

# Comparison of different measuring methods for body temperature in lactating cows under different climatic conditions

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The aim of the research described here was to compare different methods of body temperature (BT) measurements in dairy cows. It was hypothesised that reticular temperature (RET) values reflect the physiological status of the animals in an equivalent way to rectal (RT) and vaginal (VT) measurements. RT, VT and RET temperatures of twelve lactating Holstein–Friesian cows were measured over five consecutive days in June and October 2013. While RT and VT were manually measured three times a day, RET was automatically recorded at 10 min intervals using a bolus in the reticulum. For comparison with RT and VT, different RET values were used: single values at the respective recording times (RET-SIN), and mean (RET-MEAN) and median (RET-MED) values of 2 h prior to RT and VT measurements. Overall, body temperatures averaged  $38.1 \pm 0.6$ ,  $38.2 \pm 0.4$ ,  $38.7 \pm 0.9$ ,  $38.5 \pm 0.7$  and  $38.7 \pm 0.5$  °C for RT, VT, RET-SIN, RET-MEAN and RET-MED, respectively. RT and VT were lower than all RET measurements, while RET-SIN and RET-MED were higher than RET-MEAN ( $P < 0.001$ ). RET-MEAN and RET-MED values were higher in the morning, whereas RT and VT were greatest in the evening ( $P < 0.001$ ). Overall, records of RT and VT were strongly correlated ( $r = 0.75$ ;  $P < 0.001$ ). In contrast to RET-SIN and RET-MEAN, RET-MED was higher correlated to RT and VT. In June, coefficients were higher between all methods than in October. Relation of barn T to RT and VT was stronger when compared to RET measurements. RET-SIN was higher correlated to barn T than RET-MEAN or RET-MED. Correlation between VT and barn T was strongest ( $r = 0.48$ ;  $P < 0.001$ ). In summary, RET-MED showed highest correlation with VT and RT. However, single RET measurements (influenced by water or feed intake) can lead to extreme variations and differences to single VT and RT values.

**Keywords:** Body temperature monitoring, reticular temperature, rectal temperature, vaginal temperature, dairy cows.

Body temperature (BT) is regularly used by milk producers and veterinarians to monitor physiological and pathological processes, e.g. heat detection or diseases (Hicks et al. 2001; Bewley et al. 2008a; Liang et al. 2013). Continuous BT measurements could be very useful for health monitoring programs in dairy cows, however, it has to be kept in mind that BT is also influenced by various external and internal factors like season, breed, milk yield, parity, time of day, climatic conditions or water and feed intake and even by the measuring method (Bewley et al. 2008a; Suthar et al. 2012; Adams et al. 2013; Liang et al. 2013). Several studies investigated the potential and risks of different

methods and body locations for BT measurements in dairy cows (Firk et al. 2002). The various locations included the udder (Firk et al. 2002), rectum, vagina (Suthar et al. 2012; Lee et al. 2014), rumen (Bewley et al. 2008a; AlZahal et al. 2011; Adams et al. 2013; Liang et al. 2013), peritoneum (Lefcourt & Adams, 1996) and tympanum (Hahn et al. 1990; Bergen & Kennedy, 2000). Some of the methods are done manually, while others are based on automatic systems. Automatic BT monitoring systems can facilitate the identification of illness, heat stress, general physiological stress and oestrus (Fordham et al. 1988; Bewley et al. 2008a) given that the available implants and boluses enable a continuous temperature recording. In all cases the merit of the data depends on the economic costs (e.g. labour extensive) to record them, the accuracy, and the risks of injury for the animals.

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Predictions of the normal core BT of a dairy cow vary between 38.6 to 39.2 °C (Piccione & Refinetti, 2003). Other authors reported that BT ranges from 38.2 to 39.0 °C depending on the measurement methods used (Prendiville et al. 2002). Estimating the relationship between different methods, Sievers et al. (2004) found, for instance, a highly significant correlation between intraruminal and rectal temperature ( $r=0.92$ ). Bewley et al. (2008a) estimated lower correlations between reticular and rectal temperature (0.65).

The measurement of rectal temperature is relatively easy and for economic reasons is still the most commonly used (Hicks et al. 2001), but the use of rumen/reticulum boluses to monitor BT is a time- and labour-saving alternative and is, therefore, of increasing importance (Adams et al. 2013). Only automatic measurements can record changes of temperature within a short period of time which can be used as an indicator of animal health (Dye 2007; AlZahal et al. 2008), animal activity (Gebremedhin et al. 2011), changes in animal physiology such as ovulation (Culmer, 2012) and various types of animal-environment interactions (Hillman et al. 2005). However, if such methods are used e.g. for diagnostic reasons, correlations to temperatures measured with other methods and the repeatability of the values has to be known. Equations to adjust reticular temperatures to rectal- and vaginal-based scales may increase the utility of such cattle temperature monitoring systems. To the authors' knowledge direct comparisons between reticular, rectal and vaginal temperatures that consider different climatic conditions and diurnal variations are not yet available. Therefore, the objective of this study was to compare these methods of BT measuring in dairy cows during a warm and cold period. Based on this the potential, the use and limitations of reticular measurements were estimated.

## Material and methods

### Study farm and animals

The study was conducted on an experimental dairy farm in the northern part of Lower Saxony, Germany, with an average yearly rainfall of 650 mm. The animals were housed in an outdoor loose-housing barn with cubicles. One side of the barn was equipped with curtains, the other one with space boards. Additionally, two manually controlled axial fans were installed above the feed alley.

The herd consisted of 75 lactating Holstein-Friesian dairy cows with an average production of 11 000 kg milk per lactation period with 3.76% fat and 3.30% protein. From the herd 12 cows, which were in first to fourth lactation, with similar lactation stage at the start of the study ( $65 \pm 19$  d in milk (DIM)) were selected. Their average milk yield was  $36.4 \pm 10.4$  kg milk during both study periods. Milking was performed with an automatic milking system with an average number of 2.7 visits per day. Cows were fed a total-mixed ration ad libitum.

### Data collection

Data were collected between the 18th and 22nd of June and 8th and 12th of October 2013. Only procedures approved for standard use under normal farm management conditions were used. Reticular temperatures (RET) were recorded using a pH and temperature sensor (smaXtec Animal Care GmbH, Graz, Austria). The bolus weight is 208 g. It has a dimension of  $132 \times 35$  mm and contains a microprocessor, a memory space, an internal antenna and battery. The average operation time of the bolus battery is 300 d. The bolus cannot be removed (smaXtec animal care sales GmbH, Graz, Austria). The boluses were orally given in March 2013. Regarding earlier experiences it can be assumed that the bolus resided in the cow's reticulum (Bewley et al. 2008a). RET was recorded every 10 min with an accuracy  $\pm 0.25$  °C. Data were transferred once a month using the smaXtec mobile reader into an Excel file. Rectal (RT) and vaginal temperatures (VT) were measured manually by one single person using a standard digital thermometer. Measurements were done at 7.30 a.m., 12.00 p.m. and 17.30 p.m. while animals were restrained.

Barn climatic conditions, namely temperature in °C (T) and relative humidity in % (RH), were recorded at 15 min intervals using 9 Tinytag Data Loggers (Tinytag Plus 2 TGP-4500; Gemini Data Loggers Ltd, West Sussex, UK) with an accuracy of 0.01 °C and 3%, respectively. Data Loggers were placed over the feed alley, resting and waiting areas at heights of 1.2 and 3 m. For one cow data were missing for June and for two cows for the October period.

### Statistical analysis

Barn T and RH were averaged over the nine loggers. RH values lower than 25% and higher than 98% were deleted as measurement errors. Based on these averages, the temperature-humidity-index (THI) was calculated using the following formula (NRC, 1971):

$$\text{THI} = (1.8 \cdot T + 32) - (0.55 - 0.0055 \cdot \text{RH}) \cdot (1.8 \cdot T - 26)$$

Different reference periods for RET were used for comparison with RT, VT and barn T and THI. Those were either the single reticular temperature (RET-SIN) value, which was recorded closest to the RT and VT measurement as well as the mean (RET-MEAN) and median (RET-MED) values for the 2 h preceding the respective RT and VT measurement.

Statistical analysis was conducted using the statistical software package SAS version 9.3 (SAS Institute Inc., Cary N.C., USA). Arithmetic means, standard deviation, minimum and maximum values were estimated using Proc Means. Pearson correlation coefficients (Proc Corr) were calculated for paired comparisons of RT, VT, RET-SIN, RET-MEAN, RET-MED and barn T averaged and separated for both study periods and times of day.

Proc Mixed was used to analyse the effects of month and time of day on RT, VT and RET values with the following

**Table 1.** Descriptive statistics of rectal (RT), vaginal (VT) and reticular (RET, single value (RET-SIN), 2-h-mean value (RET-MEAN), 2-h-median value (RET-MED)) temperatures and barn temperature (T) and THI averaged and separated for the two study periods in June and October

Item	N	Mean	SD	Min	Max
<b>Total</b>					
RT (°C)	293	38.1	0.6	36.5	40.2
VT (°C)	293	38.2	0.4	37.3	40.4
RET-SIN (°C)	293	38.7	0.9	33.9	41.2
RET-MEAN (°C)	293	38.5	0.7	36.9	41.5
RET-MED (°C)	293	38.7	0.5	37.3	41.6
T (°C)	293	19.1	5.3	11.0	32.9
THI	293	65.2	8.0	52.0	83.5
<b>June</b>					
RT (°C)	158	38.3	0.6	36.5	40.2
VT (°C)	158	38.3	0.5	37.4	40.4
RET-SIN (°C)	158	38.9	1.0	33.9	41.2
RET-MEAN (°C)	158	38.6	0.8	36.9	41.5
RET-MED (°C)	158	38.9	0.6	37.3	41.6
T (°C)	158	22.9	4.3	16.5	32.9
THI	158	71.1	6.0	61.5	83.5
<b>October</b>					
RT (°C)	135	37.9	0.4	36.7	38.9
VT (°C)	135	38.1	0.3	37.3	38.7
RET-SIN (°C)	135	38.5	0.6	35.6	39.6
RET-MEAN (°C)	135	38.4	0.5	37.0	39.6
RET-MED (°C)	135	38.6	0.4	37.3	39.6
T (°C)	135	14.6	1.7	11.0	17.4
THI	135	58.3	2.9	52.0	62.7

statistical model:

$$Y_{ijklmn} = \mu + \text{Meth}_i + \text{Mo}_j + (\text{Meth} \times \text{Mo})_{ij} + T_k \\ + D_l(\text{Mo})_j + \text{Cow}_m + e_{ijklmn},$$

where  $y_{ijklmn}$  is the  $n$ th measurement of RT, VT, RET-SIN, RET-MEAN or RET-MED for method  $i$  in month  $j$  at time of the day  $k$  at day  $l$  in month  $j$  of cow  $m$ ,  $\mu$  is the intercept;  $\text{Meth}_i$ ,  $\text{Mo}_j$ ,  $T_k$  and  $D_l(\text{Mo})_j$ ,  $(\text{Meth} \times \text{Mo})_{ij}$  are the fixed effects for method  $i$  ( $i = \text{RT, VT, RET-SIN, RET-MEAN, RET-MED}$ ), month  $j$  (June, October), time of day  $k$  ( $1 = 7.30 \text{ a.m., } 12.00 \text{ p.m., } 17.30 \text{ p.m.}$ ), day  $l$  in month  $j$  ( $l = 1-5$ ), interaction between method  $i$  and month  $j$ , and  $\text{cow}_m$  is the repeated effect of cow  $m$  ( $m = 1-12$ ) and  $e_{ijklmn}$  is residual error.

## Results and discussion

### Descriptive statistics

The average barn T and THI were  $23.0 \pm 4.3$  °C and  $71.1 \pm 6.0$  in June and  $14.6 \pm 1$  °C and  $58.3 \pm 2.9$  in October. T ranged between 11.0 and 32.9 °C, THI between 52.0 and 83.5 (Table 1). The ambient conditions did vary in the range, which is normal for a region in middle Europe. Figure 1 shows the course of the hourly minimum (T Min) and maximum (T Max) barn temperatures during the 5-d study periods in June and October.

Overall means ( $\pm$ SD) of body temperatures for RT, VT, RET-SIN, RET-MEAN and RET-MED were  $38.1 \pm 0.6$ ,  $38.2 \pm 0.4$ ,  $38.7 \pm 0.9$ ,  $38.5 \pm 0.7$  and  $38.7 \pm 0.5$  °C,

respectively (Table 1). The values are in the range of earlier studies for RT (38.8 °C) (Bewley et al. 2008a), RET (38.3–38.9 °C) (AlZahal et al. 2011) and VT (38.4–38.8 °C) (Lee et al. 2014). Lowest temperature values with a minimum of 33.9 °C occurred for a RET-SIN measurement, while the highest temperature was recorded for a RET-MED value over a 2 h period (41.6 °C). The low RET-SIN values were probably caused by the intake of water and/or food around the time of measurement. Several authors showed that water consumption reduces ruminal temperature for a period of time proportional to the water temperature and amount of water ingested. It takes between 20 min and 2 h for ruminal temperature to recover (e.g. Yamada et al. 2001). This limits the use of single RET measurements, while on the other hand continuous records lead to stable values.

Compared to October, values were 0.2 to 0.3 °C higher in June (Table 1), which has to be kept in mind when BT is interpreted in relation to the physiological status of an animal. Again, this favors a high frequency of measurements. An increase with increasing ambient values was shown (e.g. Suthar et al. 2012). The frequencies of the recorded body temperatures are shown in histograms (Fig. 2).

Lowest variation was observed for VT and RET-MED. As previously presented by Bewley et al. (2008a) the most frequently observed temperatures ranged between 38 and 40 °C with a low variation between the measurements. However, it has to be kept in mind that these authors

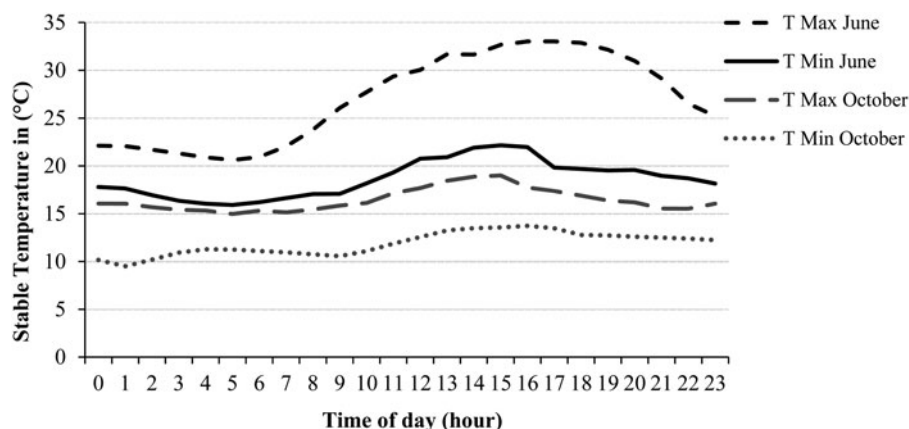


Fig. 1. Course of the hourly minimum (T Min) and maximum (T Max) barn T during the 5-d study periods in June and October.

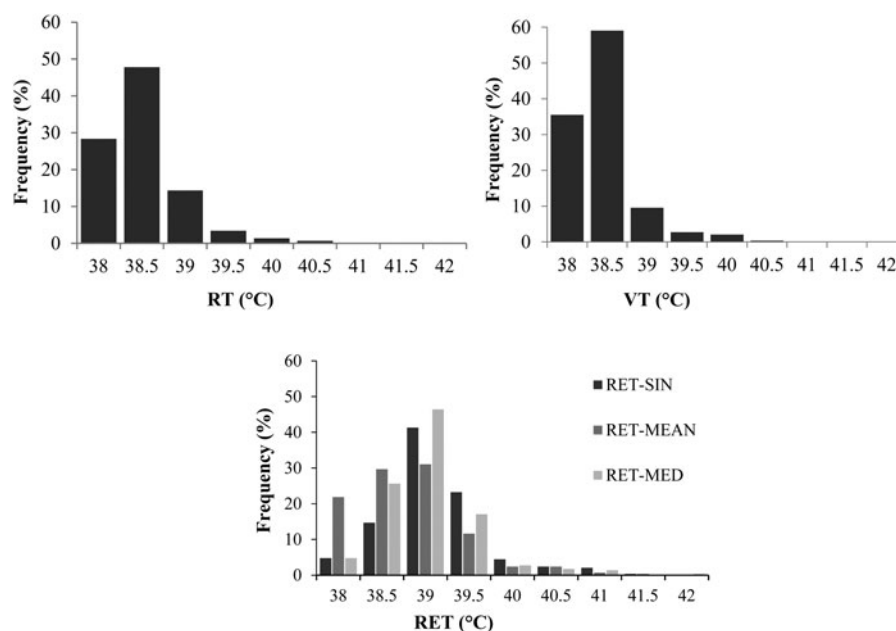


Fig. 2. Histograms (%) of rectal (RT) (left above), vaginal (VT) (right above), reticular (RET) (left below) reticular single value (RET-SIN), reticular 2-h-mean value (RET-MEAN), reticular 2-h-median value (RET-MED) (right below) temperatures recorded during the trial.

excluded low temperatures caused by water intake from their statistical analysis. Low values, occurred for RET-SIN values in the present study, may be explained by water intake around times of bolus measurements (Bewley et al. 2008b). Though independent of water ingestions, RT and VT values were similarly distributed as RET records, but on a lower level. The highest number of records was found around 39 °C, followed by 38 and 40 °C. Bewley et al. (2008a) also found the highest number of records for RT at 39.0 °C. Beside the water intake, the metabolic heat production influences the RET values, which leads to increasing temperatures when compared to VT and RT (Bewley et al. 2008a).

#### Correlations between measuring methods

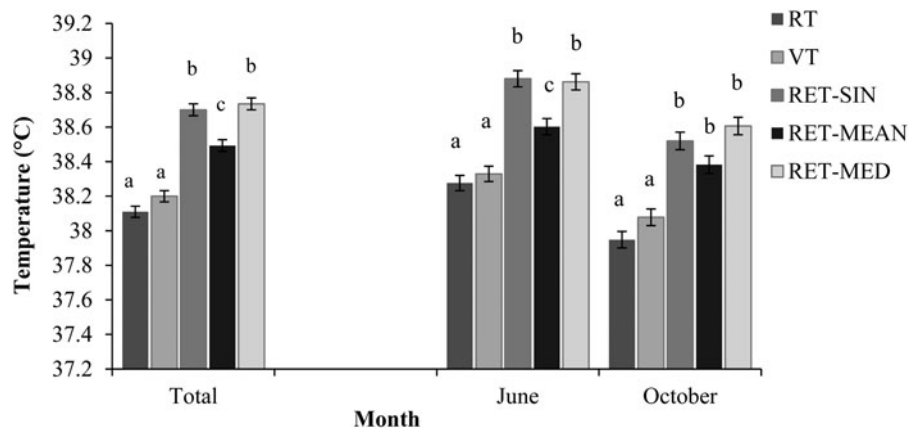
The overall Pearson correlation coefficient between RT and VT estimated for 293 paired observations was 0.75 ( $P < 0.001$ ). The correlation values between VT and RET-SIN, RET-MEAN and RET-MED were slightly stronger when compared with RT ( $P < 0.001$ ) (Table 2).

However, values were much lower when compared with Sievers et al. (2004). They showed highly significant correlations between intraruminal and rectal temperatures. The difference may be caused by the fact that their estimation was only based on 36 observations. The values of the present

**Table 2.** Correlation coefficients for rectal (RT), vaginal (VT) and reticular (RET, single value (RET-SIN), 2-h-mean value (RET-MEAN), 2-h-median value (RET-MED)) temperatures separated for the two study periods in June and October, differentiated by the time of day

Item	N	RT VT	RET-SIN		RET-MEAN		RET-MED	
			RT	VT	RT	VT	RT	VT
Total	293	0.75***	0.40***	0.48***	0.43***	0.46***	0.48***	0.53***
June								
Total	158	0.81***	0.41***	0.52***	0.48***	0.54***	0.53***	0.60***
Morning	52	0.77***	0.29*	0.42**	0.36**	0.63***	0.42**	0.69***
Midday	53	0.77***	0.57***	0.67***	0.52***	0.53***	0.62***	0.62***
Evening	53	0.86***	0.44**	0.56***	0.66***	0.73***	0.63***	0.71***
October								
Total	135	0.52***	0.24**	0.24**	0.23**	0.12	0.19*	0.09
Morning	45	0.46**	0.36*	0.57***	0.37*	0.46**	0.23	0.33*
Midday	45	0.60***	0.40**	0.38**	0.48***	0.54***	0.43**	0.42**
Evening	45	0.31*	0.09	0.16	0.23	0.19	0.29	0.18

\*\*\* =  $P < 0.001$ , \*\* =  $P < 0.01$ , \* =  $P < 0.05$

**Fig. 3.** Least Square Means of rectal (RT), vaginal (VT) and reticular (RET, single value (RET-SIN), 2-h-mean value (RET-MEAN), 2-h-median value (RET-MED)) temperatures separated by month and time of day. Different letters show significant differences ( $P < 0.05$ ) between methods within months.

study are rather similar to those found by Bewley et al. (2008a) and Burns et al. (2002). They estimated correlations between RT and RET of 0.65 and 0.50, respectively. With regard to the correlations between RT, VT and RET, it is important to keep in mind that RET measurements can be influenced by water and feed intake, while RT and VT measurements are limited by human error in locating the probe, duration of measurement, presence of air and faeces in case of RT (Bewley et al. 2008a) and the handling of an animal before and during measurement procedure. For example Lee et al. (2014) have shown that short-finger anchors led to different VT when compared with long-finger anchors. Additionally diurnal and seasonal variation may also have an impact on BT (Piccione & Refinetti, 2003). This may also have influenced the correlation values estimated for BT measurements and month. In this study correlation values were higher in June than in October. This could be explained by the fact that, with an increase of ambient temperature, body temperature

increases to a higher level with smaller differences between the RT, VT and RET values. The correlations within the study periods were highest for the evenings in June and for mornings and middays in October. In contrast to October, observations of RET-MED temperatures in June were more closely correlated to RT and VT than measurements of RET-SIN and RET-MEAN. Bewley et al. (2008a) described stronger correlations during spring and fall than for summer and winter periods. These results may again simply indicate that the relationships between the methods are different in relation to environmental conditions. This observation corresponds also to the concept of increasing correlations with deviating BT (Bewley et al. 2008a).

#### Effects of month and time of day on body temperature measurements

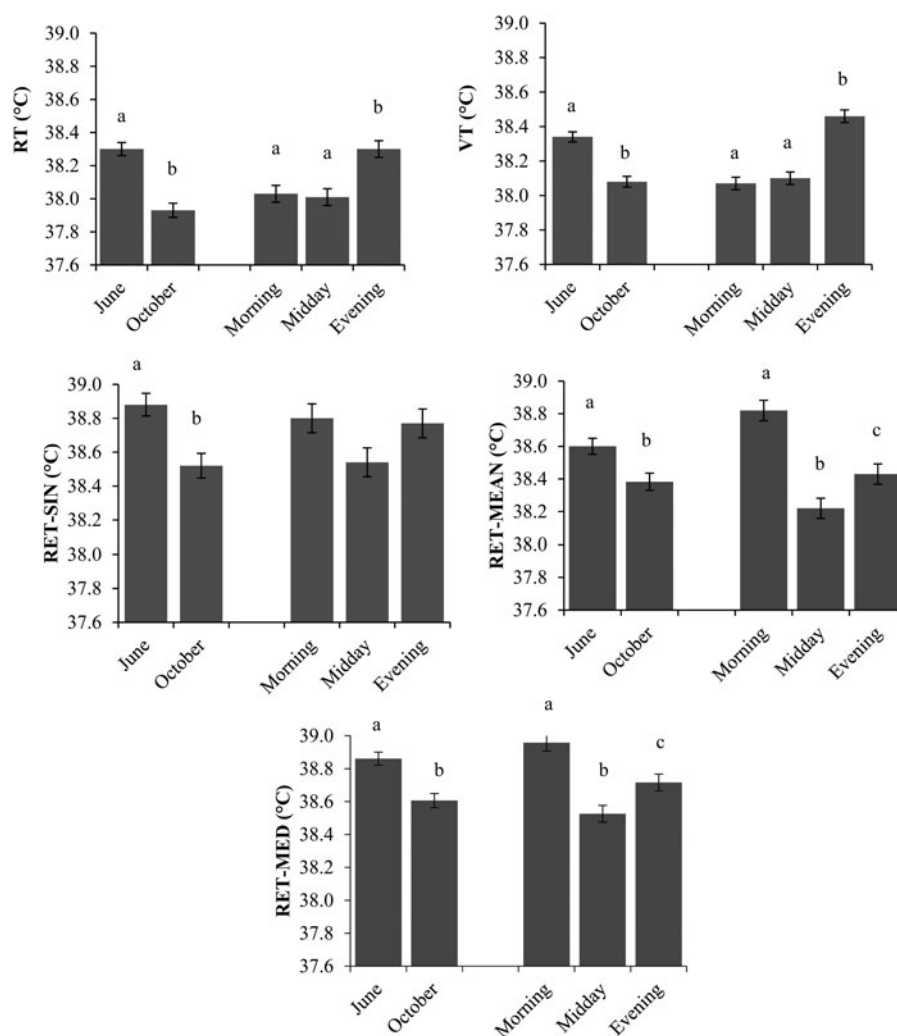
Over both study periods, RT did not differ from VT ( $P > 0.05$ ; Fig. 3). Alike, RET-SIN and RET-MEAN were not significantly



**Table 3.** Correlation coefficients between rectal (RT), vaginal (VT) and reticular (RET, single value (RET-SIN), 2-h-mean value (RET-MEAN), 2-h-median value (RET-MED)) and barn temperature (T) separated for the two study periods in June and October

Period	N	RT-T	VT-T	RET-SIN-T	RET-MEAN-T	RET-MED-T
Total	293	0.41***	0.48***	0.24***	0.08	0.16**
June	158	0.26***	0.41***	0.16*	-0.06	-0.02
October	135	0.31***	0.40***	-0.01	-0.14	-0.08

\*\*\* =  $P < 0.001$ , \*\* =  $P < 0.01$ , \* =  $P < 0.05$

**Fig. 4.** Least Square Means of rectal (RT), vaginal (VT) and reticular (RET, single value (RET-SIN), 2-h-mean value (RET-MEAN), 2-h-median value (RET-MED)) temperatures separated by month and time of day. Different letters show significant differences ( $P < 0.05$ ) between the months and time of day.

different ( $P > 0.05$ ) from each other, while RET-MED was significantly lower ( $P < 0.001$ ). When comparing the two study periods, differences between RT or VT and the three RET-temperature values were higher.

The slight difference could have been caused by the necessity of restraining animals to collect RT and VT temperatures. This may have caused stress that finally leads to altered temperatures. Therefore, intraruminal measuring

methods are probably more reliable because collecting temperatures cause less human intervention, which may provide a more accurate measure of temperature in dairy cattle (Hahn et al. 1990; Prendiville et al. 2002). Several previous studies (e.g. Sievers et al. 2004; Bewley et al. 2008a) already favoured an intraruminal measuring system towards manual methods. Meanwhile, such rumen or reticular temperatures have been established as effective measures of

core BT (Hicks et al. 2001; Prendiville et al. 2002). Nevertheless, it has to be kept in mind that ruminal or reticular temperatures are generally approximately 0.5 °C greater than core BT resulting from the heat production of ruminal microorganisms (Bitman et al. 1984; Hicks et al. 2001; Prendiville et al. 2002). Another study even reported ruminal temperatures to be 0.71 °C (shade) and 0.49 °C (no shade) greater than intraperitoneal temperatures, although no differences were observed when ambient temperatures increased above 25 °C (McVicker et al. 2001).

#### Effects of barn climate and measurement time

Correlation coefficients between barn T, RT and VT were greater when compared with correlations of RET-measurements (Table 3). This may also be explained by the earlier mentioned assumption, that RET measurements can also be influenced by water and feed intake. This will lead to lower correlations with respect to environmental temperatures. Lowest correlations between T and RT values occurred in October. Over both study periods, values ranged between  $r = 0.08$  ( $P > 0.05$ ) and  $r = 0.24$  ( $P < 0.001$ ), whereas VT was strongest correlated with barn T ( $r = 0.48$ ;  $P < 0.001$ ). The increase of the values with the change of external factors (e.g. increasing ambient temperature) was described earlier (Bewley et al. 2008a).

The effect of several measurement times of the day for morning, midday and evening on BT observation methods is given in Fig. 4. Highest RT and VT were observed in the evening (38.3 and 38.5 °C), when compared with morning (38.0 and 38.1 °C) or midday (38.0 and 38.1 °C) measurements ( $P < 0.001$ ), respectively. In contrast, all RET-temperatures were highest in the morning (38.8–38.9 °C), followed by evening (38.4–38.8 °C) and midday (38.2–38.5 °C) ( $P < 0.001$ ). In general, highest temperature values occurred for RET-MED, while lowest temperatures were recorded for RT. Diurnal changes in BT were described earlier. They are related to animals activities like water intake, feeding and milking. The changes are also related to the amounts of feed and water intake as well as milk yield (Igono et al. 1985; Piccione & Refinetti, 2003). These factors could not be taken into consideration in this study.

#### Conclusions

The intention of this study was to estimate the relationship between different methods of BT measurements in lactating cows under various climatic conditions. Mean values of RT were not significantly different, while variation differed. The RT was strongly correlated with VT, demonstrating that both provided a similar estimate of body temperature in dairy cattle. The relationship between RT, VT and RET was lower. The dynamics of the reticulorumen environment, particularly related to feed and water intake, likely influenced this relationship and might limit the suitability of RET in an automated temperature-monitoring system. Besides, data recorded with the methods studied here partly differ in their

potential use. While the precision of VT and RT measurements is affected by the competency of the operator using them and other circumstances (e.g. depth and duration of probe insertion and the presence of air or faeces in the rectum (RT) (Lee et al. 2014), the use of a rumen bolus is relatively independent from these factors. Beyond, the use of boluses is less labour intensive, time consuming and cannot result in injuries (Bewley et al. 2008a). However, the impact of water and feed intake is a major limiting factor of measurements from the reticulorumen. The frequently measured values are stable over time and allow a much better use within health monitoring programs, because diurnal variation, and particularly the impact that feed and water intake and milking will have, can be excluded. However, it is important to remember that no method can be considered to be the ideal benchmark for core BT.

It can be concluded, that all methods were subjected to more measurement error and variation than anticipated. Nevertheless, these results are encouraging for the use of newly available technologies for monitoring RET.

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