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Author:

Edwards, Robert

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**A prospective cohort study of foot
temperature and claw horn disruption
lesions in non-lame dairy cows.**

Robert Edwards

**A dissertation submitted to the University of Bristol in accordance with the
requirements for award of the degree of Master of Science in the Faculty of
Health Sciences.**

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Abstract

Claw horn disruption lesions (CHDL's) are the cause of a large proportion of lameness in dairy cows. The early diagnosis and prompt treatment of lameness lesions have been shown to be key elements to controlling the disease on farm. The present study sought to establish whether the repeated measurement of foot temperature, recorded from the plantar pastern area using infrared thermometry, could accurately predict the presence and severity of CHDL's in non-lame post-partum dairy cows.

For this purpose the temperature of the hind feet of 112 cows were recorded three times a week for the first eight to ten weeks of lactation after which they were trimmed and examined for the presence of CHDL's. Three - level, multilevel analysis, found ambient temperature and parity to be significant predictors of foot temperature. The lesion scores in six of the fifteen different categories used to score disease were also found to be positive and significant predictors of foot temperature when added to the model.

Analysis using receiver operating characteristic curves showed all significant predictive models to have similar poor levels of accuracy at predicting disease. Accuracy was poor whether the models were predicting the presence and absence of disease defined as lesion score ≥ 1 versus no lesions, or lesion scores in the fourth quartile of scores versus lesions scores in quartile one to three.

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Contents

Abstract.....	2
Acknowledgements.....	3
Contents.....	5
List of tables.....	7
List of figures.....	8
Abbreviations.....	9
1.0 Introduction.....	10
2.0 Materials and methods.....	13
2.1 Animal management.....	13
2.2 Assessment of eligibility.....	13
2.3 Measurement of foot temperature.....	14
2.4 Claw examination.....	15
2.5 Assessment and analysis of claw photographs.....	15
2.6 Scoring of claw horn disruption lesions.....	17
2.7 Statistical analysis.....	18
3.0 Results.....	19
3.1 Basic descriptive statistics.....	19
3.2 Foot temperature predicted from ambient temperature, number of days in milk and Measurement occasion.....	21
3.3 Foot temperature predicted by parity.....	22
3.4 Selection of final predictive models.....	23
3.5 Foot temperature predicted from disease categories.....	32
3.6 Receiver operating characteristic analysis.....	34

4.0 Discussion.....	37
4.1 Foot temperature predicted from claw horn disruption lesions and time after calving.....	37
4.2 Foot temperature predicted from lesions of medial and lateral claws.....	40
4.3 The accuracy of predictive models.....	41
4.4 Foot temperature and digital dermatitis.....	43
5.0 Conclusions.....	45
References.....	47
Literature review.....	56

List of Tables

Table 1: Definition of severity scores for claw horn disruption lesions.....	17
Table 2: Descriptive statistics for ambient, foot and udder temperatures recorded during the study.....	19
Table 3: Descriptive statistics for lesion scores recorded for the 15 disease categories.....	20
Table 4: Parameter estimates and their significance from the multilevel analysis of effect of ambient temperature and DIM on foot temperature.....	21
Table 5: Descriptive statistics for foot temperature of heifers and cows.....	22
Table 6: Parameter estimates and their significance from the multilevel analysis of effect of ambient temperature and parity on foot temperature.....	23
Table 7: Descriptive statistics of lesion scores in the 15 disease categories for heifers and cows.....	24
Table 8: Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature.....	25-28
Table 9: Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature taken within the first 45 days of lactation only.....	23-31
Table 10: Area under the ROC curve evaluation of predicted foot temperature and the presence and absence of disease, for each significant disease category.....	35
Table 11: Area under the ROC curve evaluation of predicted foot temperature and the presence and absence of disease, for each significant disease category, for the first 45 days of lactation only.....	35

List of Figures

Fig 1: Example photograph of a foot after trimming and examination to be further analysed using Image J software.....	16
Fig 2: Graph showing foot temperature and DIM (with foot temperature predicted from the significant general linear model that included ambient temperature and a quartic polynomial function of DIM).....	22
Fig 3: ROC Curve of Foot sum weighted sole lesion scores (Area Under the Curve (AUC) 0.68 for lesion scores in the fourth quartile versus all other lesion scores, based on measurements collected from the first 45 days of lactation only).....	36

Abbreviations

AUC	Area Under the Curve
CHDL's	Claw Horn Disruption Lesions
COEFF	Coefficient
DIM	Days In Milk
ITT	Infrared Thermometry
N	Number of observations
ROC	Receiver Operating Characteristic
SE	Standard Error
SD	Standard Deviation
SST	Sole Soft Tissue
UK	United Kingdom

1.0 Introduction

The mean prevalence of lameness in dairy herds in the United Kingdom (UK) has shown little improvement in the last 20 years (Clarkson and others, 1996; Whay and others, 2003; Barker and others, 2010; Griffiths and others, 2018) and in many herds it continues to be a significant welfare problem and disease cost (Dolecheck and others, 2018).

Claw horn disruption lesions (CHDL's) which include sole haemorrhage, sole ulcer and white line disease (haemorrhage and separation) are the cause of a large proportion of lameness in dairy cows (Murray and others, 1996; Van Amstel and Shearer, 2006; Newsome and others, 2016). The different lesions frequently occur together at a similar stage of lactation, indicating a common aetiology (Manske and others, 2002), and typically reach peak severity nine to sixteen weeks after calving (Leach and others, 1997; Offer and others, 2000).

Claw horn disruption lesions are the end result of compression of the corium beneath the third phalanx which leads to haemorrhage and inflammation of the former (Lischer and others, 2002). Normal hormonal and metabolic events that occur around the time of calving cause mechanical weakening of the abaxial hoof suspensory apparatus, which permits greater movement of the third phalanx within the hoof capsule; likely acting as a primary factor in the aetiology of CHDL's (Tarlton and others, 2002). Prolonged compression or greater peak forces on the germinal epithelium beneath the axial aspect of the flexor tuberosity can cause haemorrhage, the formation of poor quality horn and ulcers in the "typical sole ulcer site" (Van Amstel and others, 2006) and prolonged compression or greater peak forces on the germinal epithelium beneath the abaxial margins of the third phalanx may give rise to haemorrhagic lesions of the white line (Ossent and Lischer, 1998). The presence of more severe CHDL's has been linked to many factors, some of which include: body condition and thickness of the digital cushion (Bicalho and others, 2009; Newsome and others, 2017 Part 2), the presence of bone proliferation on the caudal aspect of the third phalanx (Newsome and others, 2016), bedding type (deep straw versus cubicles) (Bergsten and other, 2015), and difference in claw height between medial and lateral claws (Bryan and others, 2012).

Cure rates for lameness are greater the earlier it is identified and treated (Reader and others, 2011) and fortnightly mobility scoring has proven itself superior to conventional farm control measures in this regard (Leach and others, 2012; Groenevelt and others, 2014). It has been reported, however, that milk yield can be reduced for a period of six to eight weeks before a lameness event is identified by fortnightly mobility scoring (Reader and others, 2011). This indicates that some lameness lesions adversely affect production, with an initial level of discomfort that does not cause detectable changes in gait. Given that cattle are prey animals it is likely that the display of pain associated with claw lesions is suppressed until a threshold is reached, and thus detection using mobility scoring systems may be delayed (O'Callaghan and others, 2003).

The health and welfare of dairy cows would benefit greatly from the ability to detect biologically significant CHDL's before they cause lameness.

Infrared thermometry (ITT), of the plantar pastern area, has been successfully used to identify feet with or without lesions based on differences in the recorded maximum foot temperature (Main and others, 2012; Wood and others, 2015). Sole ulcer, sand crack and digital dermatitis caused increases in foot temperature (not adjusted for ambient temperature) of 2.51, 2.06 and 2.16°C respectively, compared to feet with no lesions, (Main and others, 2012). In the study conducted by Wood and others (2015) in which foot temperature was recorded every two weeks, the maximum adjusted foot temperature was found to be highest at the time of lameness detection. This study also found that the foot temperature which was recorded six weeks after lameness detection, and treatment, was lower than the foot temperature which was recorded six weeks prior to the point of lameness detection. The authors concluded that the period of inflammation associated with a lameness lesion may extend at least six weeks prior to the behavioural signs of lameness being apparent.

It would be useful to further investigate the relationship between foot temperature, measured repeatedly over time, and different severity CHDL's in non-lame cows. If the future presence and severity of CHDL's was shown to be positively associated with repeated foot temperature measurements this would further support the theory that inflammation of the corium is a possible cause of increased foot temperature (Wilhelm and others, 2015; Oikonomou and others, 2014) and that foot temperature

is a proxy measurement for the inflammation status of the corium. A positive association between CHDL's and foot temperature would allow the accuracy of foot temperature to predict CHDL's to be tested. If foot temperature was shown to be an accurate predictor of CHDL's the repeated measurement of foot temperature would allow feet that are likely to develop severe CHDL's to be identified at the time when the original insults to the corium were occurring - which would allow for changes in management, accommodation and possible corrective hoof trimming to take place.

Correct examination of the relationship between foot temperature and CHDL's relies on using a scoring system for CHDL's that accurately reflects the biological significance of lesions. It is currently not known if haemorrhage of the white line and the sole are of equal biological importance, or if severity is more important than the extent of haemorrhage, or whether a combination of the severity and extent provides a more accurate assessment (Leach and others, 1998; Wilhelm and others, 2015). Because of this, researchers in the past have found it difficult to objectively classify sole and white line haemorrhages (Wilhelm and others, 2015) and many different scoring systems have been used previously (Leach and others, 1998; Offer and others, 2000; Le Fevre and others, 2001; Laven and others, 2004; Wilhelm and others, 2015). Examination of the relationship between foot temperature and the different component parts of CHDL's may help answer the question as to which system of scoring lesions more accurately reflects the inflammation status of the corium.

This longitudinal study evaluated the relationship between the maximum temperature of the hind feet of clinically non-lame dairy cows, measured repeatedly using ITT, and CHDL's present 80 – 100 days after calving. Different lesion scoring systems were used in an attempt to best examine the relationship between foot temperature and the extent and severity of lesions of the white line and sole. The aim of this study was to investigate whether ITT could be used, in non-lame dairy cows, as an accurate method to predict the presence and severity of CHDL's.

2.0 Materials and Methods

2.1 Animal management

The present study was carried out on a dairy farm in north Powys, with a herd size of 850 Holstein/Friesian cows and average 305-day milk yield of 9913kg. The farm was selected for convenience based on cows being milked three times a day on a rotary parlour, adequate handling facilities and farm managers and staff willing to accommodate the study. Cows were observed for data collection during the first 80-100 days of their lactation between January and June 2016. Milking cows were housed on mattress cubicles bedded with dried recycled paper shreds, with fresh bedding added twice weekly. Cubicle alleys were automatically scraped. Milking cows were fed a partial mixed ration with concentrate feeding “topped up” in the parlour. All milking cows were foot bathed 5 times a week, using 3% Formalin. Freshly calved cows were moved to the milking cow group within 24 hours of calving. Dry cows were housed in deep sand bedded cubicles, with cubicle alleys scraped once daily. Dry cows were fed a high straw content total mixed ration. Three to five days prior to calving, cows were moved onto a straw yard. All cows received regular foot trimming twice per lactation.

2.2 Assessment of eligibility

Cows which were parity three or younger, in the first 14 days of lactation and not receiving treatments were enrolled into an initial group of eligible cows. The mobility of all such cows was assessed on a 0-3 scale using the DairyCo Mobility Score system, as recommended by Bell and Huxley (2009): 0 representing good mobility (walks with even weight bearing and rhythm on all four feet, with a flat back), 1 Imperfect mobility (uneven rhythm or weight bearing of steps or shortened strides; affected limbs not immediately identifiable), 2 impaired mobility (uneven weight bearing on a limb that is immediately identifiable and/or obviously shortened strides that is usually accompanied with an arch to the back), 3 severely impaired mobility (cannot keep up with the healthy herd). Cows that scored 0 or 1 were judged to be clinically non-lame and enrolled into the study. Cows remained in the study until they received routine claw examination and trimming at 80 – 100 days of lactation.

Study cows had their mobility scored once a week by the same person, who did not have access to previous mobility score results. Scoring was performed by observing each cow from her right hand side, as she walked six to eight steps on a solid concrete surface from the exit of the rotary parlour to the entrance of the handling race.

No sample size calculations were performed and cows were enrolled (approximately eight to twelve cows each week) based on the number of cows that could be feasibly foot trimmed by one researcher each week over the time period available for the study. The previous lameness records of cows enrolled into the study were not consulted.

Cows were removed from the study if they became clinically lame (mobility score ≥ 2), ill or exited the herd.

2.3 Measurement of foot temperature

During the period of study, all enrolled cows had the temperature of both hind feet recorded three times a week using a hand-held infrared thermometer (product code N85FR, Maplin Electronics, Manvers, Rotherham, UK). All temperature recordings were taken at evening milking time (9pm – 11pm), while cows were standing half way round the parlour rotation. The feet were left dirty and unwashed in accordance with the findings of Stokes and others (2012). The infrared thermometer used in the study recorded a maximum temperature, with a reported accuracy $\pm 0.1^{\circ}\text{C}$. The positioning of the thermometer was determined by the crossing of two laser beams at a 15 cm distance, allowing the thermometer to be held at a fixed distance from the foot. The thermometer was moved in a standardised manner, so as to measure temperature from the plantar aspect of the pastern immediately proximal to the heel bulb and distal to the accessory digits, as described by Main and others (2012). An “automatic data hold facility” ensured that the maximum temperature was detected (and subsequently retained) while the thermometer was directed at the foot. The same infrared thermometer was used to measure the maximum skin temperature of the udder, to investigate the effect of skin temperature on foot temperature.

A digital voice recording app (Quickvoice) was used on a phone to store cow and foot identity and foot and udder temperature measurements, at the time of data collection. All data were then entered in to a spread sheet (Microsoft Excel 2010).

The ambient temperature beside the parlour was recorded to investigate the effect of environmental temperature on foot temperature. The thermometer used had a reported accuracy of +/- 1°C (product code L55AJ, Maplin Electronics, Manvers, Rotherham, UK). Ambient temperature was recorded at the beginning and end of each evening's data collection, with the mean of these two readings used for further analysis.

2.4 Claw examination

The hind feet of study cows were examined and trimmed 80-100 days after calving. This coincided with the time all cows normally received routine mid-lactation claw trimming, as part of the lameness control plan in place on the farm at that time. With each cow restrained in a crush the hind feet were raised in turn, cleaned and trimmed using a 5-Step trimming technique (Toussaint Raven, 1989). After trimming, a black marker pen was used to outline haemorrhagic lesions of the sole and sole ulcers and haemorrhagic lesions of the white line. An 8mm diameter paper circle was also stuck to the ground contact area of the lateral claw for scaling purposes. A blue plastic board was held behind the foot to give a contrasting background, upon which the identity of the cow and foot was displayed. The ground contact area of the claws was then photographed with a digital camera (Samsung WB250F, Samsung Electronics Co, China) with lens parallel to the ground contact area at a distance of about 30cm.

Any cases of digital dermatitis were recorded, using the 5 M-stage scoring system (Dopfer and others, 1997).

2.5 Assessment and analysis of claw photographs

Once all field data collection was completed a total of 224 photographs were evaluated over five consecutive days by the same person (see Fig 1). Photographs were analysed on a personal computer using Image J software (downloaded from www.imagej.en.softonic.com). Using the 8mm paper circles for calibration, the area

of the sole and the length of the white line was measured for each claw. The area of the sole was calculated as the area within the perimeter of the white line, cranial to a curved line drawn between the estimated positions of the axial and abaxial grooves, which marked the junction of the heel bulb (Leach and others, 1998). The area of all haemorrhagic lesions of the sole and the length of all haemorrhagic lesions of the white line were measured, in mm² and mm respectively.



Fig 1: Example photograph of a foot after trimming and examination to be further analysed using Image J software.

Each haemorrhagic lesion was scored for severity using a scale 1-5, based on the intensity of colour of the lesion, and sole ulcers were scored on the scale 6-7, based on the area of exposed corium (Leach and others, 1998) (see Table 1).

Table 1: Definition of severity scores for CHDL's.

Visual appearance of the lesion	Severity score
Haemorrhage	
Diffuse red in horn	1
Stronger red colouration	2
Deep dense red	3
Port colouration	4
Red raw, possibly fresh blood	5
Sole ulcer	
Corium exposed	6
Severe sole ulcer-major loss of horn	7

2.6 Scoring of claw horn disruption lesions

The CHDL's of each foot were scored in 15 different disease categories. Firstly lesions of lateral and medial claws were scored using the following six disease categories: maximum lesion score in the white line, proportional length of white line lesion(s), weighted white line lesion score, maximum lesion score in the sole ulcer region, proportional area of sole lesion(s), weighted sole lesion score.

The proportional length of each white line lesion was calculated by dividing the length of each lesion by total length of the white line, and the proportional size of each sole lesion was calculated by dividing the size of the lesion by the total area of the sole. The weighted score was generated by multiplying the proportional size of the lesion by its respective severity score, and was described by Leach and others (1998) as an attempt to combine the effects of the size and severity of lesions.

A further three disease categories were then assigned to provide overall foot scores, which were generated by summing the respective lesion categories of the medial and lateral claw: foot sum weighted white line lesion scores, foot sum weighted sole lesion scores, and foot sum weighted white line and sole lesions scores.

This scoring system was chosen because it allows foot lesions to be assigned to medial or lateral claws, the white line or the sole and severity as well as proportionate length/size can be considered when looking at associations with foot temperature during the post calving period.

2.7 Statistical analysis

All data were merged into a single spread sheet (Microsoft Excel 2010). A series of general linear models were fitted where individual parameters were tested as predictors of foot temperature. The parameters tested included ambient temperature, parity, udder temperature, and the lesion scores for each of the 15 different disease categories. These models were fitted using the multilevel software MLwiN version 3.00 (Rasbash and others, 2017) which allowed the repeated measurement of foot within cow to be accounted for within the models. Models that contained one individual variable were tested against the Standard Normal Distribution for significance ($P \leq 0.05$). A chi-squared test of the change in log likelihood ($P \leq 0.05$) was used to determine which variables were retained in models containing multiple variables. The accuracy of predicted foot temperature to correctly identify claws/feet with or without disease was tested using receiver operating characteristic (ROC) curves (IBM SPSS Statistics version 23). Foot temperatures were predicted from the significant model which included adjustments for ambient temperature and parity. Accuracy was measured by area under the ROC curve (AUC) (Carter and others, 2016). The presence or absence of disease was defined as claws/feet with 1) a lesion score ≥ 1 , versus a lesion score of zero; 2) a lesion score in the fourth quartile versus a lesion score in quartile one to three of lesion score.

3.0 Results

3.1 Basic descriptive statistics.

One hundred and fifty two cows were enrolled between January and June of 2016. Forty cows were removed from the study; 36 due to clinical lameness (mobility score ≥ 2), and four due to other illnesses, with 112 cows completing the study consisting of 50 parity 1 (heifers) and 62 parity >1 (cows). The dataset consisted of 7160 assessments of foot temperature of 224 hind feet of 112 cows. The foot temperature of both hind feet of cows was recorded on average 32 times, between seven and eighty seven days post calving, on average.

Descriptive statistics for ambient, foot and udder temperatures recorded during the study are shown in Table 2.

Table 2: Descriptive statistics for ambient, foot and udder temperatures recorded during the study

	Range	Median	Mean (SD)	N
Ambient temperature (°C)	5.2 – 26.3	13.6	15.8 (4.8)	3580
Foot temperature (°C)	11.2 – 35.9	27.8	28.1 (3.5)	7160
Udder temperature (°C)	10.4 – 38.3	31.5	31.9 (2.3)	3580

Of the 224 feet examined, at foot trimming, 53 feet had no haemorrhagic lesions of the white line and sole, 24 feet were affected by digital dermatitis, and one foot had a sole ulcer. Descriptive statistics for lesion scores recorded for each of the 15 different disease categories are listed in Table 3.

Lateral claws had numerically greater mean lesion scores than medial claws in all six disease categories. The mean proportional length of white line lesion(s) ($P < 0.001$), mean weighted white line score ($P < 0.001$), mean proportional area of sole lesions ($P < 0.001$) and mean weighted sole lesion score ($P < 0.001$) were all higher in lateral claws compared to medial claws.

Table 3: Descriptive statistics for lesion scores recorded for the 15 disease categories.

Disease category	Range	Median	Mean (SD)
Lateral claw, maximum score in the white line	0 – 4.00	1.00	0.68 (0.97)
Lateral claw, proportional length of white line lesion(s)	0 – 66.10	10.15	10.04 (14.08)
Lateral claw, weighted white line lesion scores	0 – 155.60	20.30	15.33 (26.24)
Lateral claw, maximum lesion score in sole ulcer region	0 – 5.00	3.00	1.11 (1.36)
Lateral claw, proportional area of sole lesions	0 – 57.50	12.40	10.91 (12.80)
Lateral claw, weighted sole lesion score	0 – 106.5	22.10	18.83 (24.29)
Medial claw, maximum score in the white line	0 – 4.00	0	0.499 (0.90)
Medial claw, proportional length of white line lesion(s)	0 – 63.40	0	6.71 (12.16)
Medial claw, weighted white line lesion scores	0 – 145.60	0	10.65 (22.12)
Medial claw, maximum lesion score in sole ulcer region	0 – 6.00	0	0.18 (0.70)
Medial claw, proportional area of sole lesions	0 – 45.80	1.65	2.21 (5.81)
Medial claw, weighted sole lesion score	0 – 53.20	4.95	3.79 (9.56)
Foot, sum of weighted white line lesion scores	0 – 259.70	20.30	25.80 (36.63)
Foot, sum of weighted sole lesion scores	0 – 109.50	27.05	22.85 (26.94)
Foot, sum of weighted white line and sole lesion scores	0 – 266.90	47.35	48.47 (51.04)

3.2 Foot temperature predicted from ambient temperature, number of days in milk and measurement occasion.

A general linear model was developed in which foot temperature was predicted from ambient temperature and the number of Days in Milk (DIM) during the post calving period (Table 4). Ambient temperature was represented in the model by a variable centred round the general mean ambient temperature for this study (15.8°C), which showed that for every 1°C increase above ambient temperature, foot temperature was increased by 0.192°C. Days in Milk was found to be significant predictor of foot temperature. Further examination of the relationship between foot temperature and DIM involved the addition of variables which represented first, second, third and fourth degree polynomial functions of DIM. Foot temperature was found to be best predicted, in the model, by a variable which represented a fourth degree (quartic) polynomial function of DIM (Fig 2).

Table 4: Parameter estimates and their significance from the multilevel analysis of effect of ambient temperature and Days in Milk (DIM) on foot temperature.

Parameter	COEFF	SE	P value
Constant	26.526	0.167	0
Ambient temperature*	0.192	0.008	<0.001
DIM ¹	0.13	0.02	<0.001
DIM ²	-0.004	0.001	<0.001
DIM ³	0	0	<0.001
DIM ⁴	0	0	0.001

*Ambient temperature – general mean of ambient temperature

The relationship between ambient temperature and the occasion within the study that foot temperature was measured was examined. Measurement occasion was found to be a significant predictor of ambient temperature (P<0.001). The positive association between measurement occasion and ambient temperature meant that ambient temperature was rising as the study proceeded for each cow. Since ambient temperature was a significant predictor of foot temperature, the former could act to mask the true effect that claw lesions had on foot temperature. Separate models were used to test the ability of each individual disease category to predict foot temperature. To avoid the potential for ambient temperature to confound the results a further parameter was added to each model which represented a first order

interaction between the disease category variable being tested and the term for ambient temperature. Addition of this parameter would allow the model to take into account all possible combinations of lesion score for that disease category and ambient temperature.

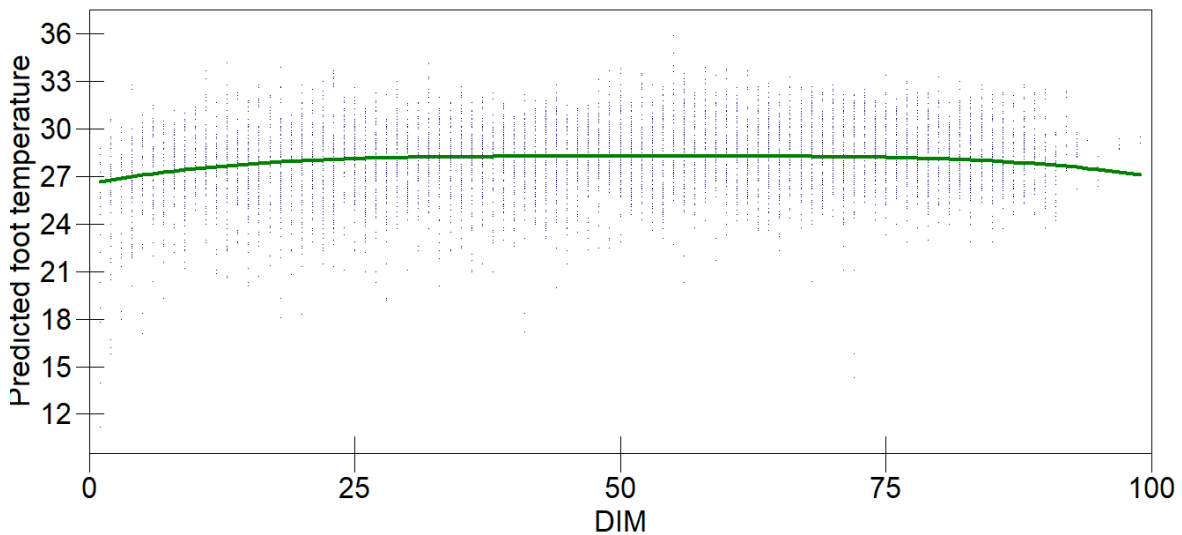


Fig 2: Graph showing foot temperature according to Days in Milk (DIM).

3.3 Foot temperature predicted by parity.

Heifers had a greater range of foot temperature and a numerically higher mean foot temperature than cows (see Table 5).

Table 5: Descriptive statistics for foot temperature of heifers and cows.

Foot temperature (°C)	Range	Median	Mean (SD)	N
Heifers	11.2 – 32.2	26.95	27.1 (2.8)	3186
Cows	15.8 – 33.7	28.60	26.7 (2.8)	3974

After adjusting for ambient temperature, the foot temperature of heifers was 0.756°C higher than cows ($P < 0.001$) (see Table 6).

Heifers had higher mean lesion scores than cows for 12 of the 15 disease categories ($P < 0.001$) (see Table 7). Cows had higher mean lesion scores than heifers for lateral claw maximum score in the white line ($P < 0.033$) and lateral claw weighted white line lesion score ($P < 0.033$). There was no difference in mean lesion score between cows and heifers for the disease category lateral claw proportional length white line lesion(s).

Table 6: Parameter estimates and their significance from the multilevel analysis of effect of ambient temperature and parity on foot temperature

Parameter	COEFF	SE	P-value
Constant	29.26	0.23	
Ambient temperature*	0.222	0.007	<0.001
Parity**	-0.756	0.141	<0.001

*Ambient temperature – general mean of ambient temperature

** Parity – 0 = Heifer, 1 = cow

After accounting for all the possible effects of increased lesion scores recorded for heifers, parity remained a significant predictor of foot temperature and was therefore retained as an explanatory variable in all further models. However because parity was also likely to mask the effect of disease on foot temperature, it was decided that an additional parameter should be added to each predictive model. This parameter represented a first order interaction between the disease category variable that was being tested and the term for parity. Addition of such a parameter would allow the model to take into account all possible combinations of lesion score for that disease category and parity.

3.4 Selection of final predictive models.

The final models which predicted foot temperature were constructed starting with the following variables: ambient temperature (centred round its general mean), parity, each disease category variable, and the inclusion of two further parameters representing first order interactions between the disease category variable and ambient temperature and the disease category variable and parity. The models listed in Table 8 represent what were considered to be the best models for each disease category variable. In an effort to better identify the period of time when insults to the corium would have been responsible for the CHDL'S that were identified, a second series of similarly constructed models were developed from a dataset that was restricted to foot temperature measurements recorded from the first 45 days of lactation only (see Table 9).

Table 7: Descriptive statistics of lesion scores in the 15 disease categories for heifers and cows.

Disease category	Range		Median		Mean (SD)	
	Heifers	Cows	Heifers	Cows	Heifers	Cows
Lateral claw, maximum score in the white line	0 - 4	0 - 4	0.5	2	0.66† (0.91)	0.69† (1.03)
Lateral claw, proportional length of white line lesion(s)	0 – 47.2	0 – 66.1	5.65	32.9	10.07 (13.00)	9.68 (14.81)
Lateral claw, weighted white line lesion scores	0 – 128.0	0 – 155.6	5.65	65.8	14.86† (23.78)	15.49† (28.35)
Lateral claw, maximum lesion score in sole ulcer region	0 - 5	0 - 4	3	2.5	1.42* (1.45)	0.83* (1.23)
Lateral claw, proportional area of sole lesions	0 – 55.7	0 – 57.5	17.3	2.74	14.89* (13.33)	7.60* (11.77)
Lateral claw, weighted sole lesion score	0 – 106.5	0 – 95.4	31.9	5.52	27.02* (26.89)	12.09* (20.47)
Medial claw, maximum score in the white line	0 - 4	0 - 3	1	0	0.73* (1.02)	0.28* (0.69)
Medial claw, proportional length of white line lesion(s)	0 – 63.4	0 – 60.4	8.4	0	9.26* (13.80)	4.26* (9.99)
Medial claw, weighted white line lesion scores	0 – 111.5	0 – 145.6	16.8	0	14.61* (24.19)	6.81* (19.21)
Medial claw, maximum lesion score in sole ulcer region	0 - 5	0 - 4	0.5	0	0.30* (0.90)	0.09* (0.49)
Medial claw, proportional area of sole lesions	0 – 45.8	0 – 13.7	22.9	1.65	4.00* (7.81)	0.62* (2.17)
Medial claw, weighted sole lesion score	0 – 53.2	0 – 46.2	26.6	4.95	6.69* (12.08)	1.25* (5.58)
Foot, sum of weighted white line lesion scores	0 – 139.1	0 – 259.7	22.45	65.8	28.76* (33.57)	22.57* (38.82)
Foot, sum of weighted sole lesion scores	0 – 109.5	0 – 95.4	58.5	10.47	34.51* (29.13)	13.07* (21.25)
Foot, sum of weighted white line and sole lesion scores	0 – 222.8	0 – 266.9	80.95	76.27	62.90* (49.65)	35.65* (49.63)

*P<0.001, †P<0.033

Table 8: Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	29.299	0.236	<0.001	29.278	0.262	<0.001	29.782	0.271	<0.001
Ambient temperature*	0.217	0.007	<0.001	0.234	0.009	<0.001	0.223	0.007	<0.001
Parity	-0.766	0.142	<0.001	-0.756	0.147	<0.001	-1.041	0.161	<0.001
Lateral claw maximum score in the sole ulcer region recoded (0-3=0, 4-5=1)	-0.357	0.242							
Lateral claw maximum score in the sole ulcer region recoded (0-3=0, 4-5=1) x Ambient temperature*	0.058	0.023	0.011						
Lateral claw proportional area sole lesions				-0.001	0.006				
Lateral claw proportional area sole lesions x Ambient temperature*				-0.001	0.001	0.030			
Medial claw maximum score in the white line							-0.823	0.241	0.0006
Medial claw maximum score in the white line xParity							0.500	0.167	0.003

*Ambient temperature – general mean of ambient temperature

Table 8 (cont): Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	29.308	0.246	<0.001	29.563	0.262	<0.001	29.324	0.235	<0.001
Ambient temperature*	0.235	0.008	<0.001	0.230	0.007	<0.001	0.218	0.007	<0.001
Parity	-0.768	0.145	<0.001	-0.918	0.157	<0.001	-0.781	0.142	<0.001
Medial claw proportional length white line lesions	-0.003	0.006							
Medial claw proportional length white line lesions x Ambient temperature*	-0.002	0.001	<0.001						
Medial claw weighted white line lesion score				-0.022	0.010	0.028			
Medial claw weighted white line lesion score x Ambient temperature*				-0.001	0.0003	0.012			
Medial claw weighted white line lesion score x Parity				0.013	0.007	0.042			
Medial claw maximum lesion score in sole ulcer region							-0.175	0.097	
Medial claw maximum lesion score in sole ulcer region x Ambient temperature*							0.024	0.01	0.018

*Ambient temperature – general mean of ambient temperature

Table 8 (cont): Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	29.467	0.247	<0.001	29.214	0.240	<0.001	29.929	0.340	<0.001
Ambient temperature*	0.223	0.007	<0.001	0.234	0.008	<0.001	0.222	0.007	<0.001
Parity	-0.848	0.146	<0.001	-0.735	0.142	<0.001	-1.121	0.195	<0.001
Medial claw weighted sole lesion score	-0.017	0.008	0.028						
Foot sum weighted white line lesion scores				0.0008	0.0019				
Foot sum weighted white line lesion scores x Ambient temperature*				-0.0005	0.0002	0.012			
Foot sum weighted sole lesion scores							-0.022	0.008	
Foot sum weighted sole lesion scores x Parity							0.014	0.006	0.017

*Ambient temperature – general mean of ambient temperature

Table 8 (cont): Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature.

Parameter	COEFF	SE	P value
Constant	29.874	0.354	<0.001
Ambient temperature*	0.237	0.009	<0.001
Parity	-1.102	0.203	<0.001
Foot sum weighted white line and weighted sole lesion scores	-0.011	0.005	0.0278
Foot sum weighted white line and weighted sole lesion scores × Ambient temperature*	-0.0003	0.0001	0.015
Foot sum weighted white line and weighted sole lesion scores × Parity	0.007	0.003	0.016

*Ambient temperature – general mean of ambient temperature

Table 9: Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature taken within the first 45 days of lactation only.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	29.255	0.461	<0.001	29.290	0.437	<0.001	29.462	0.377	<0.001
Ambient temperature*	0.245	0.013	<0.001	0.246	0.013	<0.001	0.248	0.013	<0.001
Parity	-1.047	0.267	<0.001	-1.054	0.255	<0.001	-1.125	0.223	<0.001
Lateral claw weighted sole lesion score				-0.026	0.012	0.03			
Lateral claw weighted sole lesion score x Parity				0.018	0.008	0.03			
Medial claw maximum score in the white line							-1.198	0.333	0.0003
Medial claw maximum score in the white line xParity							0.764	0.230	0.001

*Ambient temperature – general mean of ambient temperature

Table 9 (cont): Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature taken within the first 45 days of lactation only.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	28.965	0.376	<0.001	29.039	0.366	<0.001	28.992	0.345	<0.001
Ambient temperature*	0.246	0.012	<0.001	0.247	0.013	<0.001	0.249	0.013	<0.001
Parity	-0.888	0.224	<0.001	-0.902	0.219	<0.001	-0.832	0.204	<0.001
Medial claw proportional length white line lesions	-0.040	0.025							
Medial claw proportional length white line lesions x Parity	0.033	0.033	0.046						
Medial claw weighted white line lesion score				-0.026	0.014				
Medial claw weighted white line lesion score x Parity				0.018	0.366	0.044			
Medial claw proportional area sole lesions							-0.036	0.018	0.046

*Ambient temperature – general mean of ambient temperature

Table 9 (cont): Parameter estimates and their significance from multilevel analysis of effect of ambient temperature, parity and disease category lesion score on foot temperature taken within the first 45 days of lactation only.

Parameter	COEFF	SE	P value	COEFF	SE	P value	COEFF	SE	P value
Constant	29.041	0.342	<0.001	29.684	0.472	<0.001	29.475	0.493	<0.001
Ambient temperature*	0.250	0.013	<0.001	0.247	0.013	<0.001	0.245	0.013	<0.001
Parity	-0.851	0.203	<0.001	-1.247	0.270	<0.001	-1.176	0.283	<0.001
Medial claw weighted sole lesion score	-0.026	0.011	0.016						
Foot sum weighted sole lesion scores				-0.033	0.012	0.006			
Foot sum weighted sole lesion scores x Parity				0.021	0.008	0.009			
Foot sum weighted white line and weighted sole lesion scores							-0.014	0.006	0.02
Foot sum weighted white line and weighted sole lesion scores x Parity							0.009	0.004	0.016

*Ambient temperature – general mean of ambient temperature

3.5 Foot temperature predicted from disease categories.

Of the models that included foot temperature from the entire period of study, six disease categories were found to be positive and significant predictors of foot temperature and four disease categories were found to be negative and significant predictors of foot temperature.

1. Lateral claw maximum score in the sole ulcer region when recoded from 0-5 to a binary score (0-3=0, 4-5=1) when included in an interaction term with ambient temperature had a positive and significant effect on foot temperature.
2. Lateral claw proportional area sole lesions when included in an interaction term with ambient temperature had a negative and significant effect on foot temperature.
3. Medial claw maximum score in the white line had a negative and significant effect on foot temperature, and Medial claw maximum score in the white line when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.
4. Medial claw proportional length white line lesions when included in an interaction term with ambient temperature had a negative and significant effect on foot temperature.
5. Medial claw weighted white line lesion score had a negative and significant effect on foot temperature and when included in an interaction term with ambient temperature had a further negative and significant effect on foot temperature. Medial claw weighted white line lesion score when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.
6. Medial claw maximum lesion score in sole ulcer region when included in an interaction term with ambient temperature had a positive and significant effect on foot temperature.
7. Medial claw weighted sole lesion score had a negative and significant effect on foot temperature.

8. Foot sum weighted white line lesion scores when included in an interaction term with ambient temperature had a negative and significant effect on foot temperature.
9. Foot sum weighted sole lesion scores when included in an interaction term with parity had a positive and significant effect on foot temperature.
10. Foot sum weighted white line and weighted sole lesion scores had a negative and significant effect on foot temperature and when included in an interaction term with ambient temperature had a further negative and significant effect on foot temperature. Foot sum weighted white line and weighted sole lesion scores when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.

Of the models that included foot temperature from just the first 45 days of lactation, six disease categories were found to be positive and significant predictors of foot temperature and two disease categories were found to be negative and significant predictors of foot temperature.

1. Lateral claw weighted sole lesion score had a negative and significant effect on foot temperature and when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.
2. Medial claw maximum score in the white line had a negative and significant effect on foot temperature and when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.
3. Medial claw proportional length white line lesions had positive and significant effect on foot temperature.
4. Medial claw weighted white line lesion score had positive and significant effect on foot temperature.
5. Medial claw proportional area sole lesions had negative and significant effect on foot temperature.
6. Medial claw weighted sole lesion score had negative and significant effect on foot temperature.

7. Foot sum weighted sole lesion scores had a negative and significant effect on foot temperature and when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.
8. Foot sum weighted white line and weighted sole lesion scores had a negative and significant effect on foot temperature and when included in an interaction term with parity had a positive and significant effect on foot temperature and the addition of this term provided a significant improvement in overall model fit.

Notably, none of the disease categories involving the white line of the lateral claw were found to be positive and significant predictors of foot temperature in either series of models. Digital dermatitis was not found to be a significant predictor of foot temperature in any of the models fitted in the present study.

3.6 ROC analysis.

The area under the ROC curve for disease categories that had been found to be positive and significant predictors of foot temperature (after adjusting for the effect of ambient temperature and parity) ranged from 0.54 to 0.68 for the entire study period, and 0.52 to 0.68 for the first 45 days of lactation only, (see Tables 10 and 11).

There appeared to be little difference in accuracy whether the presence or absence of disease was defined as having no lesions versus a lesion score ≥ 1 or having a lesion score in the fourth quartile of scores versus claws/feet with lesion scores in quartile one to three of lesion scores.

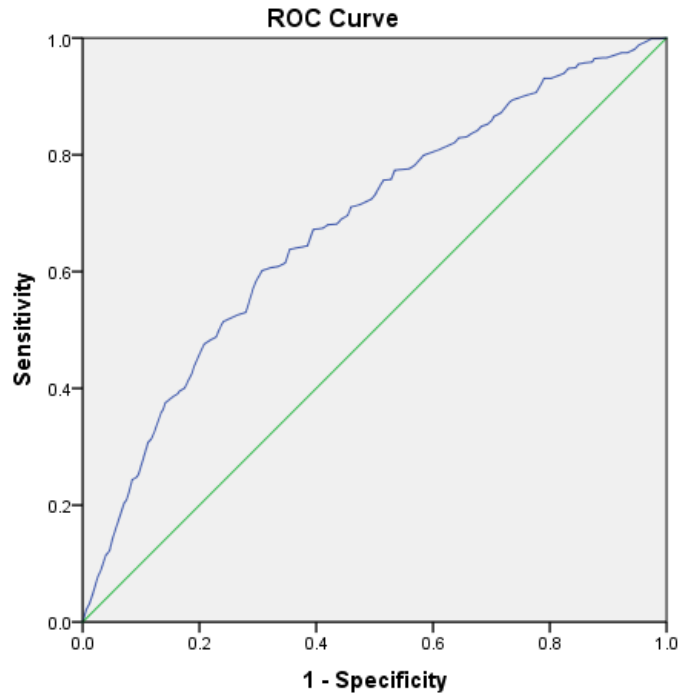
Considering lesions of the foot as a whole, the highest AUC value was achieved by foot sum weighted sole lesion scores (AUC = 0.68), when the presence of disease is defined as lesion scores \geq fourth quartile score and the absence of disease defined as lesion scores $<$ fourth quartile score (see Fig 3). The point where the minimum distance line crosses the ROC curve denotes the optimum sensitivity and specificity (Carter and others, 2016). With reference to Fig 3, the optimum sensitivity and specificity is 63% and 64% respectively, which would be achieved by setting the foot temperature threshold at 27.6°C.

Table 10: Area under the ROC curve evaluation of predicted foot temperature and the presence and absence of disease, for each disease category previously found to be positive and significant predictors of foot temperature.

Disease category	Area under the curve (lesion versus no lesions)	Area under the curve (lesions in 4th quartile versus other quartiles)
Lateral claw maximum score in the sole ulcer region	0.67	0.54
Medial claw maximum score in the white line	0.64	0.60
Medial claw weighted white line lesion score	0.64	0.63
Medial claw maximum score in the sole ulcer region	0.68	0.55
Foot sum weighted sole lesion scores	0.65	0.68
Foot sum weighted white line and weighted sole lesion scores	0.64	0.63

Table 11: Area under the ROC curve evaluation of predicted foot temperature and the presence and absence of disease, for each disease category previously found to be positive and significant predictors of foot temperature, for the first 45 days of lactation only

Disease category	Area under the curve (lesion versus no lesions)	Area under the curve (lesions in 4th quartile versus other quartiles)
Lateral claw weighted sole lesion score	0.57	0.52
Medial claw maximum score in the white line	0.64	0.64
Medial claw proportional length of white line lesions	0.64	0.59
Medial claw weighted white line lesion score	0.64	0.61
Foot sum weighted sole lesion scores	0.66	0.68
Foot sum weighted white line and weighted sole lesion scores	0.64	0.62



Diagonal segments are produced by ties.

Figure 3: ROC Curve of Foot sum weighted sole lesion scores (AUC 0.68 for lesion scores in the fourth quartile versus all other lesion scores, based on measurements collected from the first 45 days of lactation only).

4.0 Discussion

The aim of the present study was to investigate whether the repeated measurement of foot temperature using ITT, during the post calving period, could be used as an accurate method to detect feet with CHDL's in clinically non lame dairy cows (mobility score <2, scale 0-3). Lesions were scored using fifteen different categories to allow any relationship between foot temperature and the severity and extent of haemorrhagic lesions of the white line and sole to be fully investigated.

4.1 Foot temperature predicted from claw horn disruption lesions and time after calving.

The present study successfully demonstrated that claw horn disruption lesions, measured at 80-100 days after calving, were significant predictors of foot temperature during the post calving period. Overall the lesion scores in six of the fifteen different disease categories were found to be positive and significant predictors of foot temperature, and these were: 1) lateral claw maximum lesion score in the sole ulcer region, 2) medial claw maximum lesion score affecting the white line, 3) medial claw weighted white line lesion score, 4) medial claw maximum lesion score in the sole ulcer region, 5) foot sum weighted sole lesion scores, 6) foot sum weighted white line and sole lesion scores.

These results differ from the findings of Wilhelm and others (2015) who were not able to demonstrate any meaningful correlation between foot temperature, measured once, in the first week after calving, and claw lesions, measured eight weeks after calving. This study was designed around the fact that it takes two to three months for haemorrhages to become visible at the ground contact area of the claw following insult to the corium (Nocek, 1997). In the present study CHDL's were measured on the ground contact area of the claws after therapeutic trimming. This likely means that the lesions which were recorded were the result of insults to the corium that took place less than two to three months earlier. In the present study foot temperature was measured on average 32 times in the first 87 days of lactation, which meant it was not necessary to attempt to precisely pinpoint the time when insults to the corium would have resulted in the lesions recorded at foot trimming. The repeated measurement of foot temperature is likely one of the reasons why an association

between foot temperature and CHDL's was proven in the present study and not in the study of Wilhelm and others (2015).

It is difficult to explain why four of the disease categories were found to be negative and significant predictors of foot temperature. However, many of the negative coefficients of these disease category variables were very small (especially in the models generated from the full dataset). This indicates that such negative correlations, although significant, were likely to be very small.

The digital cushion has the important role of dissipating the concussive forces transferred through the third phalanx onto the underlying corium (Raber and others, 2006). Bicalho and others (2009) reported that the thickness of the digital cushion decreases steadily starting in the first month of lactation and reaches a nadir at 120 days after calving. They also found that thickness of the digital cushion is highly negatively associated with the prevalence of sole ulcers and white line disease, and is a strong predictor of lameness. Oikonomou and others (2014) demonstrated that the thickness of the digital cushion is negatively associated with sole temperature. They suggested that reduced thickness of the digital cushion may allow increased mechanical forces to act on the corium, causing inflammation and increased sole temperature and the likely future development of CHDL's.

The repeated measurement of foot temperature, in the present study, allowed the effect of DIM on foot temperature to be examined. Figure 2 shows predicted foot temperature rising after calving to reach a plateau after approximately 50 days, which is continued at that level before dropping off at 80-100 days after calving. If it is assumed that maximum foot temperature, like sole temperature, is a possible measure of inflammation within the corium then the DIM parameter is acting as a proxy measurement for disease processes within the foot. This would indicate that inflammation of the corium increases after calving and reaches a maximum level approximately 50 days in to lactation and is then maintained until approximately 80-100 days after calving, before declining.

Recent work by Newsome and others Part 2 (2017) showed the future likelihood of severe sole haemorrhage, sole ulcers and lameness were associated with thin sole soft tissues (SST) (combination of the digital cushion and the corium) but not a

reduction in thickness of SST in the post calving period, which is in contrast to the findings of Bicalho and others (2009). This indicates that the severity of CHDL's (and presumably the prior level of inflammation within the corium that gives rise to them) can vary independently of changes in thickness of SST.

The second series of models that only considered the reduced dataset (restricted to foot temperature measurements recorded from the first 45 days of lactation) were developed in an attempt to better identify the period of time when insults to the corium would have caused the CHDL'S identified in the study. Statistical analysis of the reduced dataset produced results which were very similar to those given by analysis of the full dataset; six disease categories were found to be positive and significant predictors of foot temperature, with four of these six disease categories being significant in the analysis of both datasets, and those were: medial claw maximum score in the white line, medial claw weighted white line lesion score, foot sum weighted sole lesion scores, foot sum weighted white line and weighted sole lesion scores. Such findings indicate that claw horn development lesion scores in these disease categories can positively predict foot temperature when restricted to the first 45 days of lactation and when extending to 80-100 days after calving.

In the reduced dataset parity was included as an interaction term in all six of the models which contained disease category variables that were positive and significant predictors of foot temperature, but was only included in four out of the six models developed from the full dataset. This might suggest that parity confounded the effects of disease on foot temperature more during the first 45 days of lactation than compared to the entire period of study.

The feet of animals were not examined for CHDL's at the start of the study and it is likely that a number of animals would have had some lesions at that time (Offer and others, 2000; Wilhelm and others, 2015). It is unlikely that the failure to identify and record any such lesions would have affected the results obtained in the present study. If animals had moderate to severe lesions, which were progressing, when they were enrolled, it is likely that they would have been identified as clinically lame by the weekly mobility scoring that was performed as part of the study (Groenevelt and others, 2014). In fact 36 animals were removed during the course of the study due to clinical lameness. The fact that only one animal had a sole ulcer identified at

claw trimming and examination is also testament to the fact that animals which completed the study did not have advanced CHDL's. Any cows, at the start of the study, affected by less severe CHDL's would be the result of damage to the corium that occurred two to three months earlier (Nocek, 1997). These "historic" lesions would be worn away from the ground surface of the claws over the course of the study from standing and walking activity, and further removed by the therapeutic trim performed on each foot at 80-100 days after calving.

4.2 Foot temperature predicted from lesions of medial and lateral claws.

More disease categories of the medial claw were found to be predictive of foot temperature than of the lateral claw (three out of six versus one out of six categories respectively). None of the disease categories involving the white line of the lateral claw were predictive of foot temperature. This was an unexpected finding since lateral claws were more affected by lesions than medial claws, in both the white line and the sole, which is in agreement with Nikkhah and others (2005) and Wilhelm and others (2015). Alsaad and others (2015) measured the temperature of lateral and medial hind claws prior to trimming. They found that the temperature of the coronary band and the skin just proximal to it was warmer in lateral claws compared to medial claws. This was thought to be because lateral claws bear more weight than medial claws causing them to be more affected by CHDL's and have a faster rate of horn wear and compensatory growth, which likely requires an increased supply of blood and nutrients to the corium. In the present study foot temperature was measured by the technique described by Main and others (2012) which did not entail recording the temperature of individual claws, but rather the maximum temperature from the area just distal to the accessory digits and proximal to the coronary band of the heel. If the skin of the heel bulb of the lateral claw was warmer than that of the medial claw then it is this temperature that would be recorded as the overall foot temperature, which would lead us to expect that lesion scores of disease categories of the lateral claw should be better predictors of foot temperature than lesion scores of disease categories of medial claw.

The vascular systems of the medial and lateral claw, distal to the main arterial supply and venous drainage, have not been shown to be different in clinically normal cattle

(Vermunt and others, 1992). Inflammation of the corium of either claw should therefore have similar effects on skin temperature in the plantar pastern area, which does not explain why foot temperature is better predicted by lesion scores of disease categories of the medial claw compared to the lateral claw. Some CHDL's have been shown to be associated with changes to the normal vascular supply to the claw. Sole haemorrhage, sole ulcers and white line disease, have all been associated with a greater numbers of anastomoses and an increased number of bulbar vessels (Singh and others, 1994). These additional arterial tributaries originate, at least in part, from the proper plantar digital artery and the abaxial proper plantar digital artery (Vermunt and others, 1992). Greater blood flow in these vessels, is likely to increase heat radiated from the skin (Alsaad and others, 2015), overlying the heel bulb of the claw. These findings therefore also suggest that the disease categories of the lateral claws, with their higher lesion scores, should be better predictors of foot temperature than that of the medial claws.

4.3 The accuracy of predictive models.

The model which predicted foot temperature, after adjusting for ambient temperature and parity, was not found to be an accurate means of identifying claws/feet with or without disease lesions for the categories that had previously been found to positively and significantly predict foot temperature. There was very little difference between AUC values for disease categories of the sole versus the white line, or the medial claw versus the lateral claw or between individual claws and the scoring systems used to assess the foot as a whole. It was thought that foot temperature might have more accurately predicted the presence of lesions for disease categories considered to be of high biological importance eg lateral claw maximum lesion score in the sole ulcer region, and the disease categories that considered the foot as a whole, in which lesion scores represented the total damage/inflammation of the corium of the white line and sole over both claws. This, however, was not the case. Evaluation of ROC curves also showed that the model used to predict foot temperature was no more accurate at identifying feet with lesions scores in the fourth quartile of scores versus all other scores than it was at identifying feet with or without a lesion, and this was the same for the significant disease categories tested from the dataset restricted to the first 45 days of lactation.

The fact that models were constructed which demonstrated positive associations between predicted foot temperature and the lesion scores for numerous disease categories but were unable to accurately identify feet with or without lesions can be explained by one of two ways.

Firstly although a variation in lesion scores existed between individual cows within each disease category it is possible that the higher scoring lesions did not occur frequently enough, or they simply were not severe enough to be biologically significant. This means that although damage/inflammation of the corium occurred at some point which caused a detectable increase in foot temperature which could be correlated with a claw lesion that was detected a number of weeks later, the association was weak such that the predicted model was not accurate at differentiating between the presence and absence of disease, which was defined in a number of different ways. The objective of the present study was to investigate whether ITT could be used as an accurate method to predict the presence and severity of CHDL's in non-lame cows and in so doing establish whether or not such technology might be suitable for the early identification of high risk cows, before they become lame. It is unfortunate that it was not possible to record the CHDL'S on the feet of the 36 cows which were removed from the study, because of lameness. This additional information would have enabled the lesion scores of freshly lame cows and non-lame cows to be compared and would have therefore provided valuable information about the relative severity of the lesion scores recorded in the non-lame cows and the frequency with which they occurred. It was not possible to examine the feet of lame cows excluded from the study because of the farm policy to treat all lame cows within 48 hours of lameness detection and because of the farms ability to accommodate a more complex study.

Secondly it is possible that the system used to score the CHDL's did not place sufficient emphasis on the more severe lesions. It is not easy to score CHDL's objectively and in a manner that reflects their likely biological significance and many different scoring systems have been used in previous studies (Leach and others, 1998; Offer and others, 2000; Le Fevre and others, 2001; Laven and others, 2004; Wilhelm and others, 2015). In the present study maximum lesion score (as an indicator of severity), proportional length/area (as an indicator of extent) and weighted scores (as an indicator of severity and extent) were all well represented

amongst models that were significant predictors of foot temperature. Leach and others (1998) used a geometric series $x = 2^{(n-1)}$ to score severity, with the intention of giving greater emphasis to the more severe lesions that would likely be of greater biological significance. Thomas and others (2015) used a similar weighted scoring system which was proven to be relevant in a clinical situation whereby cows with higher lesion scores were proven to take longer to recover from lameness. It might therefore have been more appropriate to have used a similar weighted system for scoring lesion severity, in the present study. Such a system might have resulted in a stronger association between predictive foot temperature and lesion scores and the new predictive models being more accurate at differentiating between the presence and absence of disease.

It should also be noted that this research was conducted on just one farm. It is likely that the association between foot temperature and CHDL's varies between farms and it is therefore possible that foot temperature may be a more accurate predictor of CHDL's on other farms. Stokes and others (2012) found that the temperature of feet with CHDL's did not differ significantly if the feet were dirty or clean, and it is for that reason that feet were left dirty prior to recording foot temperature in the present study. However the hygiene of feet of many cows in the study were sub optimal; largely a result of the farm using automatic scrapers, having relatively narrow cubicle and feed alleys and not providing a loafing area. Hence it is possible that the feet of cows measured in the present study were dirtier than the feet of cows recorded by Stokes and others (2012), which may have had effects on foot temperature and its association with CHDL's.

4.4 Foot temperature and digital dermatitis.

Digital dermatitis was found to affect twenty four feet. Unlike previous studies, which have used the same technique of ITT (Main and others, 2012; Stokes and others, 2012) digital dermatitis was not found to be a significant predictor of foot temperature in any of the models fitted in the present study. At the time of foot examination the digital dermatitis lesions were all categorised as small M3 lesions, which is recognised as a non-infectious stage of the disease (Dopfer and others, 1997; Alsaad and others, 2014). It is not possible to say whether any of these lesions may have been at an infectious stage earlier in the period of study, although none of

the affected cows were judged to be clinically lame at any point during this time. The difference in results between this study and Main and others (2012) and Stokes and others (2012) might be explained by them having a higher number of cases with infectious lesions (M2 and M4), however M stage categories were not described in these studies.

5.0 Conclusions

Many different factors increase the risk of CHDL's. Some of these include: high 305 day milk yield in the previous lactation, body condition score ≤ 2.75 at dry off, age (\geq third lactation), previous history of CHDL's (Foditsch and others, 2016), loss of back fat thickness after calving and thin sole soft tissue thickness (Newsome and others, 2017), difference in claw height between medial and lateral claws (Bryan and others, 2012), and housing in cubicles with sparse bedding (Barker and others, 2009; Griffiths and others, 2018). To reduce the prevalence of lameness due to CHDL's aggressive preventative measures should be adopted for high risk cows (Foditsch and others, 2016), which should include their early identification.

Main and others (2012) were the first to demonstrate detectable differences in foot temperature, within a herd, using a low cost, hand-held infrared thermometer and recognised that further investigation into the use of this technique might be worthwhile. If it was demonstrated that infrared thermometry could accurately predict the development of severe CHDL's, this low cost technology might be suitable for use in an automated system that monitored foot temperature, for example in robotic milking stations or in rotary parlours.

The present study demonstrated that, after adjusting for ambient temperature, ITT of the plantar pastern area of non-lame cows was able to detect changes in foot temperature over the course of the post calving period, which was presumably associated with the disease process of the lesions, namely the inflammatory status of the corium. This research also found that ITT was able to detect the presence of elevated foot temperature associated with CHDL's, after adjusting for ambient temperature and parity. More specifically foot temperature predicted the severity of haemorrhage in the sole ulcer region of lateral claws and the severity of haemorrhage in the white line and the weighted white line lesion scores of medial claws. The weighted white line lesion scores and the weighted sole lesion scores summed over both claws were also predicted by foot temperature. Despite these significant associations, foot temperature was not found to be an accurate predictor of presence or absence or the severity of CHDL's.

As mentioned earlier it may prove beneficial to repeat the statistical analysis using a different system of scoring lesion severity such as that used by Thomas and others

(2015). It is however possible that foot temperature may only be an accurate predictor of more severe haemorrhagic lesions of the white line and sole. Further studies on a larger number of cows with severe CHDL's, and preferably on multiple farms, would be needed to establish this.

According to Sogstad and others (2005) cows are most at risk from lameness five to seven months after calving, with the highest incidence of white line disease and sole haemorrhage occurring between three to five and five to seven months after calving, respectively. Herds in which routine preventative foot trimming of cows 60 – 100 days after calving have a significantly lower mean adjusted lameness prevalence (14% versus 23.3%) than herds which do not employ the practice (Griffiths and others., 2018). In the absence of readily available technology capable of accurately identifying cows at high risk of developing CHDL's, most dairy farmers would therefore be well advised to adopt the practice of early lactation preventative trimming of all cows.

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Literature review

Introduction

Lameness in dairy cows, in the UK, has been recognised as a widespread problem with serious animal welfare and economic implications, for over 20 years (Whitaker and others, 2004). Studies conducted during this time suggest that the problem may not be improving and that there continues to be a large variation in lameness prevalence between different farms; the mean (range) prevalence of lameness was estimated to be 20.6% (2.0 to 53.9) (Clarkson and others, 1996), 22.1% (0 to 50) (Whay and others, 2003) and 36.8% (0 – 79.2) (Barker and others, 2010). Previous studies have highlighted the benefits of improved detection of lameness and prompt treatment (Leach and others, 2012; Groenevelt and others, 2014) with research now looking at identifying subclinical lesions before cows are identified lame by conventional mobility scoring techniques (Nikkhah and others, 2005; Wilhelm and others, 2015).

Lameness and welfare

Lameness is painful, and causes cows to change their behaviour, posture and gait in attempts to reduce their level of discomfort (Whay, 2002.) Lamé cows spend a larger proportion of their time lying down (Singh and others, 1993; Walker and others, 2007) and a smaller proportion of their time socialising and interacting with their environment (Walker and others, 2007). Lamé cows spend more time standing in cubicle stalls (Cook and others, 2007), and lying out of cubicles (Galindo and Broom, 2002) and at milking time enter the parlour later (Hassall and others, 1993) meaning they are spending more time standing in the collecting pen. The proportion of time spent feeding may be the same (Singh and others, 1993) or reduced (Galindo and Broom, 2002) in lamé cows.

Lamé cows display a heightened sensitivity to pain (hyperalgesia) (Whay and others, 1997; Whay and others, 1998) that is positively associated with mobility score (Tadich and others, 2013). Cows that are lamé because of sole ulcers and white line disease may display hyperalgesia for 28 days after treatment (Whay and others, 1998), highlighting the extended period of pain cows experience with certain

lameness lesions. Lamé cows may also perceive normally innocuous stimuli as painful (Ley and other, 1996)

Economic and production losses caused by lameness

Lameness is the second most costly production disease/health problem in English dairy herds (Kossaibati and others, 1997), with an initial case of lameness estimated to cost around £323.47 (Willshire and Bell, 2009).

Lameness increases the risk of culling throughout lactation (Rajala-Schultz and others Part II,III, 1999; Booth and others, 2004). This is, however, confounded by milk yield and reproductive status, which means that often the culling of lame cows is delayed until the end of lactation when milk yield has declined (Rajala-Schultz and others Part II, III, 1999), which represents an important potential welfare problem.

Lameness more commonly affects high yielding older cows (Green and others, 2002; Archer and others, 2010) and causes a mean (range) reduction of 357kg (160 to 550kg) to potential 305 day lactation yield (Green and others, 2002). The magnitude of milk yield reduction depends on the severity and duration of lameness, the stage of lactation when it occurs and the type of lesion (Green and others, 2002; Amory and others, 2008; Archer and others, 2010; Reader and others, 2011).

Lame cows are less likely to ovulate and ovulate lower oestrogenic follicles, have less intense oestrus behaviour and have lower luteal phase milk progesterone concentrations, compared to non-lame cows (Morris and others, 2011). Mild and severe lameness extends the calving to first insemination by four and six days and the calving to conception interval by 18 and 16 days respectively compared to non-lame cows (Orgel and others, 2016) which is in agreement with Garbarino and others (2004) and Collick and others (1989).

Claw horn disruption lesions

Claw horn disruption lesions constitute a subset of non-infectious lesions that includes sole haemorrhage, sole ulcer and white line disease (Newsome and others, 2016).

Sole ulcer is considered to be the most important cause of lameness in dairy cows and is responsible for 28% of lameness events (Murray and others, 1996; Van Amstel and Shearer, 2006). Sole haemorrhage causes 8% of lameness events (Murray and others, 1996) and when affecting $\geq 50\%$ of the sole or a smaller area with deep intense colour increases the risk of lameness (Odds Ratio 2.35) (Manske and others, 2002). With the exception of Webster (2001), studies have failed to demonstrate a positive association between sole and white line haemorrhage and mobility score (Offer and others, 2000; Tadich and others, 2010; Wilhelm and others, 2015).

Claw horn disruption lesions frequently occur together at similar stages of lactation (Manske and others, 2002; Offer and others, 2000). Positive associations exist, at the claw level, between white line haemorrhage and sole haemorrhage, and sole haemorrhage and sole ulcer (Manske and others, 2002). Haemorrhagic lesions of the sole reach peak severity at nine to sixteen weeks post calving (Leach and others, 1997; Offer and others, 2000) which is when sole ulcers are most commonly diagnosed (Logue and others, 2004). Such associations suggest that CHDL's may share the same causative mechanisms and represent different developmental stages of the same disease process. In fact when many series of cohorts of cattle are examined it is possible to follow the increasing severity of sole contusions until they give rise to a discrete sole ulcer (Leach and others, 1997; Offer and others, 2000; Le Fevre and others, 2003).

The aetiopathogenesis of CHDLs

The aetiopathogenesis of CHDLs is multifactorial and still poorly understood (Bicalho and others, 2009). It is well documented that CHDL's most frequently affect the lateral hind claws (Le Fevre and others, 2001; Wilhelm and others, 2014), which is consistent with the pressure distribution on claws as described by Van der Tol and others (2002). It is now widely accepted that most CHDL's originate from contusions within the claw horn capsule (Logue and others, 2004; Newsome and others, 2016). Prolonged compression of the corium leads to capillary damage, haemorrhage, thrombosis, and cellular inflammatory reaction (Ossent and Lischer, 1998). Prolonged compression of the corium beneath the axial aspect of the flexor

tuberosity can cause haemorrhage and ulcers in the “typical sole ulcer site”, and haemorrhage within the white line may be caused by prolonged compression of the corium beneath the abaxial margins of the third phalanx (Ossent and Lischer, 1998).

The third phalanx is suspended from the abaxial hoof wall by laminar attachments and on its axial aspect receives a smaller area of support from fibre bundles of the distal interdigital cruciate ligaments (Lischer and others, 2002). This asymmetrical suspension of the third phalanx causes it to tilt in an axial direction when under load (Lischer and others, 2002). Physiological and metabolic events around the time of calving cause mechanical weakening of the abaxial hoof suspensory apparatus (Tarlton and others, 2002), which may increase the mobility of the third phalanx within the hoof capsule (Lischer and others, 2002).

The digital cushion has the important role of dissipating the concussive forces transferred through the caudal aspect of the third phalanx onto the underlying corium, during foot strike and loading (Raber and others, 2006), and its thickness is highly negatively associated with the prevalence of sole ulcers and white line disease, and is a strong predictor of lameness (Bicalho and others, 2009). The thickness of the digital cushion (DCT) is positively correlated with body condition score and it decreases steadily during the first 120 days after calving (Bicalho and other, 2009). The middle and abaxial pads of the digital cushion contain significantly less Arachidonic acid, which may be the result of its utilisation for prostaglandin synthesis during episodes of active inflammation in the underlying corium (Raber and others, 2006). Utilisation of fatty acids from parts of the digital cushion may impede its future ability to dissipate forces which may instigate a self-perpetuating cycle: increased forces acting on the corium, additional inflammation and further utilisation of fatty acids and diminished cushioning capacity of the digital cushion, etc (Newsome and others, 2016). A recent randomised control trial testing four treatments for CHDL's has provided further evidence that inflammation is likely to play an important role in the aetiopathogenesis of CHDL's (Thomas and others, 2015). This study found that administering a non-steroidal anti-inflammatory treatment in addition to a therapeutic trim and applying a block to the non-lame claw produced the best recovery rates of new cases of lameness due to CHDL's. Both reducing inflammation and weight bearing on the sole appear to be important in the healing process of CHDL's.

A recent retrospective cohort study found an association between the presence of bone proliferation on the caudal aspect of the third phalanx and the previous lifetime occurrence of CHDL's (Newsome and others, 2016). The authors believe that such bone proliferation may be caused by inflammation in the surrounding tissues, associated with active CHDL's and/or by mechanical trauma to the periosteum, arising due to the suboptimal dissipation of load forces by the digital cushion. They further believe that these disease processes may form part of another self-perpetuating cycle that predisposes the cow to a higher risk of future lameness: local inflammation/direct trauma to the periosteum, causing bone proliferation giving rise to greater point-forces acting on the corium, additional inflammation and further stimulus for bone proliferation. Bone proliferation also often develops on the abaxial aspect of the third phalanx (Tsuka and others, 2012) which may have a causative role for inflammation and haemorrhage within the laminar corium which becomes visible in the white line as it grows out, becoming a risk area for separation, impaction and infection (white line disease) (Newsome and others, 2016).

Lameness detection

Recent work suggests that early diagnosis and prompt treatment of lameness is a key aspect of managing the disease (Reader and others, 2011; Leach and others, 2012; Groenevelt and others, 2014).

Reader and others (2011) found that the chance of recovery from lameness was negatively associated with the duration of lameness; cows that had been lame for less than two weeks and two to four weeks had greater rates of recovery compared to cows that had been lame for 18 weeks (Odds Ratio 3.63 and 2.51, respectively). A possible reason for this might be explained by some of the findings of Newsome and others (2016) who reported that cows that had experienced more lameness in the 12 month period prior to slaughter had greater bone proliferation on the third phalanx.

The merits of fortnightly mobility scoring and prompt treatment over conventional farm practices of lameness control are well described (Leach and others, 2012; Groenevelt and others, 2014). These studies found that the time to lameness treatment was reduced by 38-65 days, which resulted in the treatment of less severe

foot lesions, which had higher cure rates. Both studies identified that conventional farm practices, unfortunately, failed to identify many cases of lameness.

Mobility scoring is quick, inexpensive and easy to perform. However, it can become time consuming when used to score the whole herd, and it is a subjective assessment that frequently results in discrepancies between observers (Whay and others, 2003). When farmers and scientific researchers assessed the same cows with a widely accepted mobility scoring system the farmers' estimated prevalence of lameness was close to the researchers estimated prevalence of severely lame cows (Leach and others, 2010). Reasons for this may include desensitization and habituation of farmers when lameness surrounds them, and under-recognition of the importance of mild cases of lameness (Whay, 2002). Given that cattle are prey animals it is likely that the display of pain associated with claw lesions is suppressed until a threshold is reached, and thus detection using mobility scoring systems may be delayed (O'Callaghan and others, 2003). This theory is supported by the findings of Reader and others (2011) who reported that reductions in milk yield could be recorded for a six to eight week period before lameness was diagnosed by fortnightly mobility scoring, indicating that some lameness lesions adversely affect production, with an initial level of discomfort that does not cause detectable changes in gait.

Since the 1980's various technologies have been investigated for their potential to be used as automated lameness detection systems, with the aim of providing regular, quickly available, accurate, objective measurements to help dairy farmers manage lameness in their herds (Van Nuffel and others, 2015).

Infrared Thermography and Infrared Thermometry (ITT).

Infrared Thermography is a non-invasive diagnostic technique that has been used widely in human and veterinary medicine to detect changes in surface temperature, which is associated with inflammation. The thermographic images are a pictorial representation of the heat gradients generated by an object, with variations in heat represented by different colours (Eddy and others, 2001). Infrared thermometry is a similar non-invasive technique. Thermometers often have an "automatic data hold facility" which displays and saves the maximum temperature recorded from the

surface of an object, and have the advantage of being less expensive than thermal imaging cameras (Main and others, 2012).

Infrared Thermography and ITT have both been used to investigate the association between foot temperature and the occurrence of lameness and the presence of foot lesions, in numerous studies within the last 12 years (Nikkah and others, 2005; Alsaad and Buscher, 2012; Main and others, 2012; Wilhelm and others, 2015; Wood and other, 2015).

Foot temperature is positively associated with mobility score, with sole temperature of feet from severely lame cows being 2.54°C higher than feet from sound cows, after adjusting for ambient temperature (Oikonomou and others, 2014).

In a longitudinal study where foot temperature was measured alongside routine fortnightly mobility scoring, adjusted foot temperature was found to be highest at the point of lameness detection, which was also significantly higher than foot temperature recorded both 2 weeks before and after lameness detection (Wood and others, 2015).

Nikkah and others (2005) found that cows that were less than 200 days into lactation had an increased temperature at the coronary band and a greater incidence of sole haemorrhage than cows that were more than 200 days into lactation. The study also found that sole haemorrhage occurred more frequently in lateral than medial claws, and that the temperature of the coronary band was higher over the lateral claw compared to medial claw, relative to the temperature of a control area of skin higher up the leg. From these results the authors suggested that skin temperature measurement using infra-red thermography might be able to detect inflammation associated with sole haemorrhage in early/mid lactation cows, and recommended further investigation of the relationship between foot temperature and hoof lesions.

Alsaad and Buscher (2012) used infra-red thermography to record the maximum temperature of the coronary band and found that digital dermatitis, on its own, and a group of other lesions (sole haemorrhage, white line disease, sole ulcer, heel-horn erosion, double sole and interdigital growths) caused feet to have a significantly higher temperature than feet which had no lesions. Stokes and others (2012) made very similar findings, with temperature differences existing irrespective of whether

the feet were dirty, cleaned or lifted in a crush, (maximum temperature measured over the plantar aspect of the pastern using infra-red thermography). The fact that temperature did not differ significantly between feet with digital dermatitis and the other skin or CHDL's was taken to indicate that infra-red thermography was not sensitive enough for lesion specific detection.

Main and others (2012) and Wood and others (2015) used ITT to record the maximum temperature over the plantar area of the pastern, and both studies provided further confirmation of the temperature difference that exists between feet with and without lesions. Sole ulcer, sand crack and digital dermatitis caused increases in foot temperature (not adjusted for ambient temperature) of (2.51, 2.06 and 2.16°C respectively), compared to feet with no lesions, (Main and others, 2012). Using multilevel analysis Wood and others (2015) predicted that the presence of a lesion increased foot temperature by 0.62°C at the average ambient temperature of the study and for every 1°C increase in ambient temperature foot temperature was predicted to rise by 0.276°C. Differences in mean foot temperature could not be detected between specific lesion types, however, further categorisation of lesions highlighted groups of lesions such as claw horn lesions (sole haemorrhage, sole ulcer, sole separation, white line disease, sole puncture, impacted stones) which tended to have a higher foot temperature than feet without lesions (P=0.06).

After adjusting for the effects of mobility score and ambient temperature Oikonomou and others (2014) demonstrated that the sole temperature of lateral hind claws, measured at the typical sole ulcer site, was correlated with the thickness of the digital cushion (as measured by ultrasound); sole temperature was 2.96°C higher for claws in the first quartile compared to the fourth quartile of digital cushion thickness. The authors postulated that this association may be the result of the likely poorer cushioning capacity of thinner digital cushions leading to greater contusion and inflammation of the corium beneath the third phalanx.

Wilhelm and others (2015) investigated the use of infra-red thermography of the sole as a possible diagnostic technique for the early detection of subclinical laminitis. The weight bearing surface of claws was examined during the first week and at the end of the eighth week after calving, which involved an assessment of haemorrhage and

measurement of temperature using infra-red thermography. An interval of approximately two months between examinations was used because it is recognised that blood, from damaged blood vessels within the corium, which is incorporated into newly formed horn, takes approximately two months before it becomes visible as haemorrhages at the trimmed weight bearing surface of the claw (Nocek, 1997). The results of the claw-score analysis showed a deterioration in claw health occurred during the first two months of lactation, with heifers experiencing a larger increase in claw score than cows. At the second examination a significant difference was found to exist in the claw score and temperature of the lateral compared to the medial hind claws; lateral claws having higher claw scores and being warmer. However, no clear correlation could be demonstrated between claw temperature after calving and the claw lesion score recorded eight weeks later. The authors put forward several possible reasons for why no such correlation was found. Firstly they outlined that it is not easy to objectively score claw haemorrhages and indicated that the scoring system they used may have affected their results. The scoring system in the study classified slight haemorrhage of the sole as four times worse than slight haemorrhage of the white line. This system may therefore over-estimate the severity of haemorrhages of the sole and might not accurately represent the lesions present on the claw. The authors also postulated that the increase in temperature caused by inflammation is not strong enough to heat the claw horn measurably, and that maybe the temperature of the ground contact area of the claw is not suitable to draw conclusions on the inflammation status of the corium, with the technique used in their study. It is also suggested infra-red thermography was possibly performed too early in the disease process and that additional examination times, may have yielded different results.

Summary

Claw horn disruption lesions frequently occur together at similar stages of lactation. They may share some of the same causative mechanisms and in some instances may represent different developmental stages of the same disease process. The more severe lesions such as sole ulcer and white line disease are important contributors to lameness in dairy cows. The aetiopathogenesis of CHDL's is complex. Systemic factors such as weakening of the suspensory apparatus after

calving, thickness of the digital cushion and bone proliferation on the third phalanx have important roles as do external mechanical forces applied to the foot associated with standing times and walking surface etc. The end result is that inappropriate forces act on the corium causing inflammation, which allows blood to leak into newly formed horn that may be of inferior quality. It takes approximately two months for such newly formed horn to reach the ground contact area of the hoof, and if lesions are severe a lameness event may occur after a further variable period of time. The ability to be able to measure the “health status” of the corium within the foot would provide huge advantages in the early identification of feet that were likely to develop severe CHDL’s and possible lameness events.

Future studies should look to further investigate the relationship between foot temperature and the presence of CHDL’s with the aim of establishing whether repeated examination using ITT is a diagnostic technique that could be used to accurately detect inflammation of the corium and the likely development of biologically significant CHDL’s.