# Effects of a monensin controlled release capsule on reticulorumen temperature and pH determined using real-time monitoring in fresh dairy cows

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Abstract: Monensin has been shown to decrease mortality and morbidity in feedlot cattle by reducing the incidence of acute and subacute rumen acidosis (SARA). Lately, the technique of real-time observance of reticuloruminal temperature and pH, which involves placement of indwelling pH probes in the reticulum or in the rumen has gained in popularity. In this study, we aimed to determine, using real-time monitoring in fresh dairy cows, how monensin controlled release capsules affect the reticulorumen temperature and pH. We used a slow-release rumen preparation, which emitted daily monensin doses of 335 mg. Following the manufacturer's instructions, the preparation was administered through an oral balling gun. The following points indicate the method for determining the two experimental groups: (1) monensin-supplemented test group (TG) (a 32.4 g monensin controlled release capsule, MCRC, n = 20) and (2) a control group (CG) (capsule containing no monensin, n = 20). Both began a day after calving, and one month after calving the experiment was finished. A set of smaXtec boluses fabricated for animal care was used to measure the temperature and pH of the reticulorumen. After the first day of the study, a statistically significant difference between reticulorumen temperature in CG (38.67 ± 0.10 °C) and TG (39.08  $\pm$  0.09 °C; P < 0.01) was found. The use of the monensin controlled release capsule, which emitted daily monensin doses of 335 mg, in the first 30 days after calving increased the reticulorumen pH by 1.89% (P < 0.001), and the temperature of the reticulorumen by 0.82 % (P < 0.001). We conclude that using monensin in the form of monensin controlled release capsules reduces the risk of SARA. Real time observation of temperature and pH levels in the reticulorumen in fresh dairy cows allows for evaluation of the risk of SARA and provides the opportunity to determine the prophylactic effect of those capsules.

Keywords: cow; post-partum; diseases; SARA

Rumen health and modification of the metabolic processes and gene networks in the liver, which is an essential metabolic organ, are some of the most important factors for cow health and productivity

during early lactation (Drong et al. 2016). The term sub-acute ruminal acidosis (SARA) tends to be used synonymously with unhealthy rumen (Humer et al. 2018). Feed additives, predominantly ionophore an-

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tibiotics, are mostly used with the aim of increasing meat production, preventing disease or decreasing its occurrence, reducing the age at slaughter and therefore improving the quality of animal-derived food (Zawadzki et al. 2011). Monensin supplementation results in augmented ruminal propionate production (Van Maanan et al. 1979) and retention of nitrogen, improved digestibility of starch and dry matter (DM), a substantially enhanced amino acid flow and digestion in the duodenum, decreased in vivo and in vitro methane production as well as of bacterial proteins in the rumen. No changes in the ratios of acetate to propionate in dairy cattle receiving monensin supplementation was observed by Benchaar et al. (2006) and Mathew et al. (2011). Mortality and morbidity in feedlot cattle decreased as well when these were fed monensin, as it decreases the incidence of bloat, acute and sub-acute ruminal acidosis and bovine subcutaneous emphysema (Callaway et al. 2003). The administration of monensin controlled release capsules results in increased ruminal propionate production, and the prevalence of clinical or subclinical ketosis is decreased (BHB > 1.2 mmol/l) by 53%. Moreover, according to Drong et al. (2016), the capsule improves post-parturient hepatocyte functions in female cattle with good body condition. The ionophore monensin reportedly has various advantageous effects in ruminants. In dairy cattle, the beneficial effects encompass a lower incidence of ketosis and displaced abomasum, prevention of body condition loss, augmented production of milk and increased efficiency of milk production (McGuffey et al. 2001). The capacity of monensin to selectively inhibit Gram-positive over Gram-negative bacteria that reduce succinate to propionate allows for greater energy efficiency (McGuffey et al. 2001). Monensin may be successfully used to reduce CH4 production in ruminants, especially in intensive systems, as it has been demonstrated to increase propionate to acetate ratios and to decrease the numbers of hydrogengenerating protozoa in the rumen (Beauchemin et al. 2008). Monensin included in the diet enhanced total digestive tract fibre digestion, which indicates that monensin supplementation results in enhanced fibre digestion at post-ruminal sites. The aforementioned findings suggest potential for using monensin to improve nutrient digestion in female dairy cattle during grain-induced SARA (Osborne et al. 2004).

As Gasteiner et al. (2009) stated, the state-of-theart for determining reticuloruminal pH involves the placing of indwelling pH probes in the form of a bolus in the reticulum or in the rumen for continual observation of reticuloruminal pH values. The benefits of this technique lie in the possibility it provides for diurnal recording. On the other hand, the elimination of the chip (Penner et al. 2007) or transmission by cable to an external unit attached to the anima, are prerequisites for collecting the data. Gasteiner et al. (2009) have given a description and an evaluation of a wireless unit for data transmission with which continual measuring of reticuloruminal pH and consequently, a prolonged examination may be performed. In order to enhance the detection quality for digestive disorders at the level of an individual cow and at the level of the whole herd, the technique could be applied in precision livestock farming devices, such as rumen boluses (Stefanska et al. 2018). The reliability of observing rumen pH in real time, compared to other methods for evaluating the risk of SARA, is considerably higher (Penner et al. 2007).

In the literature, we could find no information on real-time measurements that would explain how monensin controlled release capsules, emitting monensin doses of 335 mg daily, affect reticulorumen temperature and pH. We hypothesised that monensin would increase temperature and pH in the reticulorumen. Therefore, in this study we aimed to determine how a monensin controlled release capsule affects the temperature and pH in the reticulorumen using real time monitoring in fresh dairy cows.

# MATERIAL AND METHODS

The study was carried out in Eastern Europe, at  $56^{\circ}00'$ N,  $24^{\circ}00'$ E. The study involved 40 fresh (one day after calving) and second lactation Lithuanian Black Pied dairy cows, the average calving rate of which was 60-70 cows per month. The herd comprised 550 dairy cows, and a period of one month was used for creating the groups. All cows, housed in a free-stall system, were fed a total mixed ration, and milked twice per day. The examined cows produced  $30 \pm 3$  kg/day, and 9500 kg/year of yield on average, and the average body weight was  $550 \pm 65$  kg/cow.

Detailed clinical examinations revealed no health problems in any of the cows. The somatic cell counts (SCC) in the milk of the examined cows amounted approximately to  $200 \pm 32$  thousand/ml, and milk urea was  $20 \pm 1.4$  mmol/l. The monensin controlled release capsule used for the study emitted monensin daily doses of 335 mg, when administered with an oral balling gun in accordance with the manufacturer's instructions. Two experimental groups were formed as follows: (1) Test group (TG) supplemented with monensin (a monensin controlled release capsule (MCRC) of 32.4 g, n = 20) and (2) Control group (CG) (capsule containing no monensin, n = 20). Treatment of both groups began one day after calving, and the experiment was finished one month after calving.

An indwelling, wireless data transmission system (smaXtec animal care GmbH, Graz, Austria) was used to monitor the reticuloruminal temperature and pH (Gasteiner et al. 2009). Every 10 minutes, specific "smaXtec" boluses in the reticulum provided measurements. The duration of the measurement process was one month; analysis was performed on the collected data, and it was stored on a computer. The manufacturer's instructions were followed when performing the calibration of the pH probes.

Data analysis and statistics. All statistical analyses were performed using the SPSS program for Windows 15.0 (SPSS Inc., Chicago, IL, USA). Based on the Kolmogorov-Smirnov test, the distributions of the data collected while monitoring the groups were normal. The results of the descriptive statistics are presented as the mean and standard error. Comparison of differences between the means of the groups was performed using Student's t-test. The degree and direction of the relationships among the temperatures of the contents of the abomasum and reticulorumen were determined using Pearson's correlation coefficient (r). The significance level was set at P < 0.05.

The research was performed according to the provisions of the Law of the Republic of Lithuania No. 8-500 on Protection, Keeping and Use of Animals, dated 06/11/1997 (Valstybės žinios (official gazette) No. 108 dated 28/11/1997) and orders of the State Veterinary Service of the Republic of Lithuania on Breeding, Care, Transportation of Laboratory Animals (No. 4-361, dated 31/12/1998) and Use of Laboratory Animals for Scientific Tests (No. 4-16, dated 18/01/1999). The study's approval number was PK012868.

### **RESULTS**

Altogether, in the CG, the average pH in the reticulorumen was  $5.82 \pm 0.00$ . In the TG, the average was  $5.93 \pm 0.00$ , and the difference between the two groups was statistically significant (P < 0.001). The measurements shown in Table 1 indicate that the reticulorumen temperatures were 38.89 ± 0.01 °C (CG) and 39.21 ± 0.01 °C (TG) on average. Overall, the findings demonstrate that after daily administration of 335 mg doses of monensin, the reticulorumen pH was an average of 1.89% (P < 0.001) higher compared to in the CG. At the same daily monensin dose, the reticulorumen temperature was an average 0.82 % (P < 0.001) higher than in the CG. The coefficient of reticulorumen pH variation throughout the period of analysis was 5.44% in the TG and 5.43% higher in the CG; temperature variation in the TG was 3.45%; in the CG it was 0.60% lower. As shown in Table 1, the correlations between the reticulorumen temperature and pH for both the CG and TG groups are statistically significant (r = 0.135, P < 0.001 and r = 0.124, P < 0.01, respectively). The differences between the reticulorumen temperature and pH were statistically significant (P < 0.01) over the whole course of the study and when the different weekly periods were compared (Table 2). After one day of the experiment, a statistically significant difference between reticulorumen temperature in the TG  $(39.08 \pm 0.09 \,^{\circ}\text{C}; P < 0.01)$  and CG  $(38.67 \pm 0.10 \,^{\circ}\text{C})$ was observed.

In Figures 1 and 2, the data signifying changes in the analysed traits are displayed by day and group. Analysing the figures, it is apparent that throughout the entire time period of the experiment, reticulorumen temperature (from 0.18% on day 20 to 2.27% on day 10) and pH (from 0.68% on day 20 to 4.72% on day 27) were higher in the CG than in the TG.

Table 1. Statistical analysis of the investigated traits

Traits	CG (n = 11 651)	TG (n = 11 633)	
	mean ± SEM	mean ± SEM	
pH	$5.82 \pm 0.003$	5.93 ± 0.003***	
Temperature (°C)	$38.89 \pm 0.012$	39.21 ± 0.010***	
Correlation between traits	0.124**	0.135**	

CG = control group; SEM = standard error of the mean; TG = test group

<sup>\*\*</sup>*P* < 0.01; \*\*\**P* < 0.001

Table 2. Differences between	groups during the same perio	d. The mean of all cows from one group

Group	Traits	After 1 week	After 2 weeks	After 3 weeks	After 4 weeks
Descriptiv	e statistic				
CG	pН	$5.86 \pm 0.01$	$5.74 \pm 0.01$	$5.79 \pm 0.01$	$5.90 \pm 0.01$
	temperature (°C)	$38.85 \pm 0.02$	$38.89 \pm 0.02$	$38.94 \pm 0.03$	$38.85 \pm 0.03$
TG	рН	5.94 ± 0.01***	5.85 ± 0.01***	5.86 ± 0.01***	6.12 ± 0.01***
	temperature (°C)	39.24 ± 0.02***	39.28 ± 0.02***	39.17 ± 0.02***	39.11 ± 0.026***
Correlatio	n between traits				
CG	pH – temperature	0.098**	0.134**	0.164**	0.126**
TG	pH – temperature	0.144**	0.277**	0.116**	0.083**

CG = control group; TG = test group

We also investigated the effect that time of day had on the changes in the evaluated indicators (Figures 3 and 4). In both groups, a decrease in reticulorumen temperature and increase in reticulorumen pH were observed from 6.00 a.m. to 1.00 p.m. The most

prevalent difference occurred in reticulorumen pH (Figure 3) at 4.00, 5.00 and 9.00 a.m. (higher in the CG by 5.8–5.9%), and in reticulorumen temperature (Figure 4) at 5.00 a.m. and 1.00 p.m. (higher in the CG by 4.4–4.8%).

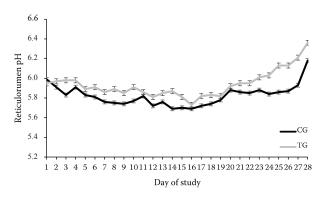


Figure 1. Reticulorumen pH during the experiment by day and group (mean ± S.E.M.)

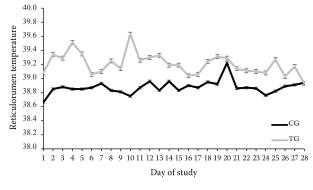


Figure 2. Reticulorumen temperature during the experiment by day and group (mean  $\pm$  S.E.M.)

Figure 3. Reticulorumen pH during the experiment depending on the hour of the day (mean  $\pm$  S.E.M.)

CG = control group; TG = test group

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CG = control group; TG = test group

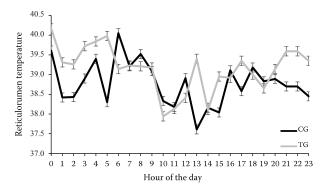


Figure 4. Reticulorumen temperature during the experiment depending on the hour of the day (mean  $\pm$  S.E.M.)

CG = control group; TG = test group

<sup>\*\*</sup>P < 0.01; \*\*\*P < 0.001

### **DISCUSSION**

Differences between the reticulorumen pH values in the two groups were observed at every point of the study. Sub-acute ruminal acidosis is defined as a condition in which pH is depressed for extended periods every day (Ingvartsen 2006). Monensin raises ruminal pH primarily by impairing the growth of lactate-producing bacteria, which would otherwise proliferate in conditions of abundant starch, as stated by Osborne et al. (2004). The prepartum administration of a monensin CRC did not increase rumen pH in multiparous cows provided with experimental diets in the transition and early lactation periods (Fairfield et al. 2007). Prado et al. (2010) failed to see any effects on bovines and water buffalos upon incorporation of sodium monensin or propolis-based products into foragebased diets. The efficiency of sodium monensin in maintaining ruminal pH at higher levels and in reducing dry matter intake is higher compared to that of a propolis ethanol extract (Silva et al. 2015). It is widely known that as reticulum pH is higher than rumen pH, SARA detection thresholds must be adapted to localisation and pH measuring methods (Sato et al. 2012). High-resolution pH kinetics, guaranteed by rumen pH sensors, are highly useful for the detection of SARA, in cases where rumen pH is examined correctly (Villot et al. 2017).

Throughout the study, the reticulorumen temperature rose by 0.82% (P < 0.001). Compared to a mixed hay diet, high amounts of grain in feed result in increased rumen temperature (AlZahal et al. 2008). Based on the devices utilised, there is a moderately linear relationship and an agreement in pH measurements with the manual technique applied. This might be because manual pH determination tends to be insufficiently accurate (Loholter et al. 2013). A statistically significant positive correlation was determined (r = 0.509, P < 0.001) between the temperatures of the contents of the abomasum and the reticulorumen. SARA status can be evaluated by observing ruminoreticular temperature (Antanaitis et al. 2016).

Real-time monitoring coupled with the use of a monensin controlled release capsule emitting daily doses of 335 mg results show that reticulorumen temperature and pH increased throughout the first 30 days after calving. It can thus be stated that the monensin controlled-release cap-

sule reduces the risk of SARA. Therefore, the risk of SARA in fresh dairy cows can be evaluated using real-time monitoring of reticulorumen temperature and pH and the prophylactic effect can be determined as well.

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