

# First Results in the Use of Milk Mid-infrared Spectra in the Detection of Lameness in Austrian Dairy Cows

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## Summary

Lameness in dairy cows is a concern for both producers and consumers. Milk mid-infrared (MIR) analysis could be an extra tool in the detection of lameness problems for farmers. The aim of this study was to test the feasibility of detecting lameness problems using MIR spectra from milk through the development of predictive models. The data for this research was provided by RINDERZUCHT AUSTRIA (2017), from their “Efficient Cow” project and were recorded between July 2014 and December 2014. The data sets used were the complete data set of 9811 records and subsets according to lactation stage, parity, breed and hoof disease. Two types of pre-processing were tried: first derivative followed by a Standard Normal Variate (SNV) transformation or second derivative followed by a SNV transformation. The first and second derivatives do not give the same results which highlights the importance of pre-processing during model development. The best results were obtained for the Heel horn erosion subset. However, the specific nature of the used data requires the addition of more data coming from varied animals and farms and validation steps before using this technology on a larger scale.

## Key words

MIR spectra, milk composition, lameness, PLS-DA

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Received: May 16, 2017 | Accepted: September 13, 2017

## ACKNOWLEDGEMENTS

I am grateful to EMR (2017) for giving me the opportunity to do this research. I would also like to thank RINDERZUCHT AUSTRIA (ZAR) for providing the data.

## Introduction

Lameness in dairy cows is a concern for both producers and consumers. Indeed, it is the third most costly health problem on dairy farms (Enting et al., 1997) with a mean prevalence of 36.9% reaching up to 79.2 % on certain farms (e.g. Barker et al. 2010). It is also a big animal welfare concern and generates the need to use antibiotics. Milk mid-infrared (MIR) analysis could be an extra tool in the detection of lameness problems for farmers without adding to their workload. Indeed, the MIR analysis is already performed in routine for most of the cows to provide at least fat and protein content in the frame of the milk recording. The idea is that lameness associated physiological, e.g. inflammation, or behavioural, e.g. feeding habits, changes have a repercussion on the milk composition and that this change can be detected through the use of MIR spectra. This could be especially useful for early lameness detection, i.e. animals are already affected before showing clear clinical signs and are potentially more easily overlooked, or for large herds where it is harder to keep track of all animals with the same level of detail on a regular basis. The objective of this study was to test the feasibility of detecting lameness problems using MIR spectra from milk through development of predictive models and to evaluate the effect of different pre-treatment methods on the sensitivity and specificity of the prediction models.

## Data collection, sub setting and calibration

The data for this research was provided by RINDERZUCHT AUSTRIA (2017), from their “Efficient Cow” project. This project, launched in 2012, collected data from 167 Austrian farms and about 5000 dairy cows. Among the data collected, there was information on animal health such as lameness and claw care. Lameness was assessed by trained technicians, using Visual Locomotion Scoring which gives a cow a score from 1 to 5. Diseases or problems of the foot or hoof were recorded by hoof trimmers. The Visual Locomotion Scoring according to Sprecher et al. (1997) gave the animals a score of 1 (normal gait), 2 (uneven or stiff gait), 3 (lightly lame), 4 (lame) or 5 (heavily lame). For the present analysis, a new variable was created out of these scores, classifying animals into non lame (1 and 2) and lame (3, 4 and 5). Next to this, milk samples gathered for routine milk recording were analyzed by MIR spectrometry using 1 FOSS FT+ and 2 FOSS FT6000 instruments. The MIR instruments were standardized using the EMR/CRA-W standardization process (Grelet et al., 2015). Only records between calving and 365 days in milk, and with a maximum of 7 days between the lameness scoring and the milk sampling for MIR analysis were used in this study. They were recorded between July 2014, start of the standardization of the spectrometers in Austria, and December 2014 on 3973 cows from 121 farms, giving a total of 9811 records. Cows were recorded on average 2.5 times. The cows consisted of 2670 Fleckvieh, 666 Holstein and 637 Brown Swiss cows. MIR spectra consist of 1060 absorbance values at different wavenumbers, ranging from 925.66  $\text{cm}^{-1}$  to 5010.16  $\text{cm}^{-1}$ . The absorbance values give information about the composition of the milk as each combination of atoms absorbs light at a precise wavenumber. Yet, not all those 1060 data points are used in the making of the prediction model. Indeed, some parts of the spectra are ‘noisy’ because of strong

water absorption. Therefore, specific parts, that contain the most information whilst also reducing the noise to a minimum, are selected. These spectral areas are: 968.1 to 1577.5  $\text{cm}^{-1}$ , 1731.8 to 1762.6  $\text{cm}^{-1}$ , 1781.9 to 1808.9  $\text{cm}^{-1}$  and 2831.0 to 2966.0  $\text{cm}^{-1}$  (Grelet, 2016). It is also important to note that lameness prediction is indirect as it is not directly measured in milk but based on modifications in milk composition; therefore, the selected parts of the spectrum are used as a whole to predict lameness directly. Lameness and MIR data were merged and inadequate records, containing only lameness or only MIR information or with more than seven days between Locomotion Scoring and milk sampling, were deleted using SAS (SAS Institute Inc., 2017). Different data sets were created. The full set (9811 records) was used as a reference. Multiple subsets, either linked to a specific period in the lactation, to specific diseases, to breed or to parity, were created to obtain more homogeneous data sets. This was done to see if homogeneity of the dependent variable definition and the associated spectra had an influence on the precision of the model. Subsets with different groupings of animals were created with only heifers (parity=1), young (parity=1, 2) and old (parity>2) cows. This was done as there is not a consensus on the influence of parity on milk composition in the literature. Some studies report this influence (e.g., Yadav et al., 2013; Yang et al., 2013) others not (e.g., Gurmessa et al. 2012). Subsets were also created for lactation stages; a factor that is known to influence milk composition (e.g., Bastin 2011). For this, the complete lactation was split into first and last half and first and last third. In order to allow differences due to health and lameness having a better chance of standing out, we also took breed into account to smooth out breed related differences. Indeed, many studies (e.g., Heinrichs et al., 1997) reported the influence of breed on milk composition. Heel horn erosion (HHE) is the dissolution and decay of the horn on the bulbs of the heel. White line disease (WL) refers to a gap between the sole and the wall often filled with feces or decayed horn masses, which can lead to an abscess if the leather skin is affected (Egger-Danner et al. 2015). For both these diseases, specific sets were created by selecting all records with the disease, adding all the records of healthy animals coming from the same farms and only keeping records where the hoof trimmer data had been collected within three weeks of the MIR analysis. However, not all animals affected by HHE or WL were also lame so two extra files were created where only diseased lame and healthy non lame animals were kept. A third of the records of every data set were randomly selected for validation. The other two thirds of each subset were selected and used for calibration. For this separation of calibration and validation, the data sets were separated by record, not by animal, even though some animals have multiple records, because the lameness status of an animal can vary over time and the animal may find itself in different living conditions, e.g. pasture in summer and stall in winter. This makes every record unique. In the case of heel horn erosion (HHE) and wall defect (WL), some MIR spectra found themselves in both data sets as the cow had one of her hoofs affected by heel horn erosion, while another had a wall defect problem. In total, from the 9811 records, 1843 had HHE and 1068 suffered from WL disease. Pre-treatment of the spectra consisted of a first or second derivative with widths of 5, using the Savitzky-Golay method, to enhance

resolution and eliminate additive baseline drift between samples. This was followed by a transformation to Standard Normal Variates (SNV) in order to standardize each spectrum into having a mean of 0 and standard deviation of 1 to correct for scattering (Fearn, 2017; Huang et al., 2010). Venetian blinds were chosen as cross-validation, which means 10% of the calibration set was randomly selected 10 times. Prediction models were done with Partial Least Squares Discriminant Analysis (PLS-DA), using the software PLS-Toolbox, by Eigenvector Research Inc., on Matlab (The MathWorks Inc., 2000). The PLS-DA is a variant of PLS regression used when the dependent variable is categorical, in this case lame vs. non lame (Fernández Pierna, 2017).

## Results and discussion

The results for different data sets are shown in Table 1. The sensitivity, i.e. lame animals predicted as lame by the model, and the specificity, i.e. non lame animals predicted as non lame, is presented for each data set for calibration and validation (Penn State Eberly College of Science, 2017). The number of latent variables used in the prediction models was chosen based on the break of slope of the Root-Mean-Square Error of Cross-Validation (RMSEcv) plot of the data set 'All'. RMSEcv is a measure of fit and the smaller this value, the better the prediction model. The break of slope is the point where adding another latent variable

does not significantly reduce the RMSEcv anymore. Because all other data sets were then compared to the former, the same number of 11 latent variables was chosen for every set. For each data set, the first row is the results obtained for using the first derivative, the second row for the second derivative.

As shown in Table 1, changing the pre-processing from a first derivative and SNV to a second derivative and SNV produces varying results, depending on the subsets. Based on these results that reflect results found by Soyeurt et al. (2011), when working on models with MIR spectra, it is important to test pre-treatment possibilities to make the most advised choice. The result for the complete number of records, 'All', is low to average with sensitivities and specificities between 53.64 and 64.51 for the first derivative and the second derivative.

Results showed that sensitivity and specificity strongly increased for smaller subsets with more precise definition of the target to be predicted. In this study we choose to reduce the variation due to known factors by making data sets smaller and therefore also reducing the variation in the spectra. This allowed obtaining better results. However, selecting very strictly on certain factors can also become a problem if the data sets become too small for good calibration as illustrated in the second to last set. An alternative way of dealing with this issue could be by

**Table 1.** Number of records (N), number of lame records (lame), sensitivity and specificity for calibration, validation and for first derivative (1st der) or second derivative (2nd der) for different data sets. \*HHE = Heelhorn erosion, \*\*WL = white line defect.

Subset	N	Lame	Pre-treatment	Calibration		Validation	
				Sensitivity (%)	Specificity (%)	Sensitivity (%)	Specificity (%)
All	9811	795	1st der	63.30	62.88	59.77	62.45
			2nd der	61.05	65.42	53.64	64.51
First half of lactation	5509	490	1st der	59.94	68.28	53.37	65.80
			2nd der	59.29	69.24	52.24	67.37
Last half of lactation	4302	305	1st der	67.98	64.13	59.80	63.59
			2nd der	61.08	67.66	55.88	66.97
First third of lactation	3806	348	1st der	70.29	54.61	66.97	57.67
			2nd der	68.20	60.05	62.39	60.60
Last third of lactation	2479	176	1st der	64.87	68.74	44.62	66.36
			2nd der	56.76	72.50	32.31	68.07
Fleckvieh	6828	578	1st der	71.13	62.03	61.58	58.87
			2nd der	67.78	65.56	54.74	61.03
Holstein	1560	121	1st der	67.53	70.41	43.18	71.01
			2nd der	54.55	77.47	43.18	78.57
Brown Swiss	1423	96	1st der	68.12	70.00	66.67	63.31
			2nd der	69.57	72.73	59.26	67.79
Heifer (parity = 1)	2792	96	1st der	73.44	67.00	56.25	64.74
			2nd der	64.06	71.56	46.88	69.63
Young (parity = 1,2)	4855	195	1st der	71.43	59.34	48.89	57.78
			2nd der	63.91	63.21	46.77	61.63
Old (parity > 2)	4956	600	1st der	67.85	59.99	60.00	61.37
			2nd der	68.35	61.91	60.49	62.41
HHE*	596	52	1st der	87.50	93.43	85.00	91.06
			2nd der	84.38	93.43	80.00	91.62
HHE* & lame	273	52	1st der	87.18	92.31	84.62	85.90
			2nd der	87.18	90.91	84.62	88.46
WL**	678	41	1st der	58.62	90.54	41.67	88.32
			2nd der	44.83	92.91	25.00	90.65
WL** & lame	465	41	1st der	80.77	89.09	53.33	83.57
			2nd der	80.77	89.44	53.33	84.29

keeping larger data sets but adding factors that take into account additional sources of variation during the process of modelling. This approach was recently successfully used to link methane to MIR data taking into account lactation stage (Vanlierde et al., 2015). Increasing the number of true positives (i.e. sensitivity) is rather critical as for the farmer it is more important to detect lame animals to treat them. However, in most cases, this percentage is still too low because missing to predict 30 to 40% of cows who need help is very inefficient. Furthermore, by chance, we have a 50% chance of classing an animal correctly in the lame or non lame categories as there are only two options. Therefore, predicting 60 or even 70% correctly due to the model seems to be only a small improvement compared to chance. These first results showed that the MIR technology potentially has to be used for very specific situations and that not all types of lameness can be predicted. Moreover, the different sources of variation need to be better controlled before the technology can be used on a larger scale with data coming from varied animals and farms.

## Conclusion

Results showed that the use of milk MIR spectra with the aim of detecting lameness in cows still needs additional research. First, models need to define precisely the target to be predicted and be more refined to take into account the different sources of variation that exist in the field, as only the most homogeneous data sets produced results that started to be interesting. Moreover, the specific nature of the used data requires the addition of more data coming from varied animals and farms and validation steps before using this technology on a larger scale.

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