differences in dry matter production of evaluated treatments of seeding into rye stubble and into minimum-tilled soil were not distinctive except for Kinze treatment (seeding into minimum-tilled soil) and they were around 108–117 % of the treatment PS-2 + Kuhn Striger).

	Soil preparation prior to / minimizing technologies								
Evaluated features		l	Rye stubble			Minin	num-tilled	soil	
Evaluated realifies	Striger + PS-2	Striger + Kuhn	Monosem	Kinze	PS-2	Monosem	Kuhn	Kinze	
Growth height (m)	1.48	1.52	1.55	1.60	1.51	1.80	1.63	1.88	
Number of plants per ha (thousand pcs)	77.3	78.2	89.3	96.0	90.7	87.1	77.3	95.1	
Grain dry matter at 105 °C (%)	63.3	62.4	62.0	62.8	61.8	60.4	60.6	64.2	
Green matter production (t.ha <sup>-1</sup> )	22.64	23.30	22.77	25.77	25.80	24.92	23.53	27.72	
Dry matter of green matter at 105 °C (%)	30.5	30.6	33.9	31.2	31.2	29.8	32.8	32.6	
Dry matter production (t.ha <sup>-1</sup> )	6.91	7.13	7.72	8.04	8.05	7.43	7.72	9.04	
Relatively	100	103	112	116	117	108	112	131	
NEL production (MJ.ha <sup>-1</sup> )	41 736	43 636	46 629	48 964	48 542	45 249	47 169	55 054	

 Table 1
 Results of pilot experiment with minimizing technologies of maize seeding (Velke Opatovice, 2015).

Footnote: Minimizing technologies are listed in ascending order according to dry matter (t.ha<sup>-1</sup>), separated by soil preparation method.

In comparison with other technologies Kinze minimizing technology with both methods of soil preparation prior to seeding reached higher number of plants per ha (96 thousand pcs in treatment A, resp. 95.1 thousand pcs in treatment B), growth height (1.60 m, resp. 1.88 m), higher green matter production (25.77 t.ha<sup>-1</sup>, resp. 27.72 t.ha<sup>-1</sup>) and NEL production (48 542 MJ.ha<sup>-1</sup>, resp. 55 054 MJ.ha<sup>-1</sup>). The technology of strip seeding of maize with PS-2 into rye stubble reached similar yield as the Kinze technology even with a lower number of plants per ha at harvest time.

#### Conclusion

The attained results demonstrated that the maize seeded by soil conservation technologies can have comparable yields to the maize seeded into minimum-tilled soil, but with lower energy demands. Strip seeding with deep cultivation (subsoiling) of a soil profile to the level of seedbed fully replaces traditional tillage technology, see Nerušil et al. (2015). Due to adverse climatic conditions in 2015 higher yield was not clearly shown.

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# Increasing of Effective Fiber at TMR by new Technology (Shredlage) for Harvest of Maize Silage

BACK

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#### Introduction

Optimal harvest time of maize for corn silage is for the whole plant in the range of 30-34% of DM. In this period it must be maize harvested in a way to ensure a successful fermentation process of ensiled forage with the minimum losses of DM during fermentation and storage at silo. Self-propelled forage harvestor was equipped with numerous knife cutter drum that was able to cut the maize and secure good fermentation process, but this device was unable to disrupt the chopped forage length wise, not sufficiently disrupt grain corn. For this reason, gradually cutting drum fitted with other blades that allow TLC shortened. Furthermore, the forage harvester gradually fitted grooved rollers so. Corn Cracer, which were supposed to squeeze the grain of corn, which was subsequently used in the rumen. However, there was the fact that the resulting corn silage at an optimum dry matter 32% TLC was below 10 mm. Such forage reduced the proportion of effective fiber in TMR (Mertens D.R. 1993, 2000, Kudrna V., 1998), which stagnated in milk production deterioration of health, especially in high-yield dairy cows. Shorter chop corn silage slowed chewing of cows (Gleaves, E. W., 1973, Harris, B. 1984, Milligan, R. A., 1981), reduces the fat content and protein content in milk. Further causes increased acidification of the rumen fluid especially when increasing proportion of concentrated feed for highly productive dairy cows. This situation reduces the pH in the rumen of cows under physiological limit of pH 5.8, which inhibits bacterial activity particularly cellulolytic bacteria and associated suppression recovery fiber (Koukolova V., et al.2004). Of those envisaged for the gradual share of maize silage in the TMR decreased and effective fiber in TMR comes in the form of chopped straw. Long term we wanted to develop a technology that would allow extending the length of cut in harvested forage, particularly maize, provided security fermentation process. Last year we focused on SHREDLAGE technology, which was developed in recent years in the US and license the technology acquired last year, CLAAS. Machines equipped with this technology it is possible this year already bought in the Czech Republic.

#### **Material and Methods**

In experiments, we focused on the quality of the fermentation of corn silage (one maize hybrid, laboratory minisilo 4 l at 25<sup>0</sup> C and fermentation 60 days) with TLC 8 mm harvested forage harvestor Claas Jaguar 690 and second silage TLC 30 mm harvested self-propelled Claas Jaguar 690 by technology Shredlage, compressibility of silage (calculating specific gravity produced silage kg.m3) assessment, the share of individual fractions on the Penn State Particle Separator (PSPS). Degradability of Organic Matter (DOM) and Degradability of Neutral Detergent Fiber (DNDF) by in sacco method in the fistule rumen of dairy cows (McDonald I. 1981). Samples of maize silage was grinded at sieves 1 and 5 mm

#### **Results and Discussion**

At the table 1 you can see results of fermentation process of maize silage harvested by Shredlage technology (TLC 30mm) and clasic method with TLC 8 mm. Results of fermentation process control groupe and trial groupe is same. Different TLC of maize have not any effect for results and we can say that both silage was well conserved.

At follow table 2 you can see of specific weight of maize silage harvested at TLC 8 mm and TLC 30 mm. Specific weight of maize silage harvested TLC 30 mm (583 kg.m<sup>3</sup>) have higher weight about 47 kg.m<sup>3</sup> than TLC 8 mm silage (536 kg.m<sup>3</sup>). This fact s very important for silage with TLC 30 mm because higher specific weight cause better compressibility for longer TLC (30 mm) compared to shorter TLC (8 mm). This effect we can explane fact that TLC silage is prepare except numerous knife cutter drum also by corn cracer which have special wheelling for lengthtways of stover and destroyed of corn for small parts. This fact s very important for fermentation proces because during tamping is air better expression from silo and first aerobic fhase is shorter for this maize silage.

At the table 3 you can see results of rumen Degradability of Organic Matter (DOM) and Degradability of Neutral Detergent Fiber (DNDF) which we measure by in sacco method. Results of DOM and DNDF are similar for bouth treatment at experiment. We can not see difference. According this results we can not used in vitro method (in sacco) for determination of quality OM and NDF. Important point of this results is that for in vitro method samples of silages we have to grind for 1 mm and this sample is weigh to the nylon bags at amount 2 g of DM. This samples at nylon bags are incubated at fistule cow for 24 hours.

Except effect of degradability OM and NDF we tested effect of the Penn State particle Separator (PSPS) at bouth silages. Results you can see at follow table 4. According results at table 4 is very important that fraction 19 mm sieve is more higher (15,8 %) at TLC 30 mm compared to TLC 8 mm (5,4 %).

N = 6	TLC 8 mm	TLC 30mm
DM %	36,48	36,30
Soluble DM %	19,45	17,22
pH	3,95	3,97
Lactic acid %	2,40	2,67
Acetic acid %	0,63	0,86
Propionioc acid %	0,08	0,08
Total VFA %	0,71	0,94
LA/VFA	3,38	2,84
Crude Protein % at DM	8,34	8,19
Starch % at DM	36,65	35,46
Crude Fiber % at DM	16,68	18,30
NDF % at DM	44,36	44,37
Ash % at DM	3,14	3,29

 Table 1
 Characteristics of fermentation process of maize silage harvested by different technology.

TLC - Theoretic Long Cut, DM - Dry matter

Table 2 Specific weight of maize silage harvested by TLC 8 mm and TLC 30 mm

N = 6	TLC 8 mm	TLC 30 mm
Specific Weight kg.m <sup>3</sup>	536	583

TLC - Theoretical Long Cut

**Table 3** Degradability of maize silage harvested for TLC 8 mmm and TLC 30 mm.

N = 6		TLC 8 mm	TLC 30 mm
DOM 24 hours	1 mm	67,32	63,32
	5 mm	61,20	57,94
DNDF 24 hours	1 mm	43,96	42,31
	5 mm	34,51	30,81

TLC - Theoretical Long Cut, DOM degradability of Organic Matter, DNDF - Degradability of Neutral Detergent Fiber

Table 4 Individual fractions on the Penn State Particle Separator (PSPS).

N = 6	TLC 8 mm	TLC 30 mm
19 mm % from total 500 g	5,4	15,8
8 mm % from total 500 g	78,2	56,6
Bottom % from total 500 g	16,4	27,6

TLC - Theoretical Long Cut, DOM degradability of Organic Matter, DNDF - Degradability of Neutral Detergent Fiber

#### CONCLUSION

Results of this experiments showes that new technology for harvest of maize (SHREDLAGE) is good way for preparing maize silage with good quality of fermnetation proces, increase of specific weight and increasing longer fraction on Penn State Particle Separator. But this kind of silages with different TLC we can not determinate by in sqcco method for measuring degradability at rumen.

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