Commercial Benefits of Routine Monitoring of Rumen pH in Dairy Cows in South West England

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Abstract

Ruminal pH has been shown to be a very important parameter for nutritional status and the disease Sub-Acute Ruminal Acidosis (SARA) (Dirksen, 1986). Therefore, monitoring the ruminal pH has been studied to recognise, quantify and subsequently control this disorder. Different methods have been available to measure the pH in the rumen over the years, with different accuracies and ease to perform. Since 2005, boluses that measure pH continuously and using wireless techniques have been used for research purposes and mainly in fistulated cows. However, nobody has reported what the farmers and their advisers make of data such as rumen pH. The challenge for farmers and their consultants is to implement feeding management and husbandry practices that avoid or reduce the incidence of SARA. The aim of this study was to see how a wireless rumen pH telemetry bolus can be used on a commercial farm, what the farmer's reaction to get the data is and how it can improve the economy and management of the farm. For this purpose, a trial on eight commercial farms was run from April to August 2013 in the South West of England. Results from this study have shown the complexity of the rumen environment and the different methods that farmers use to change it. Six farms have reported a change as result of the data. 60% of farmers and their consultants have seen a monetary benefit for the farm and 100% of farm's advisers have seen a commercial benefit for their company.

Introduction

European dairy industry is characterised by its diversification in territories and the diversification of products. However, dairy production is in a mutation period and soon will have to manage different challenges, mainly with the Common Agricultural Policy (CAP) reform. Milk price and feed stock instability, milk quota abandonment and continuous need for increases in quality from consumer make new confrontations for this sector. Therefore the dairy industry has the challenge of producing more to meet the food deficit and changing the way of producing to respect the environmental causes. It is then more difficult and uncertain to produce milk in good economic conditions.

Between diversity, specialisation and standardisation, the dairy sector is complex to define. However, scientific and technological knowledge have sufficiently advanced over the years to enable feeding management and husbandry practices to overcome the challenges and allow sustainability.

In this way, eCow ltd has developed a tool to monitor the rumen pH which has a central place in cows' health and milk production. First used in the research area, and after having proved the quality of the product, eCow decided to make this technology accessible to all commercial dairy farms. Measuring reticuloruminal pH and temperature, it can detect and prevent one of the most important economic and health problems in dairy farm and beef cattle, Sub-Acute Ruminal Acidosis (SARA).

The aim of this study was to see how wireless rumen pH telemetry bolus can be used on a commercial farm, what the farmer's reaction was to the data and how it can improve the economy and management of the farm. This paper is showing the results of a trial on different commercial farms which has been run from April to August 2013 in the South West of England. For this purpose, first what SARA is will be explained, and the different researches to detect and understand it. Then the materials and method of this experiment will be described before showing the results. The last point will be made by discussing all the results of this trial and what the potential benefits are for dairy farms.

Context of the study

Ruminant animals are adapted to digest and metabolize mainly forage diets. However, growth rates and milk production are increased substantially when cows consume high-starch diets. One consequence of feeding excessive amounts of concentrates with inadequate fibre to ruminants is SARA, also known as chronic or sub-clinical acidosis. It is a well-recognised digestive disorder that is an increasing problem in most dairy herds. Nowadays, farmers attempt to increase milk production and results from field studies indicate a presence of SARA in 11-29.3% of the early lactation cows and in 18-26.4% of the mid lactation cows (Garrett et al., 1997; Kleen, 2004; Tajik et al., 2009).

Dairy herds experiencing SARA will have feed intake depression, reduced fibre digestion, milk fat depression and impaired cows' health which often results in a high culling rate (Jorgenson et al.,1993; Nordlund, 1996).

What is SARA?

SARA is currently defined as a decrease in ruminal pH below a certain threshold for prolonged periods each day. Ruminal fluid pH is a measure of the acidity or alkalinity of ruminal contents, which has diurnal fluctuations. The normal rumen pH is in the range 5.5-6.8, depending on the diet (Cunningham, 2002). For optimum fermentation of the diet and fibre digestion, ruminal pH should be between 6.0 and 6.4 because the cellulolytic bacteria, which allows the digestion of fibre, are inhibited when pH is less than 6.0 (L. Commun, 2011). However, even in healthy cows, ruminal pH will vary below this level for short periods during the day due to the digestion of carbohydrates (e.g. Starch), from cereal grains. Grain diets are high in readily fermentable carbohydrates which mean they will be rapidly broken down and lead to volatile fatty acids (VFA) and lactic acid (Mutsvangwa and Wright).

Absorption of VFA from the rumen occurs passively across the ruminal wall. This passive absorption is enhanced by finger-like papillae that project away from the rumen wall. These papillae provide a huge surface area for VFA absorption. Ruminal papillae vary in length depending on the diet cattle are fed, which protects the animal from acid accumulation in the rumen (Krause and Oetzel, 2006). If the absorptive capacity of these cells is impaired (e.g. chronic rumenitis with fibrosis), then it becomes much more difficult for the animal to maintain a stable ruminal pH following a meal. Accumulation of VFA in the rumen is the principal cause of SARA.

Ruminants possess highly developed systems to maintain ruminal pH within a physiological range about 5.5 to 7.0. As ruminal pH begins to drop following a meal, a ruminant's first response is to stop eating (Oetzel, 2007). Depending on the amount of fibre content in the diet, animals will start ruminating and by consequence producing a large amount of buffers via their saliva which will increase the ruminal pH. Conversely, if the proportion of fibre in the diet is too low, the buffers in saliva will not prevent the decrease because of a lack of rumination, the ruminal pH will drop under a threshold for several hours a day and the animal will be considered to have Sub-Acute Ruminal Acidosis.

Two situations are likely to present the risk of SARA. First, the transition from the pregnant non-lactating state to the non-pregnant lactating state is the period during which the majority of metabolic diseases occur and where SARA is the most present. During this period, which ranges from 3 weeks before until 3 weeks after calving, the cow is switched from a high-fibre, low concentrate diet to a diet that is higher in concentrate feeds and lower in fibre. The ruminal absorption capacity for acids, due to a reduction in the length and density of rumen papillae, can decrease by 50% during the dry period. It will take several weeks for this capacity to be restored after high concentrate diets are reintroduced (Dirksen et al, 1985; Duffield et al, 1999). Secondly, further in lactation, inaccurate calculation of dry-matter-intake (DMI) leading to wrong roughage/concentrate ratio, an inadequate content of structure within the diet or mistakes in preparing of total mixed rations may produce SARA (Kleen et al, 2003).

Measuring the Ruminal fluid pH.

SARA can be a difficult condition to diagnose because its appearance is subtle. The most common clinical sign associated with this disease is reduced feed intake in order to reduce the acidity of the rumen (Mutsvangwa and Wright). However, where cows are housed and fed in groups it becomes harder to see the individual feed intake and often it is unnoticed. Other signs are reduction in milk fat content, feed conversion efficiency, decreased digestion of fibre (Lean et al. 2000), laminitis causing lameness (Nocek 1997; Owens et al. 1998), liver abscessation (Owens et al. 1998), scouring (Nocek 1997), and a higher incidence of left and right displacements of the abomasum (Shaver 1997). Nonetheless, these signs appear after an episode of Sub-Acute Ruminal Acidosis, sometimes several months later, which enhances the subtlety in the detection of the disease. Furthermore, no peripheral markers could properly predict the pH of the rumen (Enemark et al, 2004).

The only reliable and accurate diagnostic test for SARA is measuring ruminal fluid pH, which can be done by different techniques, with different ease to do and accuracies. The first technique was to use a cannulated cow and sample an amount of ruminal fluid and measure it with a portable pH meter. Smith (1941) reported the first in vivo measurement studied. After some research, it has been found that there is a difference between in situ pH measures and cannula pH measures (Smith, 1941; Dado and Allen, 1993; Garett et al, 1995). Furthermore, fistulated cows are for research purposes and require a major surgical intervention which is expensive and invasive.

Thereby, techniques able to be done on-farm have been developed. Stomach tubing has been used to collect ruminal fluid samples for pH measurement. It consists of using an oro-ruminal probe and a suction pump. However, this technique has quickly been determined inaccurate as the sample is exposed to saliva contamination (Duffield et al., 2002; Alzahal et al., 2007).

Rumenocentesis, sometimes referred to as percutaneous needle aspiration, involves inserting a needle (1.3 mm, 12.7 cm long) into the ventral rumen, and withdrawing a sample of ruminal fluid using a 10-mL syringe. Although reliable data has been measured from rumenocentesis, it is an invasive technique for the animals and there could be a problem of milk decrease, haematomas and abscess formation at the puncture site if it is not done properly (Kleen et al., 2004; Garrett et al, 1999).

These techniques allow only spot samples and thus don’t record the dynamic pattern of the pH which is probably a key to understand the pathogenesis of SARA (PLaizier et al., 1999). That also means the results are dependent on the sampling time relative to feeding because it is important, to allow proper interpretation of results, that the samples are taken a certain time after feeding which may vary depending on the diet (Nocek, 1997; Shaver, 1997; Owens et al, 1998; Lean et al., 2000). Additionally, rumen pH varies significantly among sites within the rumen and by consequence samplings have to be done at the same place each time (Duffield et al, 2004; Kimura et al, 2012).

First attempts to measure pH continuously were conducted in cattle by Lampila (1955) and Lampila and Poutiainen (1966) and in sheep by Matscher et al. (1957), Matcher (1958) and Beghelli et al. (1958). They were using indwelling glass electrodes in cannulated animals linked through a wire to a receiver located outside the rumen. This technique has been used succesfully for a long time in research areas. Moreover, this method enables recording and understanding of the diurnal fluctuation of the ruminal fluid pH. In 1993, Dado and Allen developed a system to get constant measurement of ruminal fluid pH in animals kept in stanchions but reported difficulties in maintaining accuracy of the electrode due to the invasive liquid in the rumen. This device required cleaning and calibration every 2 days to avoid electrode drift (Dado and Allen, 1993).

Further researchers have progressively improved the accuracy of pH measurement over time and increased the freedom of the animal (Keunen et al., 2002; Maekawa et al., 2002; Cottee et al., 2004; Beauchemin and Yang, 2005; Rustomo and Al Zahal, 2006; Al Zahal et al., 2007). However, they still used an external cable and consequently cannulated cows. After has been first suggested by Jorgensen et al (1993), the development of a permanent emplaced, intraruminal transmitter equipped to record and store or transmit continuous intervals of intraruminal pH, as well as resisting electrodal clogging, has been investigated (Enemark et al., 2003; Graf et al., 2005; Penner et al., 2006). These devices were used on cannulated cows due to the need for data transfer and frequent calibration but the cows were allowed to move freely. Enemark et al. (2003) experimented with a continuous recording period of 8-days with an electrode positioned in the reticulum and reported only a minor drift of the electrode over this period.

Peters (1997) studied the digestive process in penguins and used a novel pH sensor for this purpose, featuring a free-flowing junction to compensate pressure changes, which counteracts contamination of the reference electrode. A similar operating principle of this device has been used in humans to monitor the gastroesophageal acid reflux (Bravo™ pH monitoring system, Medtronic, http://www.medtronic.com/downloadablefiles/Gastro-GastroFranchiseBackgrounder.pdf). This type of probe uses “gastric telemetry” to transmit data detected by a sensor in the probe via radio signals.

Recently, a new device has been reported, using a wireless indwelling probe, called a bolus (Mottram et al, 2008). The bolus measures pH continuously, stores the data and transmits it telemetrically via an in-built radio transmitter to a receiver station located around the cow. It gave reliable data for up to 40 days (Phillips et al., 2010).

In recent times, bolus technology has been improved and a recent study using a similar bolus has reported that continuously recording rumen pH telemetry can give accurate data for over 150 days and can continue to be downloaded for some 7 months after insertion (Mottram et al., 2013). In this study boluses were used in commercial cows, the bolus is swallowed by the cow and goes directly to the reticulum. The pH measured is consequently higher than in the ventral sac (Kumara et al, 2012) and needs to be taken into consideration when interpreting the data.

Boluses have been used for a number of years to encapsulate compounds for digestion in the rumen, to identify animals, to magnetise metallic debris or to monitor rumen health. Fistulated cows have been a key part of rumen research for years and our knowledge of the dynamics of the rumen is largely been derived from studies with this type of cows. However, fistulation is a major surgical intervention and creates a highly artificial rumen which not only has a leaky plug in the side but also has the abundance of instrumentation that may interfere with the movement of rumen contents.

The technique used to measure rumen pH may affects pH value and the indwelling pH measurement and data transmitting system is a very useful and proper tool for long term measurement of ruminal pH in cows (Gasteiner et al, 2012). It is advantageous because it can detect rapid fluctuations in variables that are often more difficult to acquire with spot samples due to diurnal variation (Keunen et al, 2002; Duffield et al, 2004).

There is no defined specific critical measure to determine SARA. The threshold depends upon the method used to measure the ruminal pH, the time after feeding (for spot sampling), and the site of sampling. The time spent below this limit is also important due to the daily variation of the rumen pH. Results from studies suggest that transitory periods of low pH may be beneficial from the standpoint of protein nutrition and may result in an increased flow of non-ammonia-nitrogen and essential amino acids (Calsamiglia et al., 2002).

SARA is a subtle condition in most high-producing dairy herds, leading to unnecessary economic losses. The wireless rumen pH telemetry has been proved to provide a convenient technique for detecting SARA and with the ability for it to be done on commercial farms.

Materials and method

Device and software

The boluses used for this experiment were the boluses from eCow Ltd, distributed to farmers, vets and nutritionists, called FarmBolus. A FarmBolus is 127 mm long, with a diameter of 27 mm and weighs 207 grams. The bolus is made from different electronic, metal, glass and plastic components. It comprises a stainless steel head and internal electronics. The spine is a pH probe, routinely used in industry, which runs the length of the device into the hollow chamber of the metal head. The temperature probe is embedded in the stainless steel end cap, which has holes to allow rumen liquor to flow past the sensor without permitting direct impact of stones or grit on the glass sensing bulb. The body is then filled with green pigmented epoxy resin that encapsulates the delicate electronics and protects them against rumen liquor and obviates the need for vulnerable seals. Pictures of an assembled bolus are in figure 1.

Figure 1: Picture of an assembled bolus

Due to the stainless steel sensor's end, the centre of gravity is below the central plane and has a specific gravity of 2.7. This recipe of dimensions ensures the bolus is swallowed comfortably by the cow, guarantees that the device resides in the sump of the first stomach and in a sensor down position in cows with normal shaped reticulum. The bolus doesn't contain toxic material at doses harmful for the cow. It is swallowed by the cow and the use of a standard bolus gun is required to put the bolus into the mouth. A short period before downloading is needed to permit the bolus to reach the reticulum, which is the only restriction on operation. One reason for this waiting phase is that the bolus has a temperature switch, which activates only when the temperature is above 31°C. This allows shelf storage of 2-3 years. A calibration is made when shipped by the company and is accurate for four weeks in normal storage. Inside the cow, the drift of accuracy is less than ±0.1 pH unit per 30 days.

The bolus has different settings to define the frequency of sampling and the time, duration and frequency during which it wakes up to respond to a radio signal. Boluses measured pH and temperature every 60 seconds and took an average value every 15 minutes, and woke up every minutes for radio signal and stored up to 2700 lines of data in a .csv format. The rows of data are presented in columns representing date, time, pH, temperature and battery voltage. With these recording settings, the bolus recorded 96 lines of data per day stored over 28 days. If data was not collected, the file on the bolus was overwritten from the beginning.

Two handsets have been provided by eCow ltd to download the data (see figure 2). At the beginning, a tablet computer (Samsung Q1 Ultra) running on a Windows system was used. This device was designed for research and combined all computer tasks as well as the software provide by the company. The software enables the user to calibrate the bolus, download the data and configure the different settings for the bolus. An antenna was plugged in to enable downloading via a USB port.

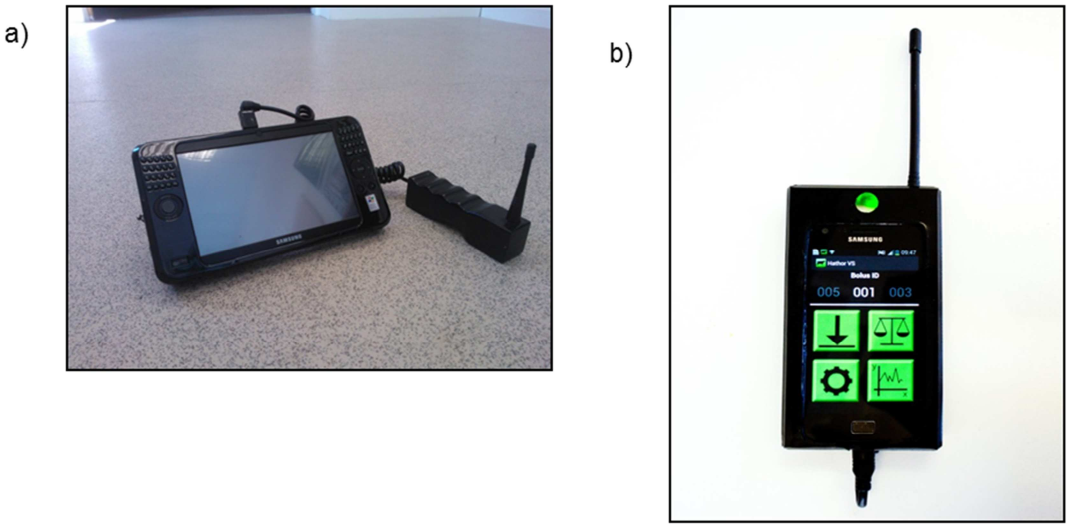

Figure 2: Pictures of both handsets used for the trial: a) Samsung Q1 Ultra and b) Samsung Galaxy S2.

Then the tablet computer was replaced by a phone (Samsung Galaxy S2). This device has been developed at the same time of the trial and consequently was a test version. Different updates improved the software and more functions were added than earlier versions, for example graph viewer. To download, an antenna was plugged in via the micro USB port of the phone, which has a power output. The antenna is part of a mould which surrounds the phone. Once the antenna is plugged in, the software comes up and is ready to download. When downloading is finished, a graph of the data is drawn on the screen and can be swiped with a finger to see the whole period of data.
The main bi-directional communication link between the bolus and the handset was operating on a frequency of 433 MHz. Each bolus only answers to a specific signal defined by its ID. That means the bolus number needs to be recorded on the handset before downloading, otherwise the bolus will never respond.

Method

In this study, eight commercial farms, referred as farm A to H, have been used, all located in the South West of England (see figure 3). The first farm has been chosen as a partner of the Royal Agricultural University. The other farms were direct customers of eCow or customers of Mole Valley Farmers so they have not been chosen for their characteristics.

Figure 3: A map representing the locations of farms involved in the trial.

There were an average number of four boluses per farm going into different type of cow (dry, fresh, early lactation) with different management, going from fully housed with Total Mixed Ration (TMR) to grazing herds. Table 1 summarises the farms and their management. The cows were chosen by the farmer, vet or nutritionist and the only advice given was to put the boluses in average cows and not in sick cows to better monitor the group rather than find the problem for one cow.

	Number of cows	Milk Yield (kg/year/cow)	Type of diet	House management
Farm A	900	11000	TMR	Fully housed
Farm B	350	12500	TMR	Fully housed
Farm C	360	11400	TMR	Fully housed
Farm D	130		TMR	
Farm E			Grazing	
Farm F		8427		
Farm G			Grazing + concentrate	Partially housed
Farm H	300		Grazing + concentrate	Partially housed

Table 1: *A table representing the information recorded on each farm.*

The day before inserting the boluses into cows, boluses were warmed up with hot water to check the radio signal response. After a positive answer, they were calibrated with two standard buffer solutions of pH 4.01 and pH 7.01 (HANNA instruments pH sachet HI 70004 and HI 70007), using the software of the handset. After calibration, an end cap was put on the steel end of each bolus to keep the sensor wet. Then at the farm, the end caps were withdrawn, the bolus numbers recorded with a corresponding cow number. The bolus was put in a bolus gun and the farmer dosed it.

Farms A and B were downloaded weekly by the author while farms C to H were on a two week cycle done by nutritionists. Depending on the farm, data was either downloaded with cows on yard, cubicles or kept on a cattle crush. Downloads were made by standing next to the cow, around the brisket, until a radio signal was established between the bolus and the handset. Figure 4 is an illustration of the downloading method. Apart from time windows, used to extend battery life, original settings haven't been changed. Time windows were set on each bolus at the convenience of the farmer and the person who came to download the data.

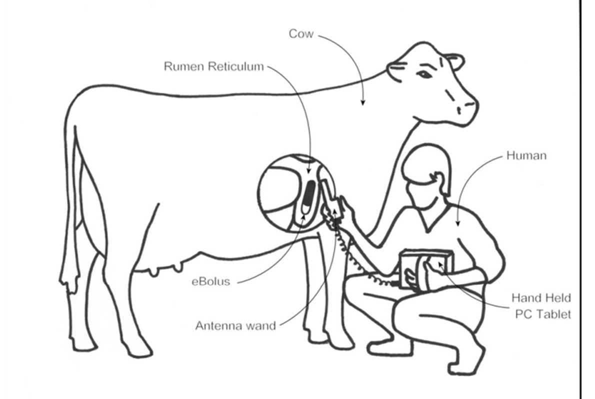

Figure 4: Illustration of the downloading method

After downloads data was transferred for analysis by USB key or email for the Q1 handset and were automatically sent by email with the Galaxy S2 when unplugging the antenna. At the first stage files were opened on Microsoft Excel (2010) to make different graphs. Secondly, these operations were performed using the software Matlab (MATLAB and Statistics Toolbox Release 2013a, The MathWorks, Inc., Natick, Massachusetts, United States). Scripts were used to produce a pH plot, a temperature plot with drinking events and a daily pH profile plot, in a quick and as automatic as possible way. These plots were systematically sent to farmers, vets and nutritionists for interpretation. Other scripts have been made to calculate the time below the threshold, the mean daily pH, the battery depletion and the risk of acidosis. These statistical results were sent when it was necessary. The data analysis was compared to milk records, cows' events and management events to see different influences in pH, temperature and routine.

At the end of the study, mid-August, two types of survey had been distributed. Both were made to get feedback from this trial and to know the satisfaction about the bolus technology. One type was aimed at the farmers and the other one was for their advisers.

Results

Graphs of pH and temperature

Data collected was presented into graphs which helped in the detection of the risk of SARA. Figure 5 gives an example of each graph produced by the data analysis. The pH range on the farms was on average between 6.8 and 5.4 (±0.1 pH).

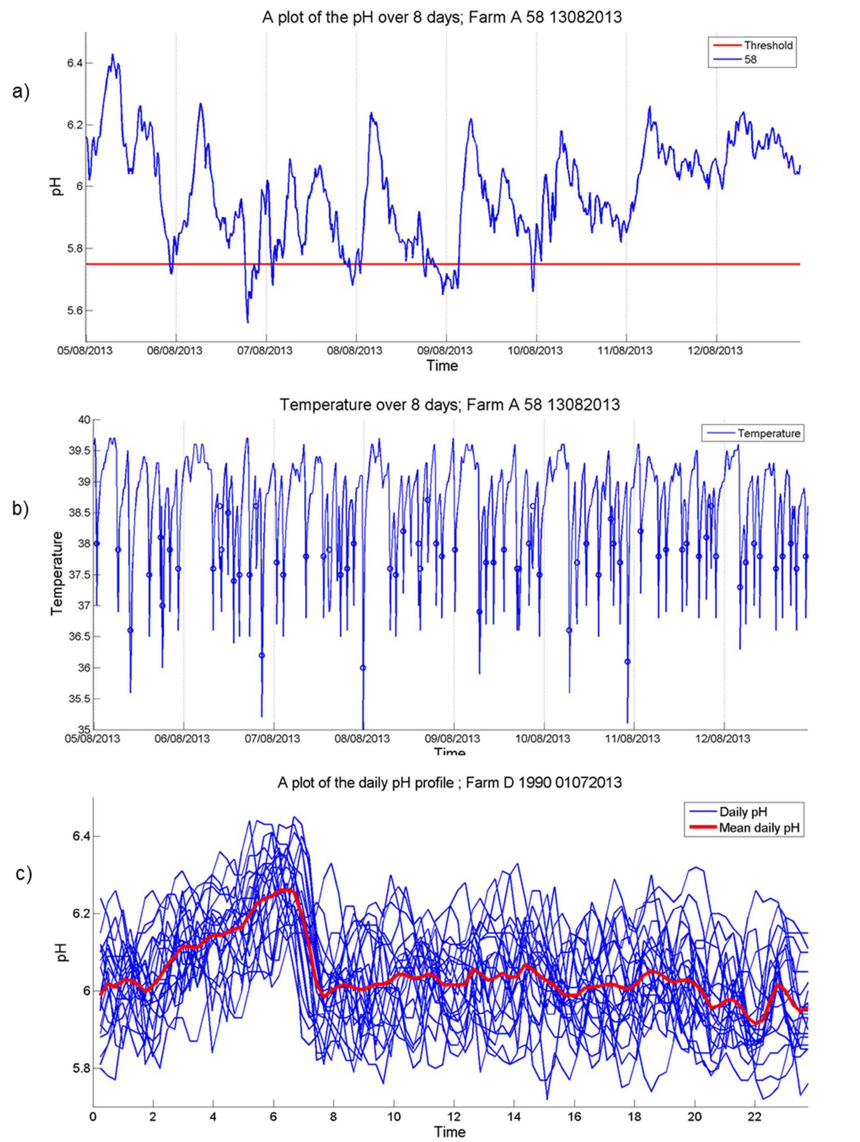

Figure 5: Example of the graphs produce for the data analysis. a) Plot of the pH over the time, b) plot of the temperature over the time and c) plot of the daily pH profile with the mean pH in red.

First data was showing five farms potentially at risk of SARA. These farms were defined at risk of acidosis because at least one cow spent time below the threshold. In total, five farms have reported to have changed the feeding management by changing the feed intake, the concentrate/forage ratio or the concentrate intake. One farm changed the time of feeding and another one did not change any feed management because no other feed than grass was available. Table 2 summarises the different changes for each farm. Results of these changes were an increase in the mean daily pH for three farms which was interpreted by a diminution of the risk of SARA. The other increased the milk yield and/or decreased the cost of the diet.

Farm	Number of boluses	Management change	Comments
A	6	Feed change	Decrease intake for heifers and increase for high yielding
B	4	Feed change	See case study
C	6	Feed change	Increase intake and stopped using acid buffer in the ration
D	4	Feed change	Change concentrate intake
E	2	Grass management	Change the time of moving the electric fence on fields
F	4	no report	
G	4	Feed change	Change concentrate intake
H	4	nothing	

Table 2: *A table of changes as result of data collected*

During the recording period, 7 cows with boluses calved. Graphs revealed a potential risk of acidosis after calving due to a big drop in pH (see figure 6). Infection has also been seen in the data, represented by an increase in temperature. One or two days after such events happened, farmers reported treating the cow for mastitis.

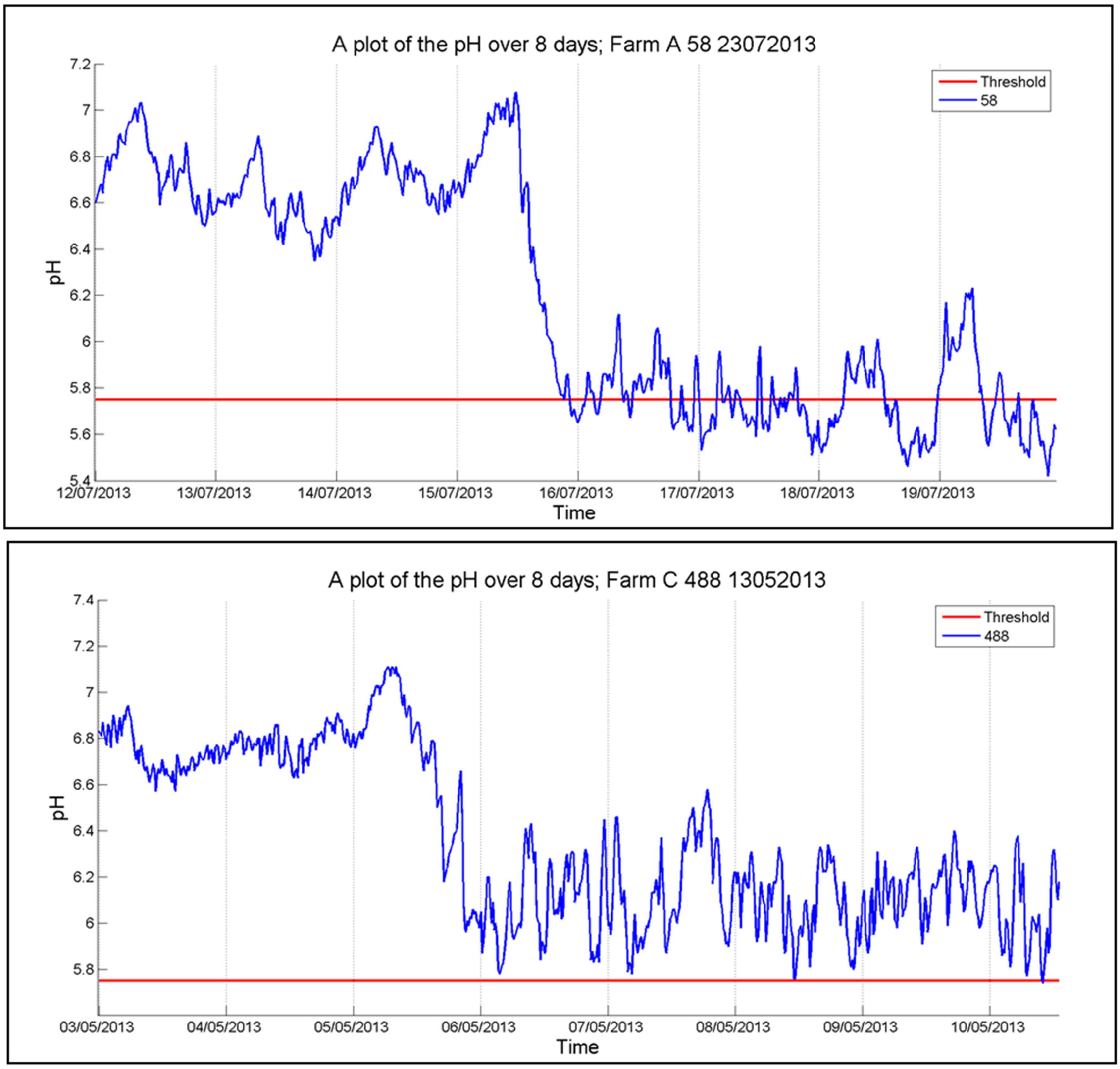

Figure 6: Graphs representing the effect of calving on the rumen pH.

Case study 1: Farm B

Farm B changed the ration twice over the trial's period. First change was on the May 17th as a result of the data recorded by the boluses. The new diet changed the concentrate/forage ratio in term of dry matter intake (DMI) from 1.27 to 1.19 but with the same intake (±0.3 kg). Table 3 shows the record of diet changes.

	Farm B 10/05/2013 (heifers + high)				Farm B 17/05/2013 (heifers + high)				Farm B 17/06/2013 (heifers + high)		
	kg fresh	%ration	DMI (kg)	%DM	kg fresh	%ration	DMI (kg)	%DM	kg fresh	%ratio	
Forage	37.1	68.6	11.2	30.2	38	69.8	11.6	30.5	45.5	76.2	13
Concentrate	17	31.4	14.2	83.5	16.4	30.2	13.8	84.1	14.2	23.8	12.4
Total	54.1	100	25.4		54.4	100	25.4		59.7	100	25.4
Diet cost (£/day)			2.98				2.83				2.23

Table 3: *Table of the ration changes.*

The figure 7 a) represents the daily mean pH over this stage. It shows the effect in daily mean pH for the cows, due to the change in diet on the May 17th. Five days after the new ration was introduced, the cows were less at risk of acidosis. New composition of the feed was £0.15/day/cow cheaper than the previous one. They kept this ration until the June 17th. Consequently, they saved £0.15/cow for 31 days which makes £4.65/cow for this period. Table 1 recorded 350 milking cows for this farm. Total benefit in feed for this month is £1,627.50.

On the 17th of June, a second change in diet occurred. Figure 7 b) is a plot of the mean daily pH over this step. The new diet modified the concentrate/forage ratio form 1.19 to 0.95 and also increased the total intake of 5kg (±0.3kg) with an augmentation in forage intake. One week after this adjustment in feed, one cow was safer from acidosis risk and the others were still in a safe pH range. Table 3 indicates that this new diet was £0.60/day/cow cheaper than earlier diet. This ration is still the current feed at the farm, which makes approximately a two months interval. Therefore, the period is 62 days, saving £0.60/cow equal £37.2/cow. This totalled a benefit in feed for the farm of £13,020 in two months.

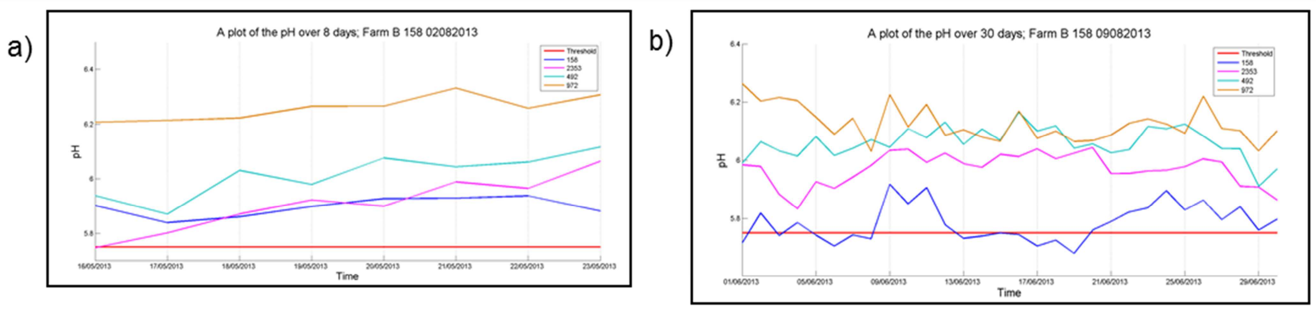

Figure 7: Mean daily pH over the first change in feed a) and the second change in feed b) for farm B.

Covering the whole period of the trial, the farm changed the ration twice and made it cheaper each time without increasing the risk of SARA for the cows. Records in milk for this period didn't show any decline in milk production or milk composition compared to the records of the year before.

In total they saved £14,647.50 in three months by changing the cost of the ration and looking at the pH level of the cows.

Figure 8 represents the pH for cow 492. This cow was moved from her group to the bull group on the July 21st. The only changes between groups were feeding time and milking time. Figure 8 b) shows that the availability of feed changed after the 21st and the cow ate later than before. In figure 8 a) and c) it is shown than the daily mean pH hadn't been changed by this movement but the cow was less at risk of SARA due to the increase of the pH nadir each day. This increase in pH meant the cow spent less or even no time under the threshold and, in term of output, has been translated to two more litres of milk per day.

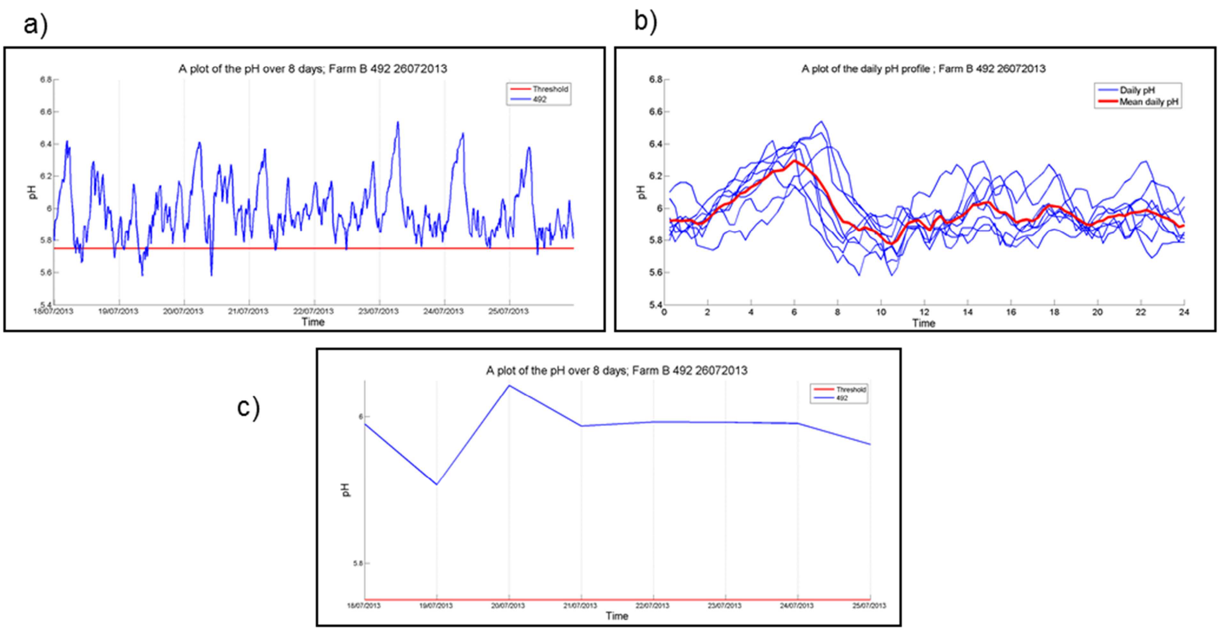

Figure 8: Graphs representing the pH of a cow that changed group.

Case study 2: Farm E

Grass management is difficult in summer time and results from Farm E show that a small change can increase the milk yield. Figure 9 represents the pH over 9 days for one cow and all verticals lines characterise midnight. So from May 29th to June 1st, the daily pattern for this cow starts with evening falls in pH as this is when the cow feeds. From the 2nd of June, this daily pattern change and the morning feed became the biggest meal of the day. This was caused by a change in management, the electric fence was usually moved in the evening but from the 2nd, it was moved in the morning. The pH decreased with this change which indicates a higher intake for this cow. She prefers to be fed in the morning because she eats more and gives more milk. Her yield also increased following the change in management and due to increased intakes as observed in the graph. A yield response of 5 more litres per day has been recorded for this animal.

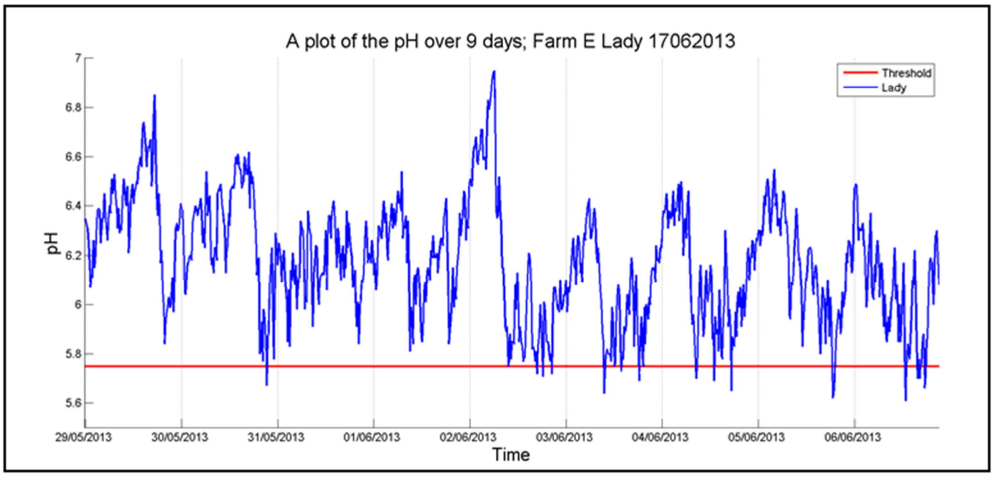

Figure 9: PH of grazing cow impact by the time fence moving

As in farm B, the time of feeding had an impact on the cow and in both cases, a positive response from the animals has been seen. However, in this case, increased intakes has pushed the cow's pH down and therefore augmented the risk of SARA.

Case study 3: Farm G

Farm G is also a grazing herd and data from this farm showed the impact of the weather and management on this type of herd. Figure 10 represents the pH recorded on four cows for a period of 8 days. Figure 10 a) and figure 10 b) represent the pH and the daily mean pH respectively. In figure 10 a) it is noticeable that the pH over the day 27th of May was higher and less variable than the other days and the 28th has been recorded as a big drop in pH for the four cows. Mean daily pH has also increased for all cows on the 27th following by a fall on the 28th. Results of this variation in pH and mean daily pH led to a decrease in milk of 2.5 litres for each cow on the 29th. Cow 950 took 4 days to get back to a pH above the threshold. The explanation from the vet and farmer was that the 27th had been a heavy rain day. Therefore cows spent time under the hedges and didn't eat a lot of grass this day but ate more the next morning.

The cows ate more on the 29th morning which explains a bigger drop than days before. However, as seen on figure 10 a) there is a smaller afternoon drop as the days before for each cow. Looking at the farmer records shows that cows were held back in the yard until all were finished milking because they had to cross a main road. The results suggest the cows ate less as a result of this disruption to their daily routine, they prioritised rest when they finally got to the field. Aside from the financial impact, a 1.5 litres drop in yield per head was recorded the next day.

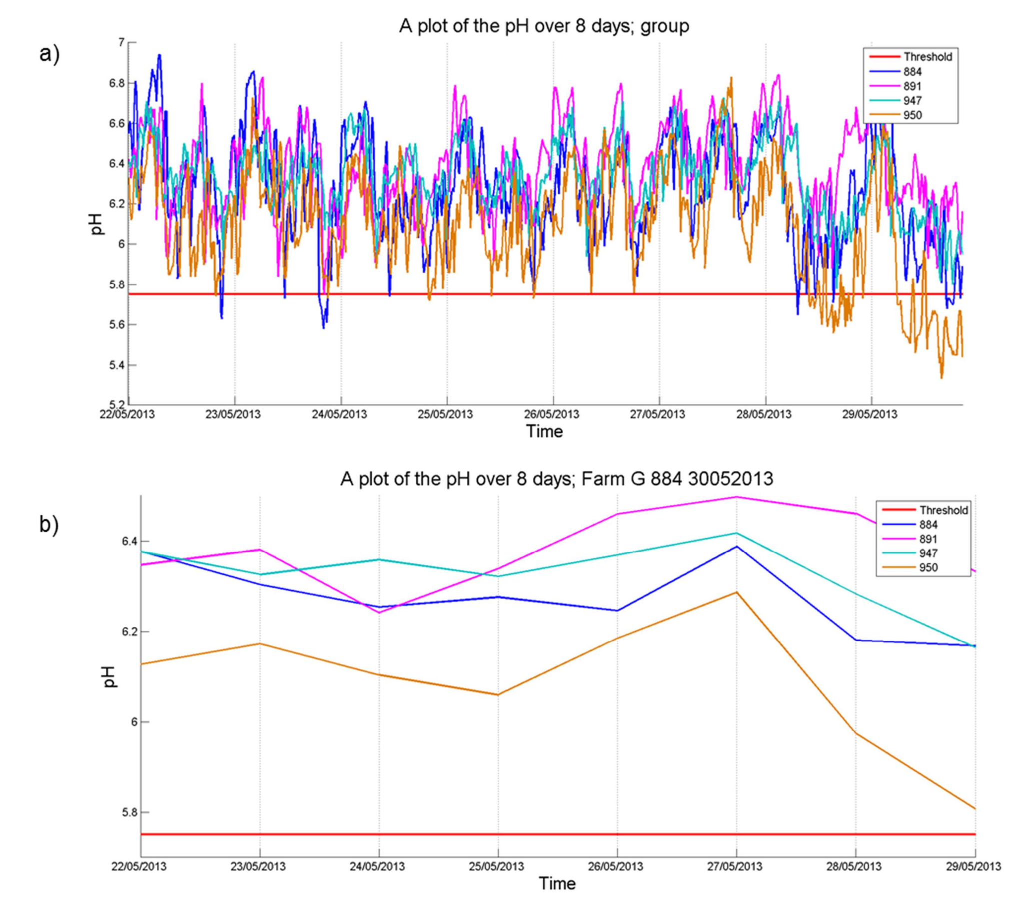

Figure 10: Graphs of pH a) and daily mean pH b) for four grazing cow

Survey

Surveys were sent to six farmers and eight advisers involved in the farms of this trial. Two farmers didn't receive the survey because their email addresses were missing. Percentages of response are 83.3% per cent for both farmers and advisers. Example of questions asked in both surveys is in annexes.

Results from surveys indicate that all farmers and advisers are familiar with smartphones but only 60% of farms have a Wi-Fi connection, which could affect the data transmission from farms to the server. 80% of farmers would like to get a weekly data stream instead of 40% for the advisers who consider that a monthly data analysis or on demand will be sufficient. None of farmers have been dissatisfied by the bolus technology and half of them are very satisfied. For the moment, only one farm has the project to use boluses continually in the future while the other will have an infrequent use. On the other hand, all vets and nutritionists have answered to advise farms to get boluses only sometimes. Their criteria to choose a farm where boluses could be placed are mainly herds with SARA problems and high yielding cows. 80% define the herd size to not enter on the choice and 60%

look at the herds with a milk yield over 9 000 litres/head/year whereas the rest doesn't find it important.

The pH graph in both forms, time line and daily profile, is the best tool in the analysis for 100% of advisers. These graphs allowed farmers, vets and nutritionists to discover the pH into their cows. For all of them who answered the question, the best learning was the feeding behaviour by means of a daily routine that can be easily disturbed and cannot be predicted with the ration.

60% of the persons questioned have seen or think that it will be a monetary benefit for farmers to use boluses because they help to appreciate the risk of SARA and consequently increasing the production and/or reducing the cost of SARA in the long term. Furthermore, all advisers have seen a commercial profit for them or their company using the boluses. The technology enables them to have better discussions with farmers, between each other and add value to their service. Therefore, every farm's consultant who answered this survey said that the wireless rumen pH telemetry could be a part of the services they provide to farmers.

Results from surveys have been positive and both farmers and advisers seem to appreciate the continuous monitoring provided with the FarmBolus. A wish to link the data to a database with all the events and milk yields has been manifested.

Discussion

Bolus and handset performances

The changeover from the tablet computer to the phone has been a total success. Nowadays, most of people use a phone and it is easier for them to understand how to download the data on the bolus when presented with a phone rather than a computer. Additionally, handling is better and easier, just one hand is needed and it is designed to be robust and waterproof against splashes in contrast to a computer. First overview of the pH line is directly available at the end of the downloading which is appreciated by farmers and vets. The new handset, for the moment, features all the bolus settings with the exception of the frequency of data storage, which is a set every 15 minutes. Inversely, it adds more settings than the Samsung Q1 for adjusting the minimum time it will take before bolus communication.

This trial has proved that farmBolus has the longest life available for the moment, which goes up to five months if managed correctly. Results from surveys showed that farmers and vets are satisfied about the product. Vets and nutritionists found it really interesting to use boluses. "*It is really useful to build up a picture of normal daily pH fluctuations on commercial farms in the South West. The pH bolus trial results have given us this insight, and highlighted the nutritional impact of management changes which force cows into unnatural daily routines.*", (J. Hamilton, eCow website, http://www.ecow.co.uk/three-counties-feeds-write-up/ July 13th 2013). Twitter is also a platform where professionals were talking about their satisfaction to have boluses in some farms. Results from the survey indicate that they will probably add rumen pH monitoring as a service provided to farm.

Figure 11 shows the new packaging solution for boluses which was developed as a result feedback from the trials. The new system keeps the sensors wet while protecting the boluses.

Figure 11: Pictures of the new method of packaging and storage.

Data

Regarding data collection, the data analysis has been improved and the time to get the data analysis has been reduced and is now less than one day. Development is in progress with Matlab which allows analysis of a file without opening it and to produce all types of graphs as well as statistical analysis in a small interval of time. In the case of this study, the files were opened before using the software because of defining the date intervals in the analysis and only getting full days. This manual check and modification of files enables simultaneously putting together previous files as the bolus overwrites the data when there is more than 2700 lines.

After having been produced, graphs need to be put together in Portable Document Format (PDF) for each cow and send by email to farmer and their advisers. The company has already found a new way of data presentation by the use of an internet web page. Farmers and advisers will have to login in to a website where all the graphs will be on one page as thumbnails. Clicking on it will make the graph bigger and easier to look at. This new approach for data presentation will be more convenient for customers. Moreover, it will be interesting to link the pH results with the milk production and other information to facilitate interpretation as seen in the survey's results.

On pH graphs, acidosis threshold has been defined as pH 5.75. The reason for this choice was that rumenocentesis was the previous technique to collect rumen samples and the SARA threshold was set at pH 5.5. Studies have found a difference between the pH in the reticulum and the pH in the rumen (Kimura et al, 2012; Lane et al; 1968; Tafai et al, 2004).Considering the results of these studies, a pH of 5.75 has been defined as the edge for SARA. However, the goal chosen in this trial was to define the risk of SARA and not its presence. This means that when the pH is under the threshold, a specialist needs to check the cow before defining the presence of SARA.

Over the trial period one cow always had a low pH and spent up to 20 hours a day under the threshold (see figure 12). A vet checked the cow several times and never found any sign to determine the presence of SARA. Furthermore, the farmer didn't find any health problem or change in milk content or milk yield. Dave Humphries, researcher at University of Reading, said the shape of the reticulum is different from one cow to another (personal communication, 29 June 2013). Hence, it is assumed that the bolus, in some cows, is closer to the centre of the rumen so closer to the reaction site where pH is lowest in the rumen. That could explain why the cow has a large difference in pH compared to the other one in the group. It is known that SARA is a subtle condition and difficult to diagnose. Signs of SARA could appear several months later and then the cow may be in acidosis in the time of recording. Nevertheless, SARA is a digestive disorder and in this way, modifying the degradation of nutrients and so the pattern of the pH line. Figure 12 shows the two cows in the same group. Cow 158 has a really low pH but the variations during the day are similar to cow 972. Then, because this case is the only case in the trial, the second assumption is believed to be the most probable, even if there is no other way than a fistulated cow to prove that the reticulum shape differs from cow to cow.

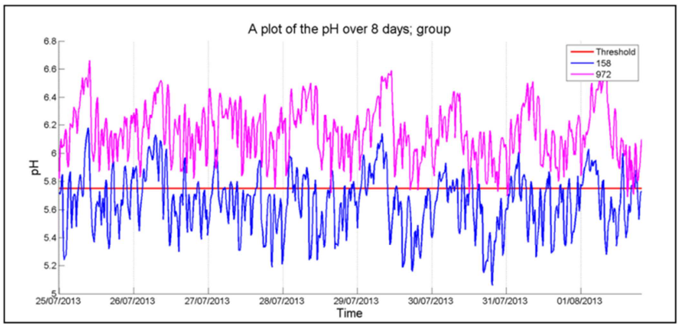

Figure 12: Graph of pH for two cows in the same group.

The diversification of the farms involved in the experiment enables recording of different daily patterns and their effects on pH value. Figure 13 shows graphs representing different types of feeding and management. Graph A represents the daily pH profile of a cow fed TMR once a day. The big drop is the feeding time for this cow and represents around 0.7 pH units. In contrast, graph B represents the daily pH profile of a grazing cow milked twice a day. There is more variation in pH due to longer interruptions for milking. Then the last graph represents the daily pH of a cow in a robotic milking farm. There is a regular daily cycle and even if it still has a main drop in pH, the variation during the day is around 0.3 pH units. This is because the cow is not interrupted for milking and she has little feeds often, resulting in low swings in rumen pH. Diverse events have also been recorded and an example of calving impact on pH and temperature is in figure 6.

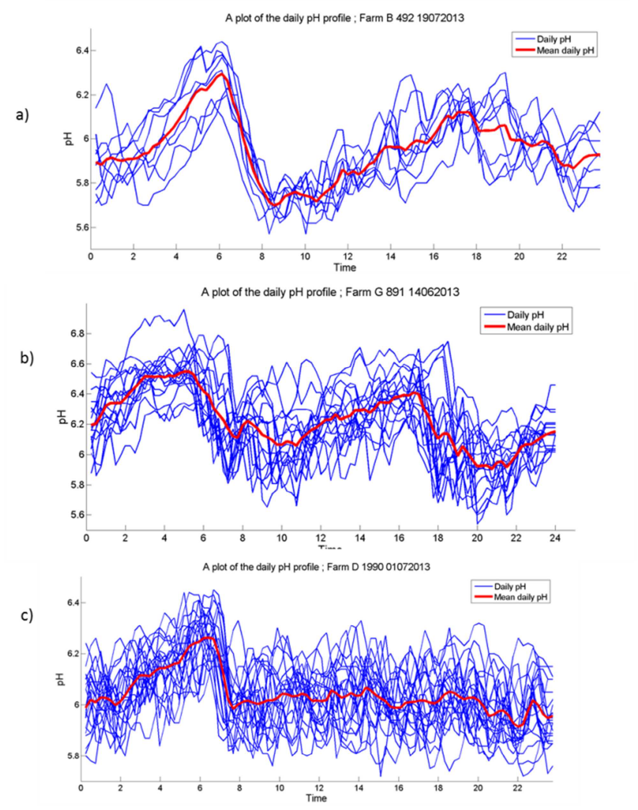

Figure 13: Graphs representing the different daily pattern: a) feed once, b) feed twice (grazing) and c) is the effect of robot milking

Benefits

Monitoring the rumen pH in cows is an important technology now available for all commercial farms. However, to be appreciated by everyone, the wireless rumen pH telemetry need to be beneficial for all persons involved. That goes from the manufacture company to the farmer, through his advisers. Toby Mottram, founder of eCow Ltd and inventor of the farmBolus, said: "*We have developed the farmbolus to provide a technology to help the dairy industry. Everyone needs to find a benefit from it otherwise I would not make it.*" (Personal communication, July 2013).

Results from this trial have shown different ways and methods to make everybody benefit and that is because boluses measure more than acidosis. As Three Counties Feed said on Twitter, cows are very sensitive to management changes and boluses record all the information to know how to look after them better. Boluses mainly record the feeding pattern and working with nutritionists allows an understanding that they can measure the energy intakes and in somehow the digestible energy. Obviously, a bolus doesn't measure quantitatively but it gives a qualitative approach that is an essential knowledge. Professionals say there are three rations on a farm, the first one is on paper, the second is the one the farmer feeds cows and the last one is the ration cows eat. The rumen bolus is measuring the last one, the most important one that has always been unknown in commercial farms. More than recording the energy intake, the bolus is about improving the rumen function and in this way, factors are not only the diets but many other variables.

Farmers involved in this trial learned new information about their cows and by getting the data have been able to test different ways to produce the most cost effective way without pushing cows over the edge of SARA. Main results from this experiment show that the rumen behaviour is complex and a multitude of actions could have an impact on it. First is the ration, principal thinking when cows show problems of acidosis. The example of farm B, showed in the case study, demonstrates the huge amount of money that can be wasted by giving an excessive amount of concentrate. In this case, decreasing the concentrate intake has reduced feed cost, increased the pH of cows which means the risk of SARA is reduced without modifying the milk output. A second change in feed, made due to being out of stock of one ingredient, would be made without bolus information and probably saved money as well. However, the concentrate ratio would approximately stay the same as before which was putting the pH of the cow too low.

In the same way, mechanisation issues could be detected. It is well known that the degree of mixing is important in nutrition and boluses could be used to detect which mixing degree or even which mixing wagon is the best to feed cows properly. This case is more adapted to TMR fed cows but grazing herds can display the symptoms of SARA and not only when winter ration is fed (Mulligan and Rafferty, 2013). As seen in the case study, grazing herd issues can be identified with rumen boluses. This problem is more about grass availability and at what time. Changing the main meal from the afternoon to the morning has made cows happier and resulted in 5 litres more per cow per day. Time of feeding is consequently important and that has been seen in grazing herds as well as fully housed herds (see cases study). Another management problem showed by boluses is the staff work. In fact, further than seeing if cows have been fed or not, boluses give an indication about how it has been done. In different farms, cows’ pH has differed dramatically from one day to another for short periods of 2-3 days which has been reflected on milk yield. Often these days were on a weekend and a conclusion has been made by farmers that on this weekend, someone else instead of usual staff was milking and/or feeding cows. The last main result for farms from this trial is the detection of heat stress. Regarding two farms, it has been observed that some cows had a lower pH than other during the month of July when a heat wave affected the south of England. They were on the same ration and no change occurred in feed, the only difference the nutritionist found was the housing shed. He noticed that one shed is less well ventilated and so cows panted, which meant no rumination and drooling saliva instead of swallowing it so the buffer effect was lost. This resulted in a gap of 10 litres minimum between cows in different shed during this period.

The results of data collection have shown a commercial benefit for dairy farmers but they are not the only ones to profit from this technology. One benefit for farms' consultants in this trial has been learning that SARA is not only due to the ration. Whilst properly formulated rations are what nutritionists spend so many hours working on, the complexity of the rumen environment needs to be appreciated. The contents of the ration influence rumen pH but many other factors do as well. Rumen pH is not a static number, it is constantly going up and down. This means by looking at the pH of cows, advisers have been able to engage in better discussion with farmers about herd management, feeding management and production but also between each other, as vets/nutritionists, about the herd health. As surveys showed, specialists in dairy farm found a commercial interest to add the rumen pH monitoring at their service to farms. One reason is that dairy specialists using such a technology, in addition to the benefit of being closer to the farm and its management, have the advantage of measuring one of the most important aspects in dairy cow and so get involved in new farms with new customers because they provide a better service. Data needs to be interpreted by professionals in cow nutrition or health to get the best information from them and that will make the difference in service quality.

Both farmers and advisers appreciate the rumen pH monitoring and found great learning from it. Some didn't evaluate any commercial benefit from this learning but results are here and the economic impact, if not direct, will be seen in the long term. Difficulties to get prices on the internet enables the comparison with only one other company who makes boluses. Averaging the herd size and milk yield for all farms of the trial give an average farm of 400 cows and 11 000 kilograms of milk/cow/year. The competitor on his website advises to put five boluses all year round which make a total of 20 boluses as they last 50 days. The only calculator displays a cost of 820€/month plus 796€ for the reader. In comparison, because the farmBolus lasts up to five months, a total of 15 boluses will be sufficient to monitor more than one year for five cows. The cost will then be £6 750 for the boluses plus £700 for the reader. A total cost of 10 636€ is predicted for the competitor while £7 450 for eCow which makes 8 694 € with a conversion rate of 1.167. Consequently, for the same number of cows monitored during one year, the products provided by eCow is almost 2 000€ cheaper. That calculation proves the competitiveness of eCow Ltd on the rumen pH monitoring market.

Conclusion

A trial has been conducted on eight commercial dairy farms in the South West of England to prove the commercial benefit of rumen pH monitoring. Results from it have demonstrated different ways to overcome challenges in dairy industry. Every type of farm has been susceptible to changes and benefits. They also showed that improvements on farms are only possible when alternative solutions are presented and technically possible.

This experiment has proven the quality of such technology and its positive impact on farms as well as in the dairy sector. Great learning has been made and it is only the starting point for the wireless rumen pH monitoring.

Annexe 1: Survey for farmer.

This survey is made up of 14 questions. Please make sure you have read all the questions, the answers will help us in our development.

About the farm

Question 1: How many milking cows do you have?

☐ 0-100 ☐ 100-200 ☐ 200-300 ☐ 300-400 ☐ 400+

Question 2: What system do you use on farm? (e.g. Dry cows grazed 50% then come in for evening feed, high yielders fully housed TMR, etc.)

Question 3: Do you have a smartphone? ☐ Yes ☐ No

Question 4: Do you use Wi-Fi on farm? ☐ Yes ☐ No

About the boluses

Question 5: How many boluses do/did you use?

Question 6: How often did you get data?

☐ Every week ☐ Every two weeks ☐ Every month ☐ On Demand
☐ Never

Question 7: What frequency would you like data?

☐ Every milking ☐ Daily ☐ Weekly ☐ Monthly
☐ On Demand

Question 8: Who interpreted and discussed the data with you?

☐ Nutritionist ☐ Vet ☐ Self ☐ Other:

Question 9: How often will you use boluses in the future?

☐ Never ☐ Sometimes ☐ Frequently ☐ Continually

Comments

Question 10: What was the greatest learning for you in the data you reviewed?

Question 11: How satisfied are you?

Very Dissatisfied	Dissatisfied	Neutral	Satisfied	Very Satisfied
☐	☐	☐	☐	☐

Question 12: What did you change as a result of the data provided?

☐ Forage ☐ Concentrate ☐ Time of feeding ☐ Micronutrients

☐ Other:

Question 13: Have you seen any monetary benefit to using the boluses?

☐ Yes ☐ No

Question 14: If yes, could you explain?

Other comments:

Thank you!!

Annexe 2: Survey for advisers.

This survey is made up of 16 questions. Please make sure you have read all the questions, the answers will help us in our development.

About the farms

Question 1: At how many farms do you use boluses?

Question 2: What is/are your criteria to advise farms to get boluses?

Question 3: What farms would you advise to get boluses?

Herds over	☐100	☐ 200	☐ 300	☐ 400	☐ 500+	☐ Doesn't matter
Herds yielding	☐ under 9 000	☐ 9 000-12 000	☐ more than 12 000			☐ Doesn't matter
Herds type	☐ Grazing	☐ Partially grazing	☐ Fully Housed			☐ Doesn't matter

About the boluses

Question 4: How often did you get data?

☐ Every week ☐ Every two weeks ☐ Every month ☐ On Demand
☐ Never

Question 5: How often would you like data?

☐ Every milking ☐ Daily ☐ Weekly ☐ Monthly
☐ On Demand

Question 6: How often would you advise farms to use boluses in the future?

☐ Never ☐ Sometimes ☐ Frequently ☐ Continuously

Question 7: How many boluses would you recommend to have in a group?

☐1 ☐2 ☐3 ☐4 ☐More

Question 8: What is/are the best tool(s) in our analysis?

☐ Time line ☐ Temperature ☐ Daily pH ☐ Dashboard

Question 9: What would you like as part of analysis?

Comments

Question 10: What was the greatest learning for you in the data you reviewed?

Question 11: What conversation do/did the boluses enable you to have with farmers? And with other vet/nutritionist?

Question 12: Have you seen any monetary benefit to using the boluses for the farmer?

☐ Yes ☐ No

Question 13: If yes, could you explain?

Question 14: Did you or your organisation find it beneficial to work with the farmers with the boluses data?

☐ Yes ☐ No

Question 15: If yes, could you explain?

Question 16: Do you think the pH data analysis could be part of your services to the farm?

Other comments:

Thank you!!

References:

Alzahal O., B. Rustomo, N.E. Odongo, T.D. Duffield, B.W. McBride, 2007, **Technical note: a system for continuous recording of ruminal pH in cattle.** Journal of Animal Science, 85, pp. 213–217.

Beauchemin KA, Yang WZ (2005) **Effects of physically effective fibre on intake, chewing activity, and rumen acidosis for dairy cows fed diets based on corn silage.** Journal of Dairy Science 88, 2117–2129.

Beghelli, V., G. Borghi and R. Matscher, 1958: **Influenza delle sostanze azotate alimentari sul pH del contenuto ruminale.** Arch. Vet. Ital. 9, 97–112.

Calsamiglia S., A. Ferret, M. Devant. **Effects of pH and pH fluctuations on microbial fermentation and nutrient flow from a dual-flow continuous culture system.** J. Dairy Sci., 85 (2002), pp. 574–579.

Cunningham JG (2002) 'Textbook of veterinary physiology.' 3rd edn. (WB Saunders: Philadelphia)

Commun. L, 2011. **Physiopathologie de l'acidose subclinique.** Le Point Vétérinaire, 321 : 56-57.

Cottee G, Kyriazakis I, Widowski TM, Lindinger MI, Cant JP, Duffield TF, Osbourne VR, McBride BW (2004) **The effects of subacute rumen acidosis on sodium bicarbonate-supplemented water intake for lactating cows.** Journal of Dairy Science 87, 2248–2253.

Dado, R. G. and M. S. Allen, 1993: **Continuous computer acquisition of feed and water intakes, chewing, reticular motility, and ruminal pH of cattle.** J. Dairy. Sci. 76, 1589–1600.

Dirksen G., H.G. Liebich, W. Mayer. 1985, **Adaptive changes of the ruminal mucosa and their function and clinical significance.** Bovine Practitioner, 20 (1985), pp. 116–120.

Dirksen, G., 1986: **Differentialdiagnostik und Therapie von Vormagen- und Labmagenerkrankung bei Kalb und Jungrind.** Prakt. Tierarzt, 69, Coll. Vet. XVIII, 92–96.

Duffield TF, Leslie KE, Sandals D, Lissemore K, McBride BW, Lumsden JH, et al. 1999, **Effect of a monensin controlled release capsule on cow health and reproductive performance.** J. Dairy Sci. 1999;82:2377–2384.

Duffield T., J.C. Plaizier, A. Fairfield, R. Bagg, G. Vessie, P. Dick, J. Wilson, J. Aramini, B.W. McBride. 2004, **Comparison of techniques for measurement of rumen pH in lactating dairy cows.** Journal of Dairy Science, 87 (2004), pp. 59–66.

Enemark, J.M.D., Jorgensen, R.J., Kristensen, N.B., 2004. **An evaluation of parameters for the detection of subclinical rumen acidosis in dairy herds.** Veterinary Research Communications, 28(8): 687-709.

Enemark JMD, Peters G, Jorgensen RJ (2003) **Continuous monitoring of rumen pH – A case study with cattle.** Journal of Veterinary Medicine 82, 1171–1178.

Garrett, E.F., K.V. Nordlund, W.J. Goodger and G.R. Oetzel, 1997. **A cross-sectional field study investigating the effect of periparturient dietary management on ruminal pH in early lactation dairy cows.** J. Dairy Sci., 80: 169-169.

Garrett, E. F., M. N. Pereira, L. E. Armentano, K. V. Nordlund, and G. R. Oetzel, 1995: **Comparison of pH and VFA concentration of rumen fluid from dairy cows collected through a rumen canula vs. rumenocentesis.** J. Dairy Sci. Supplement 1, Abstract 78, 299 pp.

Garrett, E.F., M.N. Perreira, K.V. Nordlund, L.E. Armentano, W.J. Goodger and G.R. Oetzel, 1999.

Diagnostic methods for the detection of subacute ruminal acidosis in dairy cows. J. Dairy Sci., 82: 1170-1178.

Gasteiner J., T. Guggenberger, J. Häusler, and A. Steinwider, 2012. **Continuous and long term measurement of reticuloruminal pH in grazing dairy cow by an indwelling and wireless data transmitting unit.** Vet. Med. Int., 236956.

Graf CM, Kreuzer M, Dohme F (2005) **Effects of supplemental hay and corn silage versus full-time grazing on rumen pH and chewing activity of dairy cows.** Journal of Dairy Science 88, 711–725.

Jørgensen, R. J., R. Erdman, M. Murphy, A. C. Olsson, J. Foldager, P Nørgaard, P. D. Møller, P. H. Andersen, I. Nielsen, O. Østera˚ s, Harmoinen, K. Kadarik, M. Emanuelson, and E. Reintam, 1993: **Rumen acidosis: identification of potential areas of research. Summary of group discussion.** Acta vet. Scand. Suppl. 89, 153–154.

Keunen, J. E., J. C. Plaizier, L. Kyriazakis, T. F. Duffield, T. M. Widowski, M. I. Lindinger, and B. W. McBride. 2002. **Effects of a subacute ruminal acidosis model on the diet selection of dairy cows.** J. Dairy Sci. 12:3304–3313.

Kleen, J.L., 2004. **Prevalence of subacute ruminal acidosis in Deutch dairy herds-A field study.** Ph.D. Thesis, School of Veterinary Medicine Hanover, pp: 93-104.

Kleen, J. L.; Hooijer, G. A.; Rehage, J.; Noordhuizen, J. P. 2003, **Subacute ruminal acidosis (SARA): a review. Journal of veterinary medicine.** 2003; 50: 406-414.

Kleen, J. L.; Hooijer, G. A.; Rehage, J.; Noordhuizen, J. P. 2004, **Rumenocentesis: A viable instrument in herd health diagnosis.** Dtsch. Tierarztl. Wochenschr., 2004, 111: 458-462.

Kimura A., Sato S., Goto H., Yamagishi N., Okada K., Mizuguchi H., Ito K. 2012, **Simulataneous estimation of the pH of Rumen and Reticulum Fluids of Cows using a Radio-transmission pH-Measuremnt System.** J. Vet. Med. Sci, 2012; 74(4): 531-535.

Krause, K.M., Oetzel, G.R, 2006, **Understanding and preventing subacute ruminal acidosis in dairy herds: a review.** Anim. Feed Sci. Tech. 126, 215-236.

Lane, G. T., Noller, C. H., Colenbrander, V. F., Cummings, K. R. and Harrington, R. B. 1968. **Apparatus for obtaining ruminoreticular samples and the effect of sampling location on pH and volatile fatty acids.** J. Dairy Sci. 51: 114–116.

Lampila, M., 1955: **Preliminary studies on the variations of pH and volatile fatty acid concentration in the rumen contents of the cow.** J. Sci. Agric. Soc., 27, 142–153.

Lampila, M., and E. Poutiainen, 1966: **Systematic differences in the composition of the bovine rumen fluid between different parts of the rumen.** Ann. Agric. Fenn. 5, 68–74.

Lean, I. J., L. K. Wade, et al., 2000. **New Approaches to Control of Ruminal Acidosis in Dairy Cattle.** Asian-Australasian Journal of Animal Sciences 13(Suppl): 266-269.

Maekawa M, Beauchemin KA, Christensen DA (2002) **Chewing activity, saliva production, and rumen pHof primiparous and multiparous lactating dairy cows.** Journal of Dairy Science 85, 1176–1182.

Matscher, R., 1958: **Sul potere di neutralizzazione del contenuto ruminale verso gli acidi e gli alkali rapidamente imissi nel rumine**. Arch. Vet. Ital. 9, 113–124.

Matscher, R., G. Borghi, and V. Beghelli, 1957: **Sulla technica di determinazione del ph del contenuto ruminale.** Arch. Vet. Ital. 8, 513–534.

Mottram T., Lowe J., McGowan M., Phillips N., 2008, **A wireless telemetric method of monitoring clinical acidosis in dairy cows.** Computers and Electronics in Agriculture, 2008; 64: 45-48.

Mottram T., McCubbine J., Nimmo S.B., 2013. **A novel method of calculating a SARA index by wireless rumen pH telemetry.** Join European Conference on Precision Livestock Farming, September 10th-11th-12th, 2013 – Leuven, Belgium.

Mulligan F., Rafferty D., 2013. **SARA in the summertime.** Cow management , volume 11, 4, April/May 2013, 18-19.

Mutsvangwa T. and T. Wright (2003, April). “Sub-Acute Ruminal Acidosis (SARA) in Dairy Cows”. Ontario, Ministry of Agriculture and Food. http://www.omafra.gov.on.ca/english/livestock/dairy/facts/03-031.htm (15 June 2013).

Nocek, J. E. 1997. **Bovine Acidosis: Implications on Laminitis.** Journal of Dairy Science 80(5): 1005-1028.

Nordlund, K. V., (1996), **Questions and answers regarding rumenocentesis and the diagnosis of herd-based subacute rumen acidosis.** Proc. Annu. Conf. Am. Assoc. Bovine Pract., pp: 75-81.

Oetzel, G. R. (2007), **Subacute Ruminal Acidosis in Dairy Herds: Physiology, Pathophysiology, Milk Fat Responses, and Nutritional Management.** Preconference Seminar 7A: Dairy Herd Problem Investigation Strategies: Lameness, Cow Comfort, and Ruminal Acidosis, AMERICAN ASSOCIATION OF BOVINE PRACTITIONERS, 40th Annual Conference, September 17, 2007 – Vancouver, BC, Canada.

Owens, F. N., D. S. Secrist, et al., 1998. Acidosis in Cattle: A Review. Journal of Animal Science 76: 275-286.

Peters, G., 1997b: **A new device for monitoring gastric pH in freeranging animals.** Am. J. Physiol. 273, 748–753.

Penner GB, Beauchemin KA, Mutsvangwa T (2006) **An evaluation of the accuracy and precision of a stand-alone submersible continuous rumen pH measurement system.** Journal of Dairy Science 89, 2132–2140.

Phillips N., Mottram T., Poppi D., Mayer D., and McGowan M.R., 2010, **Continuous monitoring of ruminal pH using wireless telemetry.** Animal Production Science, 50, 72-77.

Plaizier, J. C., A. Martin, T. Duffield, R. Bagg, P. Dick, and B. W. McBride, 1999: **Monitoring acidosis in the transition cow.** J. Dairy Sci. 82 (Suppl. 1), 110.

Rustomo B, Al ZahalO(2006) **Acidogenic value of feeds. II. Effects of rumen acid load from feeds on dry matter intake, rumen pH, fibre degradability and milk production in the lactating dairy cow.** Canadian Journal of Animal Science 86, 119–126.

Smith, V. R. 1941: **In vivo studies of hydrogen ion concentrations in the rumen of the dairy cow.** J. Dairy. Sci. 24, 659–665.

Shaver, R. D., 1997. **Nutritional Risk Factors in the Aetiology of Left Displaced Abomasum in Dairy Cows: A Review.** Journal of Dairy Science 80: 2449-2453.

Tafaj, M., Junck, B., Maulbetsch, A., Steingass, H., Piepho, H. P. and Drochner, W. 2004. **Digesta characteristics of dorsal, middle and ventral rumen of cows fed with different hay qualities and concentrate levels.** Arch. Anim. Nutr. 58: 325–342.

Tajik, J., M.G. Nadalian, A. Raoofi, G.R. Mohammadi and A.R. Bahonar, 2009. **Prevalence of subacute ruminal acidosis in some dairy herds of Khorasan Razavi province, northeast of Iran.** Iranian J. Vet. Res., 10: 28-32.