**Original Paper** 

# Analytical methods in dairy cows nutrition and their application in creation of production health

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The effect of quantity of nutrients on rumen fermentation and the level of metabolic markers in blood serum were simultaneously analysed in groups of dairy cows 21 days before and 21 days after parturition with the aim to diagnose disorders in milk production in the transition period of dairy cows. Results of analysis of health disorders confirmed the following: low energy concentration in the diet insufficiently saturated with fibrous carbohydrates, followed with rapid change to concentrate type of diet after delivery resulted in insufficient adaptation of the rumen metabolism before and after rapid transition to production feeding rations after calving; the level indicative of acidification of the rumen environment. Investigation of intermediary metabolism confirmed pre- and post-partum lipomobilization, with increased values of NEFA in 68 % and 54 % of animals respectively, with liver load manifestation in 37 % and 69 % of animals, respectively.

Keywords: lipomobilization, non-esterified fatty acid, rumen, intermediary metabolism

## 1 Introduction

The transition period of the cow is generally defined as 3 weeks before to 3 weeks after calving. During this period the cows undergo large metabolic adaptation in glucose, fatty acids and mineral metabolism to support lactation and avoid metabolic dysfunction (Overton and Waldron, 2004) with negative energy balance. This reflects the reduction in dry matter intake and the immune system during the transition period increases the risk for fatty liver, displaced abomasum, retained placenta, metabolic disorders such as ketosis and milk fever, and infectious disease such as mastitis and metritis (Allen and Bradford, 2008). Energy requirements for maintenance and pregnancy during the last month of pregnancy increased and mammary requirements for glucose, amino acids and fatty acids for a milk yield of 30 kg d<sup>-1</sup> at 4 days postpartum are approximately 2.7, 2.0 and 4.5 times those of the gravid uterus during late pregnancy (Drackley et al., 2001). The nutrition goal of transition cow management is to ease the transition from pregnancy to lactation by optimizing health and improving long term milk yield, reproductive success and farm profitability. Nutritional optimalization of diet will help us to control fat mobilization and increase feed intake, improving animal health and increase profitability of dairy farms (Meijer and Peeters, 2010).

The aim of our study was to evaluate the influence of nutrition on production and health of dairy cows in their pre-partum phase (21 days before and after delivery) in production herds and parallel determination of ingestion of nutrients from total mixed ration (TMR) in relation to evaluation of rumen fermentation level and metabolites in blood serum of dairy cows and to propose procedure for biological control of nutrition, adaptation of rumen and intermediary metabolism in transition period of highly-productive dairy cows.

## 2 Material and methods

In production conditions of overall 10 herds of dairy cows with the production of 6–8 thousand litters per year, analysis of the level of nutrition and degree of rumen adaptation, as well as intermediary metabolism in the peripartal phase of dairy cows breeding was carried out. In each of the evaluated herd was analysed: – nutrients composition of feed and TMR, analysis of rumen fermentation level and markers level of protein intermediary metabolism, energy metabolism and markers of liver function. In both pre and postpartal phase were evaluated in relation to the nutrition level and fermentation as reflection of nutritionally influenced adaptation during the transition phase of dairy cows breeding.

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# 2.1 Analysis

Chemical analysis of nutrients in samples of TMR were analyzed for dry mater, crude protein (CP), acid and neutral detergent fibre (ADF, NDF) and starch according to conventional methods (Committee regulation ES No.152/2009 of 27.1.2009). Non-fibrous carbohydrates (NFC) was calculated by difference (100 - (CP + (NDF - NDF bound protein) + ash + ether extract)) and energy calculated by regression (Linn et al., 1989, NRC 2001).

**Analysis of VFA and NH<sub>3</sub>.** Samples of rumen content intended for analysis of fermentative and synthesizing capacity of the rumen were taken 4–6 hours after morning feeding by stomach cannulas and stabilised by thymol for conservation of sample. Samples of rumen fluid were strained through 4-layer of gauze, centrifuged 25 min and diluted 1 : 50. VFA in the rumen content were determined in a two-capillary isotachophoretic analyser EA 100 (VILLA LABECO, Slovak Republic). The pH of the rumen content was determined potentiometrically with portable electronic pH-meter (JP SELECTA, Spain). The level of NH<sub>3</sub> in the samples of rumen content was determined by a Kjeldahl-N using a 2300 Kjeltec Analyser Unit (Foss Tecator AB, Hoganas, Sweden).

Each parameter was presented by its mean ( $\bar{x}$ ), standard deviation (s. d.), respectively.

# 3 Results and discussion

The investigation of number of samples of TMR, conducted in dairy cows in the prepartum phase and at the peak of lactation, focused on nutritional parameters including the content, digestibility and energy content of TMR and its influence on rumen fermentation and energy metabolism in dairy cows provided results summarised in Tables 1–3.

The analysed level of nutrients in TMR (Tab. 1) reflected decrease in energy concentration in comparison with the recommended values. Increased levels of neutral detergent fibres (NDF) in dairy cow rations prior and after the parturition with mean level of NDF reaching 411.3  $\pm$ 68.0 and 361.1  $\pm$ 41.0 and 335.6  $\pm$ 47.0 g kg<sup>-1</sup> dry matter (DM).

The content of non-fibrous carbohydrates (NFC) in TMR DM reached on average  $355.1 \pm 41.5$  g kg<sup>-1</sup> before parturition and  $381.8 \pm 7.0$  g kg<sup>-1</sup> DM after parturition. This confirmed that an increased proportion of concentrate feed was supplied to the cows to compensate for the low quality of bulk feed.

State of rumen fermentation directly reflects the composition of feed rations with prevalence of structural carbohydrates and limited portion of NFC before parturition.

In dairy cows in their preparation for delivery with considerable proportion of clinically healthy animals, the average values of the analysed parameters (Table 2) varied within the limits of the reference values. The mean level of pH was below the reference value as were the individual results in half of the examined dairy cows after calving. The variability of individual VFA values, increase in propionic acid level in 44 % of animals, decreased level of acetic acid in 41 % of them and a tendency of  $C_2 : C_2$  decline to the lower limit confirmed the tendency towards rumen acidification as a consequence of low level of adaptation and rapid transition to the productive type of the feeding rations. In the examined animals, propionic acid level before delivery reached in average 17.9  $\pm$ 7.4 mmol l<sup>-1</sup> and individually, in 65 % of dairy cows, reached levels < 20 mmol l<sup>-1</sup>. Such state of rumen fermentation fails to ensure sufficient production of precursors for dairy cows glucogenesis before delivery. Starch is converted in the rumen to propionic acid and supports development of rumen papillae and proliferation of amylolytic microflora (Owens et al., 1998). Propionic acid as product of microbial fermentation in the rumen is a primary fuel controlling feed intake in ruminants fed concentrate diets. It is a primary end product of starch fermentation and production rates vary greatly among diets.

Parameters	$\frac{NDF}{\overline{x}\pms.d.}$	NFC $\overline{x} \pm s.d.$	NEL $\overline{x} \pm s.d.$	$\begin{array}{l} ADF \\ \overline{x} \pm s.d. \end{array}$
21 days before parturition (ap)	32.0-38.0%	30.0-35.0 %	6.2–6.5	23.0-25.0 %
	41.1 ±6.8	35.5 ±4.2	6.23 ±0.3	25.8 ±4.5
	60 % ↑	45 % ↑	45 % ↓	50 % ↑
21 days after parturition (pp)	30.0-35.0 %	32.0–38.0 %	6.6–6.8	21.0-23.0 %
	36.1 ±4 1	38.1 ±3.9	6.72 ±0.1	23.0 ±3.8
	60 % ↑	50 % ↑	50 % ↓	35 % ↑
Peak lactation	28.0-33.0 %	36.0-42.0 %	6.9–7.1	20.0-22.0 %
	33.5 ±4.7	39.8 ±4.0	6.76 ±0.3	20.4 ±3.7
	40 % ↑	30 % ↑	45 % ↓	20 % ↑

Table 1	Carboh	vdrate	com	nosition	of	dairv	cows	rations
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Index	21 ap	21 рр
	$\overline{x} \pm s.d.$ n = 60	$\overline{x} \pm s.d.$ n = 60
рН	6.72 ±0.3	6.18 ±0.4
NH <sub>3</sub> (mg 100 ml <sup>-1</sup> )	17.8 ±5.9	19.8 ±7.3
VFA (mmol l <sup>-1</sup> )	89.5 ±21.9	100 ±21.9
Acetic acid (C <sub>2</sub> ) (%)	67.4 ±4.4	61.0 ±16.1
Propionic acid (C <sub>3</sub> ) (%)	19.6 ±4.6	24.6 ±4.5
Ratio $C_2: C_3$	1:3.6	1 : 2.6

**Table 2**Analysed values of rumen fermentation in dairy cows

Table 3	Analysed serum values of nutrient metabolism in dairy co	ows
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Index	21 ap	21 pp
	$\overline{x} \pm s.d.$ n = 60	$\overline{x} \pm s.d.$ n = 60
Total proteins (g l-1)	69.2 ±6.9	73.9 ±7.8
Albumin (g l-1)	33.5 ±2.6	29.4 ±3.6
NEFA (mmol l <sup>-1</sup> )	0.49 ±0.2	0.56 ±0.4
Glucose (mmol l <sup>-1</sup> )	3.56 ±0.8	3.02 ±0.8
Triglycerides (mmol I <sup>-1</sup> )	0.49 ±0.3	0.36 ±0.2
Cholesterol (mmol l-1)	2.68 ±1.4	2.7 ±0.7
Bilirubin (μmol l <sup>-1</sup> )	4.69 ±2.3	5.43 ±2.3
AST (µkat l-1)	0.41 ±0.1	0.50 ±0.1

The levels of blood serum indices of intermediary metabolism of proteins, carbohydrates and fats, as well as metabolic load on the liver of dairy cows in their prepartum phase are summarized in Table 3. Concentrations of non-esterified fatty acids (NEFA) and  $\beta$ -hydroxybutyrate (BHBA) showed to the adaptation to negative energy balance. NEFA as markers of lipomobilsation reflects the magnitude of mobilization of fat from storage. BHBA indicates the completeness of oxidization ("burning") of fat in the liver (Drackley, 2010).

The observed pre-partum level in dairy cows restricts the supply of glucogenic sources for saturation of foetus needs and evokes the state of lipomobilization in prepartum metabolism of dairy cows.

Propionic acid absorbed at very high rates and is very rapidly taken up by the liver, where it is a major fuel used to produce glucose (Allen and Bradford, 2008), which is needed to increase insulin and decrease NEFA. The manipulation of rate of propionic acid production includes changes of diet. This way supported production of propionic acid, a precursor of glucose synthesis, stabilizes the energetic balance and limits lipomobilization (Van Knegsel et al., 2007). When dividing animals according to the level of propionic acid in the rumen content, the degree of lipomobilization with mean value of NEFA equal to  $0.38 \pm 0.16$  corresponded to the group with level below 20 mmol I<sup>-1</sup> while at reference values of propionic acid above 20 mmol I<sup>-1</sup> the values of NEFA reached the level of 0.31 ±0.1 mmol l<sup>-1</sup>. In the animals with decreased levels of propionic acid in the rumen, manifestation of lipomobilization with values of NEFA >0.35 mmol l<sup>-1</sup> was detected in 42 % of dairy cows while in animals with propionic acid exceeding 20 mmol l<sup>-1</sup>, manifestation of lipomobilization was observed in 18.3 % dairy cows prepartum. The influence on lipomobilization of propionic acid as the most important precursor of glucogenesis in relation to serum level of NEFA was also confirmed by direct regression relationship (r = 0.447). Diets should be formulated to decrease plasma NEFA concentration and maintain rumen fill throughout the transition period. The prevalence of cows with elevated concentrations of blood NEFA or BHBA increases, reproductive performance declines (Ospina et al., 2010).

Feeding diets containing higher proportions of NFC should promote ruminal microbial adaptation to NFC levels typical of diets fed during lactation and provide increased amounts of propionate to support hepatic gluconeogenesis and microbial protein to support protein requirements for maintenance, pregnancy and mammogenesis (Varga, 2007). Increase propionate concentration in the rumen promotes insulin secretion (Harmon, 1992), because insulin is antilypolytic, an increase in dietary NFC might decrease plasma NEFA and reduce metabolic load of liver. Such state of rumen fermentation does not secure sufficient production of precursors for dairy cows glucogenesis before delivery. The major substrates for hepatic gluconeogenesis in ruminants are propionate from ruminal fermentation, lactate and amino acids from protein catabolism. Several studies have evaluated the impact of manipulating the energy density of the prepartum diet on postpartum performance (Santos et al., 2012).

# 4 Conclusions

Nutrition and management during the pre-partum phase are essential in determining the profitability of the cow for the rest of its lactation. Proper formulation of rations regarding proteins, energy density, NDF and NFC will help to increase DMI around calving along with management of body condition while excellent quality, nutritional value and digestibility of forages will assure an excellent programme for the high producing dairy cows. Further increase in milk yield and desirable milk components requires nutritional prevention and management of productive health of herds. This should be implemented through increased intake of forage dry matter and stabilization of rumen and intermediary metabolism of high-productive dairy cows in respective phases of their nutrition.

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# References

ALLEN M.S. and BRADFORD B.J. (2008) *Optimizing Transition Cow Diets. Tri-State Dairy Nutrition Conference, April 22 and 23, 2008, Grand Wayne Center, Fort Wayne, Indiana, pp. 37–44.*  DRACKLEY, J. K, OVERTON, T. R. and DOUGLAS, G. N (2001) Adaptations of glucose and long-chain fatty acid metabolism in liver of dairy cows during the periparturient period. *J Dairy Sci*, 84(E Suppl):E100-E112, doi:http://dx.doi.org/10.3168/jds. S0022-0302(01)70204-4

DRACKLEY J.K. (2010) Feeding transition cows: An apdate and re-interpretation. Minnesota dairy health conference 2010, St. Paul, Minnesota United States of Minnesota, pp. 28–40. [Online]. Retrieved November 14, 2015 from https://conservancy.umn. edu/bitstream/handle/11299/57203/Drackley.pdf?sequence=1

HARMON D. L. (1992) Dietary influences on carbohydrates and small intestinal starch hydrolysis capacity in ruminants. *J. Nutr.*, vol.122, no.1, pp. 203–210.,

MEIJER R. and PEETERS K. (2010) The use of precision dairy farming in feeding and nutrition. The first north American conference on precision dairy management 2010 [Online]. Retrieved November 14, 2015 from http://www. deboerenveearts.nl/wp-content/uploads/2014/02/Meijer\_The-Use-of-Precision-Dairy-Farming-in-Feeding-and-Nutrition.pdf

OSPINA, P.A. et al. (2010) Association between the proportion of sampled transition cows with increased nonesterified fatty acids and  $\beta$ -hydroxybutyrate and disease incidence, pregnancy rate and milk production at the herd level. *J. Dairy Sci.*, vol. 93, pp. 595–3601. doi:http://dx.doi.org/10.3168/jds.2010-3074

OVERTON, T. R. and WALDRON, M. R. (2004) Nutritional management of transition dairy cows: Strategies to optimize metabolic health. J. Dairy Sci., vol.87, pp.105–119. doi:http://dx.doi.org/10.3168/jds.S0022-0302(04)70065-X

OWENS, F. N. et al. (1998) Acidosis in cattle: A review. J. Anim Sci., vol. 76, no.1,pp. 275–286. doi:http://dx.doi. org//1998.761275x

SANTOS J.E.P. et al. (2012) Impacts of Metabolism and Nutrition During the Transition Period on Fertility of Dairy Cows. High Plains Dairy Conference, Amarillo, Texas, pp. 97–112.

KNEGSEL Van, A. T. M. et al. (2007) Dietary energy source in dairy cows in early lactation: Metabolites and metabolic hormones. *J. Dairy Sci.*, vol.90, no. 3, pp. 1477–1485. doi:http:// dx.doi.org/10.3168/jds.S0022-0302(07)71633-8

VARGA G.A. (2007) Dietary energy density for the close-up. Dry cow – postpartum performance. Florida Ruminant Nutrition Symposium, January 30–31, 2007, Best Western Gateway Grand, Gainesville, Florida, pp. 1–14.