Measuring rumen pH and temperature by an indwelling and wireless data transmitting unit and application under different feeding conditions

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Summary
An indwelling system for monitoring reticulo-ruminal pH and temperature was assembled. Data were sampled and stored in a memory chip and could be read out via radio transmission to an external receiver. The indwelling system can be given orally, but to service the measuring units, feeding experiments and measurements were conducted using 5 rumen cannulated steers.
After calibration by using standardized pH-dilutions (pH4, pH7), pH and temperature measurements were carried out under exactly defined feeding conditions, also measuring daily DMI.
In feeding experiment 1, animals received only hay, in feeding experiment 2, animals had pasture during day and forage during night. In feeding experiment 3 animals received a diet containing forage:concentrate 50:50.
In feeding experiment 1, reticuloruminal temperature (mean 38.40±0.70° C) was significantly influenced by drinking water but it was not connected with feeding time. Mean pH was 6.49±0.39 and nadir was pH 6.14.
In feeding experiment 2, mean reticuloruminal temperature was 38.12±0.80° C and mean pH was 6.36±0.22. Nadir during pasture was pH 5.34, nadir during feeding roughage was pH 6.16. Pasture had a significant influence on reticuloruminal pH. In feeding experiment 3 mean temperature was 38.55±0.83° C and mean pH was 6.37±0.24. Nadir was pH 5.29. Decline of reticuloruminal pH was induced by feeding of concentrate.
When comparing the results of measuring standardized dilutions (pH 4, pH 7) prior and after in vivo measurements, coefficient of correlation was 0.9987. Drift pH 4 was 0.197 ± 0.070 and drift pH 7 was 0.107 ± 0.088
Results show that the presented method is a useful and proper tool for scientific applications. The measuring system can also be administered to uninjured cattle. An adapted indwelling pH measuring system will be assembled for practical purposes in future.

Keywords: Rumen acidosis, reticuloruminal pH, radio transmission, ruminants

Abbreviations: A/D Converter=analog to digital converter; CP=crude protein; DM=dry matter; DMI=dry matter intake; MJ NEL= megajoule net energy lactation; SARA=subacute rumen acidosis; SM-Band=industrial security medical band

Introduction

The decline of reticuloruminal pH under the physiological norm in cattle, mostly occurring as SARA, is a widely spread metabolic problem in dairy cattle. There is a disagreement as to a precise definition of SARA, which is a not always properly to be verified by its pathological status (DuffieldD et al., 2004, Plaizier et al., 2008). SARA may result in transient nadir of ruminal pH below 5.5 (Kleen et al 2003).

Techniques for continuous measurement of the ruminal pH were used for a series of scientific investigations (Date and Allen, 1993; Keunen et al., 2002; Nocek et al, 2002; Cotte et al., 2004; Rustome et al., 2006; Alzahal et al., 2007).
These techniques have in common that there is a rumen fistula to be laid at the animal to be examined and that the investigated data are registered in a memory chip in the rumen. In order to achieve the investigated data the memory chip has either to be removed (Date and Allen, 1993; Cotte et al., 2004; Keunen et al., 2002; Nocek et al., 2002; Penner et al., 2007; Rustomo et al., 2006) or the data are transmitted by a cable to an external unit, which is fixed onto the animal (Alzahal et al., 2007). Measurement duration is restricted because glas electrodes used in these investagions work only a few days without calibration.
In the following article investigations concerning the use of a method to measure the reticuloruminal pH by means of an indwelling probe in cattle and wireless transmission of the data will be presented.

Material and Methods
For continuous indwelling measurement of reticuloruminal pH and temperature a system was developed by researchers of Science Park Graz. These probes were tested in 5 ruminally cannulated steers under the conditions of 3 exactly defined feeding trials. The measuring unit has a break-proof synthetic case. The sensor-system is controlled by a micro-processor. Data are recorded by means of an Analog to A/D-converter and further handled by the micro-processor. The actual date, time of the day as well as the number of the earmark of the according animal can be integrated in the system. The measured data are filed in a not volatile storage of the probe and can be read out from outside the rumen by radio transmission to an external receiver at any time.
Measurement results are transmitted by wireless technique (433 MHz) to the external receiver. This receiver is connected to a laptop via USB, thus the results can be transferred and at once read out by means of an appropriately developed IT-programme. Results can be evaluated statistically, interpreted and graphically pictured by the programme. Measurement intervals are selectable by the user. For the present investigations a measurement interval of 30 minutes was selected.
Three defined feeding trials were carried out as exact trials, thus daily DMI was quantified (exception: pasture) together with weekly analyses of the ration (WEENDER-analysis, minerals and micronutrients).
After an adaption period for the respective ration conditions (14 days) the following feeding trials were carried out:

Feeding trial 1: 100 % roughage (hay) ad lib.: The animals got only hay, quality: Fiber 28.5%, CP 9.7%, 5.6 MJ NEL, feed intake was at 13.5 kg T on average.
Feeding trial 2: Daily pasture (from 4:30 a.m. until 4:30 p.m.) and forage ad lib. in the evening (5 p.m. until 4 a.m.). The basic ration in the evening always consisted of one-third hay (quality like in trial 1), grass silage (29.6% Fiber, 13.5% CP, 5.7 MJ NEL) and maize silage (20.9% Fiber, 8.6% CP, 6.3 MJ NEL), whereby the feed intake of forage was at 7.2 kg T on average. Grass from pasture had 18 % fiber, 25.5% CP and 6.4 MJ NEL on average, Dry matter intake on pasture could only be estimated.
Feeding trial 3: 50 % forage (one-third hay, grass-silage and maize silage) and 50 % concentrate were fed to the animals. Feeding in this trial did not happen ad lib., but rationed. For the ration a total food consumption of 12 kg T was taken as a basis. The basic ration always consisted of one-third grass silage, maize silage and hay (quality like trials 1 and 2). The concentrate (7.3% RFA, 18.2% RP, 7.5 MJ NEL) composed of 20 % barley, 21 % maize, 12 % wheat, 10 % dry cuts, 10 % wheat bran, 20 % bruised soya and 7 % rape extraction

coarse meal whereby each animal received 6 kg T – divided into two portions – daily. Concentrates were always given at 6 a.m. and 12 a.m..
Due to its construction type (length 120 mm, diameter 36 mm and weight 208 g, shaped like a torpedo) the probe could also be given per os to the cattle. In the present investigations the defined position of the probes was at the bottom of the reticulum, were they were placed and removed via the rumen fistula.
After calibration of the probes by means of an appropriate standardized calibration dilution (4 hours in pH 4 and in pH 7), reticuloruminal pH and temperature were measured. Hereafter the probes were removed and and standardized pH-dilutions were measured again. Results were used for validation of the results.
Statistical analysis was performed by means of GLM (Statgraphic Plus 5.1) and the Bonferroni-Holm-Test.

Results
Feed intake and behaviour of animals being equipped with a sensor were not affected by the system. After all measuring procedures the probe could be found in the reticulum, were it was placed before. The indwelling use did not damage the external synthetic material of the probes as well as the components inside were in working order.
Transmission of the data took about 1 minute and worked without any problems.

Results 100 % roughage (hay) ad lib.:
When only feeding hay, reticuloruminal temperature (mean 38.40±0.70° C) was significantly influenced by the water-uptake, the same observation could be made in trial 2 and 3 ($p<0.05$). The measured temperature did not show any relation to the feeding time, however, it did with respect to the periods of water-uptake. The mean pH was at 6.49±0.39 and the lowest measured pH (nadir) was at pH 6.14.

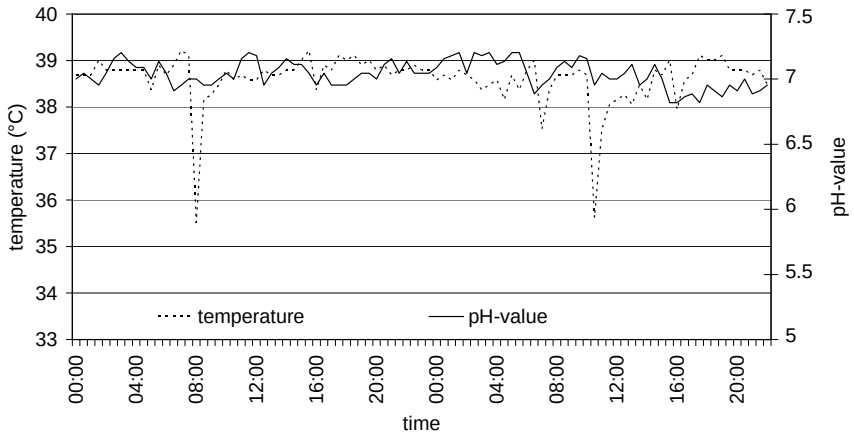

Figure 1: Diurnal variation of reticluoruminal pH and of temperature when feeding only hay

Results when providing daily pasture and forage ad lib. during night
Mean reticuloruminal temperature was 38.12±0.80 °C and mean ruminal pH was at 6.36±0.22. Nadir on pasture was pH 5.34 and nadir during night, when feeding only forage, was pH 6.16. Grazing pasture had significant negative influence on reticuloruminal pH ($p<0.05$).

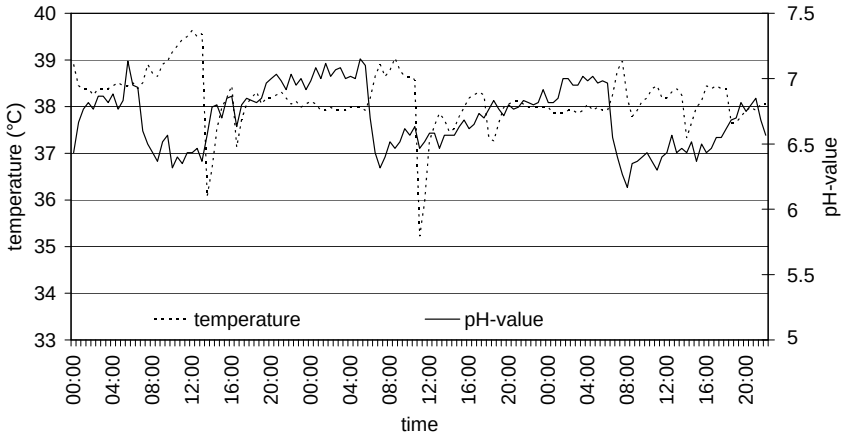

Figure 2: Diurnal variation of reticluoruminal pH and of temperature on pasture during day and feeding forage during night

Results 50% forage : 50 % concentrate
Mean temperature in the reticulorumen was 38.55±0.83 °C in trial 3 and mean pH was at 6.37±0.24. Nadir was pH 5.29. The decline of the rumen pH-value significantly correlated with the feeding of concentrate.
For validation of the measuring results the used probes were given into standardized pH-dilutions prior to and after each use. These results were compared to each other. The correlation coefficient averaged at 0.9987 for all probes.

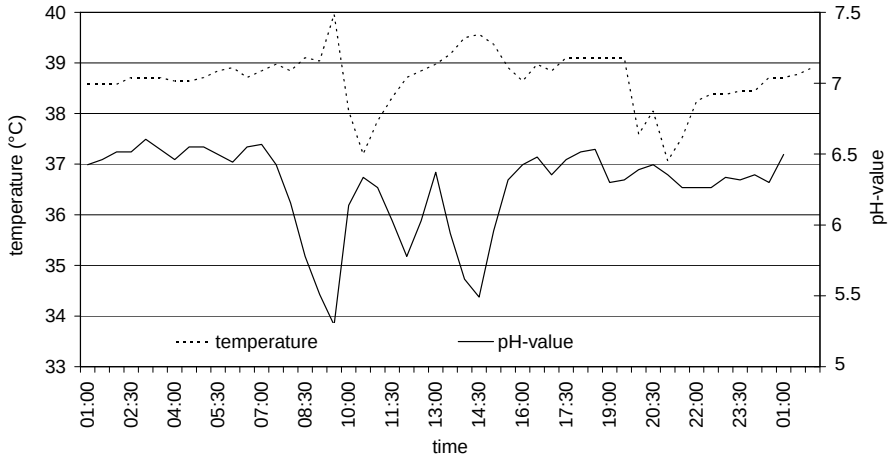

Figure 3: Diurnal variation of reticuloruminal pH and of temperature when feeding roughage : concentrate 50 : 50

Table 1: Validation of pH- probes (drift and coefficient of correlation)

	Calibration pH 4	Calibration pH 7
Number of measurements	42	42
Drift absolute	0.197 ± 0.070	0.107 ± 0.088
Drift (%)	2.44 ± 1.76	1.53 % ± 1.26

Measuring duration of probes was from 8 to 40 days without any service. During this period measured data approved to be accurate.

Discussion

Described feeding trials were carried out by using ruminally cannulated steers (n=5). Because surgery of a rumen fistula is subject to approval according to the animal welfare law and therefore restricted, we had to be content with this number of animals. The described measuring system represents the first description of a wireless data transmission of continuously investigated measurings (temperature and pH-) of the reticulorumen in cattle under exactly defined feeding conditions.

Probes were used in ruminally cannulated steers (n=5), so the service of probes and validation of measurings was ensured.

At pure feeding of roughage the temperature in the reticulorumen was significantly influenced by the water uptake (38.40±0.70 °C). This also applies to trial 2 (38.12±0.80 °C) and trial 3 (38.55±0.83 °C). The measured temperature, however, did not show a statistical relation to the feeding time in any trial. The acyclic occurring decline of temperature (negative peaks) and water uptake of the animals, however, were temporally correlated with water uptake. Reticuloruminal pH was not significantly influenced by water uptake. This circumstance can be explained by the effect of intermixture.

Feeding also has significant influence on the reticuloruminal temperature, whereby the lowest temperatures were found on pasture with additional feeding of roughage and highest temperatures were found when feeding high amounts of concentrate. Primarily, retiuloruminal temperature shows the microbiological conversion process and is therefore the highest when feeding fast fermentable carbohydrates (Oetzel, 2003). These findings were also proven in the present investigation.

In their investigations Alzahal et al. (2007) found that the ruminal temperature showed a negative correlation (R^2=0.77) to pH-nadir and concluded therefore that this can also be a hint to a consisting rumen acidosis. On the basis of the presented data it can not be concluded, that the measurement of reticuloruminal temperature can give a hint concerning the reticuloruminal pH or whether there are correlations.

Pasture with additional feeding of roughage showed the lowest temperature in the reticulorumen. On pasture the animals have also been exposed to the sun and this might have led to a higher water consumption.Additionally, pasture has the highest water content compared to the other utilized feeding stuff (about 85 %) and the fresh mass having been taken up is therefore definitely higher as when compared with dry food (concentrate, hay), whereby temperature in the reticulorumen might have been decreased more clearly and sustainably.

The mean pH-value in the reticulorumen was 6.49±0.39 in feeding trial 1 and the lowest pH (nadir) was at pH 6.14. This range is in accordance with the physiological level of the rumen pH-value (Van Soest, 1982).

In feeding trial 2 the mean pH-value in the reticulorumen was at 6.36±0.22. The nadir during the pasture period was pH 5.34 and was therefore clearly in the acidic range (VAN SOEST,

1982). Feed-intake on the pasture had significant influence on the pH-value in the reticulorumen. During and after the evening hay feeding nadir was at pH 6.16, which demonstrated the pH-regulating effect of hay being rich in fiber.
In feeding trial 3 mean pH-value was at pH 6.37±0.24. Nadir was pH 5.29 and therefore strongly acidic. The decline of the reticuloruminal pH-value was significantly correlated to the administration of concentrate. Steingass and Zebelli (2008) report that the pH-value in the reticulorumen should be at 6.32 on average in order to maintain physiological conditions and optimal conditions for fermentation. The presented results were performed with 5 ruminally cannulated steers, the daily DMI of which (12-13 kg T) was at about the half of the one of a lactating cow due to missing performance like milk production and pregnancy.
Accordingly, these indications are only representative for cattle with low food intake. Data for animals with higher DMI ought to be determined by means of according trials.
The presented measuring system allows giving of exact dates, how long the pH-value is under a defined level and it is possible to estimate the velocity of pH-decline and increase respectively. Values of < pH 5.5, < pH 5.8 and < pH 6.2 were defined as critical limitations in the present study.
When validating measuring results (see table 1) a correlation coefficient of 0.9987 could be determined for the outcome of the used probes. This method of evaluation seems to be more favourable as the method of Alzahal et al. (2007), who took rumen fluid via rumen fistula three times daily (09:00 a.m., 01:00 p.m. and 04:00 p.m.), determined the pH by means of pH-meter and compared these data to the results of the probe. The herewith determined coefficient of correlation was 0.88.
Measuring duration of probes used in our feeding trials was from 8 to 40 days without any service. Since measured data approved to be accurate, the present probe represents a promising device for the future.
Our results show that the presented indwelling system for measuring reticuloruminal pH and temperature in cattle represents an innovative and reliable basis for the clarification of scientific questions in terms of rumen physiology and rumen pathology.
Periods of acidic stress can exactly be recognized and also temporally be defined by the described system. Because the probes are to be seen potentially affected by heavy metals and other potential hazardous substances, security and stability of the probes must be guaranteed. The question whether an indwelling sensor also stays inert with a longer resting time and whether a negative effect on animal health can be excluded is still to be clarified by means of long term trials.
Only if security is guaranteed, a practical use in animals, which deliver victuals, will be able to take place routinely.
As the presented measuring probes can also be given to adult cattle per os, it is only a question of time and money, until an adapted, improved and verifiably secure system is also deployed under practical conditions – preferably in large herds – in order to control ration and animal health (“indicator animals”).

Note

For the present investigations with ruminally cannulated cattle a valid lawful licence for animal experiments according to the national regulations (Tierversuchsgesetz TVG) is at hand from the relevant office of the Styrian provincial government

References

Alzahal, O., Kebraeb, E., France, J., Froetscher, M. and McBride, B.W., 2007. Ruminal temperature may aid in the detection of subacute ruminal acidosis. Journal of Dairy Science **91**, 202-207.
Cottee, G., Kyriazakis, I., Widowski, T.M., Lindinger, M.I., Cant, J.P., Duffield, T.F., Osborne, V.R. and McBride, B.W., 2004. The effects of subacute ruminal acidosis on sodium bicarbonate-supplemented water intake for lactating cows. Journal of Dairy Science **87**, 2248-2253.
Duffield, T., Plaicier, J.C., Fairfield, A., Bagg, R., Vessie, G., Dick, P., Wilson, J., Aamini, J. and McBride, B., 2004. Comparison of techniques for measurement of rumen pH in lactating dairy cows. Journal of Dairy Science **87**, 59-66.
Keunen, J.E., Plaizier, J.C., Kyriazakis, L., Duffield, T.F., Widowsky, T.M., Lindinger, M.I. and McBride, B.W., 2002. Effects of a subacute ruminal acidosis model on the diet selection of dairy cows. Journal of Dairy Science **12**, 3304-3313.
Kleen, J.L., Hoojer, G.A., Rehage, J. and Nordhuizen, J.P.T.M., 2003. Subacute Ruminal Acidosis (SARA): a Review. Journal for Veterinary Medicine A **50**, 406-414.
Nocek, J., E., Allman, J.G., Kautz, W.P., 2002. Evaluation of an indwelling ruminal probe methodology and effect of grain level on diurnal pH variation in dairy cattle. Journal of Dairy Science **85**, 422-428.
Penner, G.B., Beauchemin, K.A. and Mutsavanga, T., 2007. Severity of ruminal acidosis in primiparous Holstein cows during the periparturient period. Journal of Dairy Science **90**, 365-375.
Plaizier, J.C., Krause, D.O., Ghozo, G.N. and McBride, B.W., 2008. Subacute rumhinal acidosis in dairy cows: The physiological causes, incidence and consequences. The Veterinary Journal **176**, 21-31.
Rustomo, B., Alzahal, O., Cant, J.P., Fan, M.Z., Duffield, T.F., Odongo, N.E. and MCBride, B.W., 2006. Acidogenic value of feeds II. Effects of rumen acid load from feeds on dry matter intake, ruminal pH, fiber degradability and milk production in the lactating dairy cow. Canadian Journal of Animimal Science **86**, 119-126.
Steingass, H. and Zebelli, Q., 2008. Strukturbewertung von Rationen für Wiederkäuer. Viehwirtschaftliche Fachtagung, 9.-10. April 2008, Bericht LFZ Raumberg-Gumpenstein **35**, 27-33.
Strabel, D, Ewy, A., Kaufmann, T., Steiner, A. and Kirchhofer, M., 2007. Rumenozentese: Eine geeignete Methode zur pH-Bestimmung im Pansensaft beim Rind?, Schweizer Archiv für Tierheilkunde **149**, 301-306.
Van Soest, P.J., 1982. Nutritional ecology of the ruminant. Comstock Publ. Association of the Cornell University Press, Ithaka and London.