Report of the Short Term Scientific Mission of Dr. Diana Sorg

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1. Purpose of the STSM

The purpose of this STSM was to compare methane concentrations in the air in a respiration chamber taken with two Laser Methane Detectors (LMD) of the same model to the methane concentrations determined by the gas analyser of the chamber while a dairy cow was in the chamber. During this experiment, the two LMDs were also compared to each other. Additionally, the influence of the cow's activity on her methane emission was analysed.

2. Work carried out

In the tie-stall, several methane profiles at different activities of the two cows entering the chambers were collected over two days prior to the chamber measurements. The activities were eating, standing idle, standing ruminating, lying idle, lying ruminating, drinking and sleeping. In total, 36 profiles of an average length of 110 sec were recorded.

Before the start of the chamber measurements, the experimental setup was tested in empty chambers. For that, the positioning of the LMDs was optimized (figure 1). The LMDs were placed next to each other in a small plastic box which was set on top of two large plastic boxes. The laser beam was directed to the centre of the six radially positioned ventilation pipes. With this setup, the laser beam was directed away from the cow but into the area where the gas for the chamber measurements was sampled. Equally important, it was reflected on an even surface. A piece of white paper was attached to the surface in order to enhance reflection.



Fig. 1: Position of the LMD in the respiration chamber. 1: LMDs in a small open plastic box 2: laser beam of the LMD 3: outgoing ventilation pipes (6 radially positioned pipes) 4: air flow 5: approximate position of the cow 6: plastic boxes (view from front towards the backdoors of the chamber, not to scale).

It was not possible to take a dynamic profile of the cow's breath and eructation by directly pointing to her mouth. The main reason for that was the lack of space due to the construction of the feeding trough in front of the cow's head, so that the mouth was not always visible. Also, presumably the cow would have been disturbed by the prolonged presence of an unfamiliar observer over the short distance (about 50 cm from her head) to an extent that would have strongly influenced her feeding behaviour and with it the results of the main experiment carried out by the Swiss scientists. The implication of this study is, however, that the technology of the LMD itself is sufficiently precise and reactive, so that it is able to reliably measure dynamic methane concentrations in ranges that are typical for dairy cows, especially when keeping in mind that it was originally designed to discriminate between very high and low methane values only, not to quantify the exact concentration.

On the first day of the chamber measurements, both LMDs were placed in chamber 1. On the second day, both LMDs were placed in chamber 2. Cow 1 was in chamber 1 during the 2 days and cow 2 was in chamber 2 during the 2 days. A continuous measurement was taken from 7.45 h until 17.45 h on the first day and from 9.00 h until 17.30 h on the second day. The measurement was only paused during lunch, during milking in the afternoon and when the batteries of the LMDs were changed (twice daily). The data were sent to the smartphones outside the chamber via Bluetooth connection.

3. Main results

Cow profiles



An example of a cow profile is given in figure 2. The activity of the cow was lying ruminating.

Fig. 2: Methane profile of cow 2 lying ruminating in the tie-stall, measured with LMD1. The high peaks are presumably eructation peaks.

Agreement of the two LMDs

We found a good agreement between the two LMDs: the Pearson correlation coefficient between them was 0.993. A linear regression for the model LMD2=LMD1 had an R^2 of 0.996 (Figure 3).



Fig. 3: Regression of the static measurements over the two days and two chambers of LMD1 and LMD2

Figure 4 shows the time course of the methane values from LMD1 and LMD2 for chamber 2 exemplarily.



Fig. 4: Methane measured with the two LMDs in chamber 2.

A linear mixed model for the LMD methane values revealed a significant effect of the LMD and of the combined Chamber-cow-day effect (CCD) (table 1). Their interaction was not significant.

Fixed effect	Р	LSMEANS (ppm-m) ± SEM		
		LMD1	LMD2	
LMD	<0.0001	960 ± 0.75	933 ± 0.76	
		chamber 1-cow 1-day 1	chamber 2-cow 2-day 2	
CCD	<0.0001	951 ± 0.77	942 ± 0.74	

Table 1: P-Value and LSMEANS of LMD methane for the fixed effects in the LMD comparison

Agreement with respiration chamber

The respiration chamber measured the average gas concentration in 700 l of exhausted air per minute and recorded the average volume of methane in litres per minute. However, the LMD recorded the cumulative concentration in ppm-m twice per second and in a gas column of 1.55 m length. So we had to convert (a) from volume to concentration in the case of the chamber values and (b) from twice per second to an average per minute and from a cumulative to an absolute concentration for the LMD values in order to compare them: from the chamber values the average concentration per minute was calculated by dividing the recorded methane volume in litres per minute by the air flow per minute (700 litres) to obtain the vol-% of methane. This in turn was multiplied by 10,000 to obtain ppm, the unit recorded by the LMD. The LMD data (2 measurements per second) were averaged for each minute. Then the absolute mean concentration in ppm was calculated by dividing the recorded LMD values in ppm-m by the length of the laser path (1.55 m).

We found a good agreement of the LMD methane with the chamber methane (table 2). Figure 5 shows a scatter plot of the values of chamber 2 exemplarily. Figure 6 shows the time course of LMD and chamber methane in chamber 2 exemplarily.

	у	Chamber 1		Chamber 2	
	х	LMD1	LMD2	LMD1	LMD2
Pearson correlation coefficient		0.93 ***	0.94 ***	0.95 ***	0.95 ***
Linear Regression y=x					
R ²		0.86	0.89	0.91	0.91
Intercept		46.5***	86.1***	-35.8***	13.4***
Slope		0.93***	0.89***	1.12***	1.07***

Table 2: Agreement of the two LMDs with the two respiration chambers



Fig. 5: Scatter plot of chamber methane against LMD methane in chamber 2.



Fig. 6: Time course of the LMD and chamber methane in chamber 2.

Methane and cow activity

Cow activity was visually recorded for each minute. The average methane of the two LMDs and of each minute was used for the analysis. In a linear mixed model with the combined Chamber-cow-day effect (CCD) and the Activity as fixed effects, CCD was not significant for the LMD methane, but it was for the chamber methane (p<0.0001) where the interaction of CCD with Activity was also significant. Cow activity had a significant influence (p<0.0001) on the methane concentrations from both chamber and LMD. Tables 3 and 4 show the LSMEANS for the average LMD and chamber methane in a linear mixed model, respectively. They are sorted from highest to lowest in order to be able to compare their ranking between the two methods. Drinking was identified by both methods as the activity with the highest methane concentration. Sleeping and moving had the lowest concentrations and were not significantly different from each other in both methods. Values with different superscripts differ significantly (p<0.05, Bonferroni corrected for multiple comparisons). Figure 6 shows the average LMD methane and cow activity in chamber 2 as an example. Figure 7 shows the chamber methane and cow activity in chamber 2 as an example.

Activity	Total length (min)	LSMEANS (ppm) ¹	SEM
Drinking	21	645 ^{bcd}	25
Lying ruminating	325	643 ^d	6
Lying idle	196	612 ^c	8
Standing	145	611 ^{bcd}	9
Feeding	183	574 ^{ab}	8
Sleeping	22	570 ^{abcd}	24
Moving	62	540 ^a	22

Table 3: LSMEANS of the LMD methane at different activities

¹Values with different superscripts in one column differ (P<0.05)

Activity	Total length (min)	LSMEANS (ppm) ¹	SEM
Drinking	21	689 ^{ab}	21
Feeding	183	681ª	7
Lying ruminating	325	665°	5
Standing	145	664 ^{ab}	9
Lying idle	196	634 ^{bd}	6
Moving	62	585 ^c	13
Sleeping	22	581 ^{cd}	24
CCD			
Chamber 1-cow 1-day 1	463	616 ^a	6
Chamber 2-cow 2-day 2	491	670 ^b	9

Table 4: LSMEANS of the chamber methane at different activities

¹Values with different superscripts in one group and column differ (P<0.05)



Fig. 7: Average LMD methane in chamber 2, coloured by cow activity.



Fig. 8: Chamber methane in chamber 2, coloured by cow activity.

4. Conclusion

Under environmental conditions in a barn it is very difficult to compare two LMDs. The two devices must not be placed too close to each other (> 10 cm) or otherwise their laser beams will be interfering. That is why slight micrometeorological changes in wind speed, direction etc. can lead to a great influence on recorded methane values when comparing two LMDs. In the chamber, these influences were minimized and we had the chance to show a good agreement between the two LMDs, telling us that at least their technology is precise and reliable. Later on in the field, both LMDs will be used in order to measure more cows at each visit to a herd. We will still include the number of the LMD as a fixed effect in our statistical model to correct for their difference, but with these data we have a good justification for using them both equally.

The good agreement with the chamber methane values shows that the technology of the LMD is able to reliably quantify exact dynamic methane concentrations in the air and especially in the range of emissions from a dairy cow (in our experience below 1500 ppm). Methane, when mixed with air in a concentration of 5 - 15 %, is explosive. Therefore, the LMD was developed to detect such dangerous concentrations in industrial settings. In ppm, this is 50.000 to 150.000 – several magnitudes above the range of our data. Still, the measurement was surprisingly precise when compared to the respiration chamber. This finding tells us that with the LMD it should be possible to discriminate between high and low methane emitting animals, which is the ultimate goal of our project.

The third result of this study was the confirmation of the influence of cow activity on methane emission. We are still developing the protocol for the measurements on commercial farms. For this purpose, this study has confirmed the strict need to at least record (if not standardize) the cow's activity when taking a methane profile with the LMD, and to compare only profiles that were taken during the same activity in the statistical analysis.

Building on the work of Chagunda et al., this is a step forward in further verifying the applicability of the LMD to measure methane emissions from dairy cows. With our work we aim to establish a working protocol that can be adopted by other scientists in METHAGENE and interested users without the need for extensive validation. By publishing our results we will make this knowledge accessible to everyone in METHAGENE and in the scientific community.

5. Future collaboration with the host institution

LMD measurements will likely be incorporated in future research projects at ETH Zurich, serving as an additional method to already established methane measurements. End of March 2015 a corresponding doctoral project proposal was submitted to the Swiss COST office. In this proposal, a collaboration with Halle University is explicitly foreseen (Letter of support provided by Prof. Hermann Swalve).

6. Foreseen publications

The data from this comparison are foreseen to be used in a publication about the validity of the Laser Methane Detector in a scientific peer-reviewed journal. However, more data will be collected in another respiration chamber in order to increase the size of the data set and to provide a greater understanding of the agreement of the LMD with the gold standard for methane measurements of dairy cows.

7. Confirmation of the host institution of the successful execution of the STSM

See the attached letter from the host institution.

8. Comments

These are only preliminary and exemplary results. A more thorough analysis will follow.