AETIO-PATHOGENESIS OF ANAEROBIC INFECTIONS ASSOCIATED WITH BOVINE LAMENESS AND SOME HUMAN DISEASES

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Introduction

In clinical veterinary practice, infectious lameness has been described in textbooks from the early 1800s. Known colloquially as 'the fouls', specific lesions were accurately described: the shag foul was a circular lesion 2 cm diameter on the heel similar to greasy heels in horses and sounds similar to digital dermatitis; the stinking foul was an interdigital necrosis that was, almost certainly, interdigital necrobacillosis; the frog foul was a space-occupying lesion within the interdigital space but without any damage to the integument, the condition we recognise now as interdigital hyperplasia (Knowlson 1819). During the 1930s, infectious causes of lameness in both cattle and sheep were considered important only with regard to diagnosis of Foot-and-Mouth disease (Woolridge 1934), not for any economic or welfare reasons.
Today, cattle lameness is considered a major welfare issue in many countries where intensive dairy farming is commonplace. Surprisingly it is two infectious diseases associated with lameness, digital dermatitis in cattle and new variant footrot in sheep, which are causing currently the greatest problem for livestock farmers. The aetiopathology of both is unclear and hence control measures are largely ineffective (Demirkan et al 1999). Treatment and control is expensive in terms of time and labour, and there is concern that disposal of footbath solutions used for treatments may pollute the environment.

Aetiology

The list of microorganisms associated with infectious lameness in cattle, mostly facultative or obligate anaerobes, is large and expanding slowly. The most common diseases and the infectious agents associated with them are listed in Table 1, together with suitable references. Until recently, it was not possible to isolate and grow Treponeme spp in pure culture and much of the evidence of their association with digital dermatitis is based currently on serology. Otherwise, all other microorganisms have been isolated from lesions and identified by standard microbiological techniques.

Risk factors for digital skin infections

Gross anatomy of the interdigital space

The vertical distance between the ground and the cranial and caudal borders of the interdigital space varies consistently in adult cattle (see Table 2). These results suggest that the caudal aspect of the interdigital space is always closer to the ground and adjacent hairy skin is vulnerable to repeated and continuous contact with slurry, especially in housed cattle. This is the case more so for hind limb digits than those of fore limbs ,ince many cows stand half-in cubicles with their hind feet in the slurry passage. The continuous contact of the hairy skin of the heel of hind limbs with slurry may cause focal hydropic maceration of the epidermis at this site and predispose it to bacterial invasion.

Microtrauma to skin

The integument of the bovine digit is designed to form an effective barrier between the environment that surrounds it and the functional tissue within it. Thus, a keratinised hard horny capsule encases the 3rd phalanx and distal interphalangeal joint of all claws. The skin of the interdigital space between each claw is also modified. Sweat glands and hair follicles are absent and the stratum corneum of the epidermis comprises of anuclear stratified squamous keratinised cells arranged in layers up to 35 cells thick. This is a very substantial flexible protective barrier to external insults. Microorganisms can penetrate such a structure only following mechanical trauma that allows opportunistic pathogens access to adhere to the deeper layers of the epidermis. Only then can keratolytic enzymes produced by pathogens such as D. nodosus, or exotoxins produced by F. necrophorum, break down the epidermal barrier to expose sensitive and susceptible underlying dermal tissue to inflammatory changes or invasion by other opportunistic pathogens.
The hairy skin adjacent to the coronary band and interdigital space is not modified from that found elsewhere. It remains an effective mechanical barrier. However, the integrity of the epidermis is affected adversely by exposure to prolonged moisture and reduced access to air. The resultant hydropic maceration may predispose skin to infection by Treponeme spp to produce clinical lesions typical of digital dermatitis (Read and Walker 1998).

Biology of anaerobic pathogens associated with human disease

Human periodontal disease has many similarities with infectious lameness in cattle; the bacterial profile, a stratified epithelium that is invaded, a 'triggering event' that precipitates epithelial and connective tissue necrosis, a susceptible host. Syphilis, like digital dermatitis, has been associated with a Treponeme infection. In periodontal disease, the focus of research has been to understand the interactions between various microorganisms isolated from diseased gingiva; with syphilis, it is the biology of Treponemes associated with their pathogenicity.
Some of these research outcomes are described below:
Attachment and colonisation of epithelial surfaces
The pathogenic Treponeme pallidum attaches to its host cells and invades epithelium whilst non-pathogenic Treponeme spp do not have this ability. This adherence factor appears to contribute to prolonged bacterial sur-

vival, motility and virulence. Cell surface structures, known as adhesins, interact with extracellular matrix molecules (EMMs) found on the host cell surface such as firbronectin, collagens and laminins. Bacterial adhesins have been found on T. denticola, a near-relative to the Treponeme spp isolated from clinical cases of digital dermatitis and new variant footrot, and exhibit specific attachment to laminins (Cameron 2003). The pathogenic Porphyromonas gingivalis has its own adherence factors that bind with host cell EMMs on the gingival epithelium (Agnani et al 2003).

Role of matrix metalloproteinases (MMPs)

MMPs are a gene family of zinc metalloenzymes associated with degradation of extra-cellular elements of host cells such as collagens and proteoglycans. There are three classes, defined according to their substrate specificity: collagenases degrade collagen produced by fibroblasts and monocyte/macrophages; gelatinises degrade denatured collagen and elastin; stromelysins have a wide specificity including proteoglycans, laminin, fibronectin and IX collagens. MMP activity is counterbalanced by host tissue inhibitors (TIMP-1). Any disease condition that upregulates MMP expression but not TIMP-1 increases matrix turnover (Nicod and Dayer 1999). Recently, Choi et al (2003) found that T. denticola can upregulate MMP expression, especially gelatinase.

Environmental influence on microbiological function

A study of pathogenic spirochaetes has produced evidence that the bacterial genome is expressed differentially according to the microenvironment that surrounds it. For example, temperature, serum deprivation and the mammalian environment may all affect synthesis of Borrelia burgdorferi proteins associated with cell regulation or signalling (Roberts et al 2002). Another example can be found related to protease production by the human colonic bacteroide Bacteroides fragilis. Its protease formation, essential for bacterial growth, is stimulated in the presence of excess carbohydrate and strongly inhibited by high peptide concentrations in the gut (MacFarlane et al 1992).

Microbial coaggregation and synergy

Examples of this exist within the microbiological profile of the healthy human intestine. Members of the genus Bacteroides ferment soluble polysaccharides such as pectin and starch; other anaerobes such as Bifidobacterum spp. and P.asacchrolytica break down insoluble complex polysaccharides like mucin. Another example relates to the anaerobe P. melaninogenicus. If this organism is added to an avirulent mixture of human oral bacteria this complex becomes uniformly infective and causes severe abscess formation in experimental animals (Socransky and Gibbons 1965). A similar synergy exists when contaminant anaerobes such as F. necrophorum and Arcanobacterium pyogenes are present in the bovine uterus post-partum, causing a severe endometritis that may result in significantly lowered fertility of affected cows.

Periodontal disease begins with the adherence of certain bacteria to the tooth enamel surface coated with salivary glycoproteins. The Gram +ve Streptococcus sanguis adheres to enamel initially. Then, bacterial coaggregation occurs which involves cell-associated lectins or fimbriae present on its cell surface. Other bacteria such as Actinomyces viscosus possess receptors that can bind specifically to these fimbriae and the bacterial profile at the gingival surface builds up to produce an initial inflammatory response. Microbial coaggregation can be inhibited by glycoproteins present in saliva (Cisar 1982).

Host humoral immune response to anaerobes

The specific activity of bovine IgG2 against spirochaetes associated with clinical digital dermatitis lesions is similar to that found for diverse strains of Gram -ve anaerobic bacteria isolated from subgingival plaque in humans. The conclusion could be that these bacteria are associated immunologically with development of their respective lesions. This humoral response could be significant in several ways:

a) IgG can bind with polymorphonuclear inflammatory cells (PMNs) to release lysosomal enzymes, thereby disrupting basement membranes and allowing microorganisms to penetrate disrupted epithelium;
b) compliment-activating antigen-antibody complexes may inhibit phagocytic activity of PMNs
c) sensitised lymphocytes release lymphokines which inhibit macrophage migration. The degree of tissue necrosis associated with different lesions may be a reflection of both the microbial profile associated with disease but also the variation in the host's humoral immune status.

Conclusions

The parallels between infectious lameness in ruminants and a variety of human diseases associated with anaerobic Gram -ve organisms are striking. Not only are the microorganism profiles similar but the humoral antibody responses comparable for healthy and diseased patients. Such comparative studies could suggest the direction that future research should take: the significance of different microbial profiles associated with lesions; host-parasite interactions and the role of humoral antibodies; virulence factors present in Treponeme spp. isolated from different sites within an animal and compared with those found in lesions.

Table 1. Microorganisms associated with infectious causes of lameness in cattle

Disease	Alternative name	Microorganisms isolated	Reference
Digital dermatitis	Mortellaro disease	Bacteroides spp	Roztocil et al (1988)
	Digital papillomatosis	Campylobacter faecalis	Sabo et al (1988)
	Papillomatous digital	Clostridium spp	Koniarova et al (1993)
	dermatitis	Fusobacterium spp.	Dopfer et al (1997)
	Strawberry footrot	Peptococcus asaccharolyticus	Choi et al (1997)
	Raspberry heel	P. saccharolyticus	Wallis et al (1997)
	Hairy warts	Peptostreptococc us spp	Demirkan et al (1999)
	Hairy footwarts	Serpens spp	Schroeder et al (2003)
		Treponema spp	
Interdigital necrobacillosis	Interdigital phlegmon	Bacteroides thetaiotaomicron	Flint and Jensen (1951)
	Infectious pododermatitis	Fusobacterium necrophorum	Hartwigk (1973)
	Foul-in-foot	Prevotella melaninogenicus	Berg and Loan (1975)
	Clit-ill	Porphyromonas asaccharolytica	Berg et al (1994)
	Hoof-rot	P. levii	Morck et al (1998)
	Panaritum		
Peracute interdigital necrobacillosis	Super foul	F. necrophorum	Cook and Culter (1995)
		P. melaninogeni-cus	
		Dichelobacter fragilis	
Interdigital dermatitis	-	Arcanobacterium pyogenes	Egerton and Parsonon (1966)
		Dichelobacter nodosus	Berg (1981)
		F. necrophorum	Masalski (1986)

Table 2. Vertical height (cm) between ground surface and cranial and caudal borders of the interdigital space in 106 front and 123 hind feet of adult cattle

Limb	Site	Perpendicular height (cm) between ground surface and interdigital space				
		Mean	Minimum	Maximum	t value	p
Front	Cranial	6.5	5.0	8.4	-47.6	0.001
	Caudal	3.6	2.3	5.4		
Hind	Cranial	6.5	5.4	7.7	-59.9	0.001
	Caudal	3.0	1.9	5.6		

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IN VITRO INFECTION OF BOVINE EPIDERMAL CELLS AND BOVINE SKIN EXPLANTS WITH TREPONEMES

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Introduction

Dermatitis digitalis (DD) is a multifactorial disease of the bovine digit. DD is an important welfare issue and it has a large economic impact. The mechanisms of its pathogenesis are still not completely understood. In addition to typical anaerobic bacteria, a variety of different Treponema phylotypes have been detected as probably involved in the pathogenesis of DD (Choi et al, 1997; Moter et al, 1998). There is a strong need for models enabling studies of virulence factors of DD (Edwards et al, 2003a). The aim of this work was to establish an in vitro model to study the role of Treponema spp. in the pathogenesis of DD, because a suitable in vitro model was not available until now. This paper reports on the successful in vitro infection of cultured epidermal cells and skin explants from the bovine digit with Treponema denticola and Treponema brennaborense.

Material and Methods

Bovine keratinocytes were isolated from the skin at the typical site of DD lesions and from the bulbar region of the claw. Cells were cultivated under standard conditions as described previously (Nebel and Mülling, 2002). Skin explants were obtained from three different sites of the bovine digit. One sample was taken from the dorsal aspect of the digit 5 cm above the claw capsule. Two samples were taken from the plantar aspect, one from the pastern region and the other from the skin just above the claw capsule. All explants were maintained in Dulbecco´s modified Eagle´s medium (Biochrom, Berlin) supplemented with 10 % fetal bovine serum (Sigma, Taufkirchen) and Kanamycin (Biochrom, Berlin) for 12 h to eliminate bacteria present on the surface of the digit. Subsequently, the explants were rinsed thoroughly with Dulbecco´s modified Eagle´s medium without Kanamycin and incubated with Treponema suspension (approximately 5 x 106 treponemes / ml) for 24 h. In all experiments a pair of explants was incubated with T. denticola or T. brennaborense and the incubations were carried out simultaneously. After incubation with the Treponema suspension the explants were fixed in Karnovsky´s solution and embedded in Epon®. Sections were prepared and examined by light and electron microscop. Keratinocytes were cultivated on cover glasses in a mixture of Dulbecco´s modified

Eagle´s medium supplemented with 10 % fetal bovine serum without antibiotics. These keratinocytes were split in two groups and then incubated with OMIZ Pat medium containing either T. denticola or T. brennaborense for up to 96 h. Every 24 h two cover glasses were rinsed twice with phosphate buffered saline and then fixed in 4 % formalin immediately. For visualisation the cover glasses were stained with the DAPI method (1μg/ml) and examined under the fluorescent microscope.

Results

Based on morphological criteria T. brennaborense and T. denticola remained alive under culture conditions for up to 96 h. Most treponemes were still showing their typical spiral shaped morphology after that incubation time. The adhesion of the treponemes to the cultured keratinocytes (Fig.1) was visible at all time points and tended to show an amplified adhesion to the keratinocytes in a time dependent manner. Especially adhesion of T. brennaborense was greater than that of T. denticola. With prolonged incubation cultured cells began to show morphologic damage and some cells detached from the cover glasses.

Investigations of the skin explants at light and electron microscopical level revealed that treponemes were detectable adhering to the surface and between the cells of the outer regions of the epidermis. Furthermore, in some explants treponemes could be detected in deep epidermal layers (Fig.2) by light microscopic examination of semithin sections stained with methylene blue. In addition electron microscopy clearly demonstrated that the treponemes invaded the epidermis via the intercellular spaces of stratum corneum and stratum spinosum.

Fig.1: Adhesion of treponemes (arrow) to keratinocytes in vitro

Fig.2: Semithin section of an infected skin explant showing treponemes (arrows) within the intercellular space in deeper layers of stratum spinosum

Discussion

Treponemes survived in cell culture under standard conditions (5 % CO2) for up to 96 h, even though all species have been described as anaerobic, although some may be considered to be microaerophilic. Adhesion of T. denticola to human gingival fibroblasts could be shown under both aerobic and anaerobic conditions (Chan and McLaughlin, 2000).

Several recent reports have implicated spirochetes in the aetiology of bovine DD (Collighan and Woodward, 1997; Moter et al, 1998; Stamm et al, 2002), a major cause of lameness in dairy cattle. Spirochetes of the genus Treponema are among the most predominant organisms visible in DD lesions (Döpfer et al, 1997). Treponemes have been identified in biopsies from DD lesions, they can be found in the stratum spinosum and dermal papillae. Their presence deep in epidermal tissue suggests they are invasive (Choi et al,1997; Moter et al, 1998) which is supported by our in vitro findings. Furthermore, serum samples from diseased animals contain elevated antibody levels to Treponema antigens (Demirkan et al, 1999).

T. denticola has been shown to adhere to basement membrane proteins such as laminin, fibronectin and type IV collagen as well as type I collagen, gelantin and fibrinogen. The binding to laminin was most prominent (Chan and McLaughlin,2000). Edwards et al (2003b) noted that T. denticola cells bound also to keratin.

Fenno and McBride (1998) reviewed cytopathic effects of oral spirochetes. In various in vitro studies of these authors, exposure of epithelial cells to T. denticola caused: visible morphological damage, cell detachment from monolayers and inhibition of proliferation, membrane blebbing, loss of tight intercellular contact and cytoskeletal rearrangement. Our results obtained from the in vitro studies with keratinocytes and explants support these observations. After incubation with treponemes over 96 h keratinocytes showed visible morphological damage and began to loosen from the cover glasses. In further studies with epidermal keratinocytes growing on sliceable membranes we will look for structural damage of the cells and their components by electron microscopy.

In periodontal disease, spirochetes have been found between cells in the junctional epithelium which are normally tightly joined. There is evidence that oral spirochetes invade tissue by migrating through the intercellular tight junctions. Spirochetes were also observed in enlarged intercellular spaces (Fenno and McBride, 1998). Our results support these findings and suggest that treponemes invade the claw tissue via the intercellular spaces of the epidermis. We suggest that the enlargement of the intercellular spaces can permit an increased infection of deeper tissue layers and facilitate the way for infection with other anaerobic bacteria.

The innate immune response is largely blamed for the significant amount of the damage caused bythe release of inflammatory mediators and cytokines. Interactions of vital T. denticola resp. T. pectinovorum with human gingival fibroblasts results in increased secretion of proinflammatory chemokines such as IL-1β, IL-6, Il-8, IL-1 and IFN-γ (Nixon et al, 2000). Therefore in a future project we will study differences of the chemokine pattern expressed by the cultured bovine keratinocytes either infected with treponemes or cultivated without treponemes.

So far, our results provide good evidence that the cell culture and explant models introduced in this study will be helpful for further investigations on the pathogenesis of DD and particularly on the mechanisms of tissue invasion. Using these models the molecular mechanisms of adhesion and tissue invasion as well as the tissue response to the infection will be studied.

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IMPACT OF HOUSING TYPE ON PREVALENCE AND SEVERITY OF DIGITAL DERMATITIS

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Introduction

Digital dermatitis is a skin condition of cattle, which usually affects the skin on the bulbs of the heel or between the digits. The lesion causes considerable pain to the cow, resulting in reduced mobility and feeding, with consequent loss of milk production. The condition can persist for months if it is untreated (Peterse 1992). This study was designed to determine the factors affecting the prevalence and severity of digital dermatitis in dairy cows, by appraising the role of the environment in its development.

Materials and Methods

This study was an observational single site study, using the ADAS Bridgets dairy herd, which had 650 Holsteins and endemic digital dermatitis. During winter housing, the hind limbs of all cattle in each of four different houses (a straw yard, two automatically-scraped cubicle yards and one tractor scraped cubicle yard) were examined every three weeks, using a modified Olympus Series 5 borescope, for digital dermatitis. The severity of lesions was scored using a system based on size, depth and colour (Laven and Hunt 2000). Table 1 shows the number of cows examined and the number of lesions observed.

Table 1: Number of cows examined and lesions observed in each of the four housing groups

	Number of cows examined	Number of lesions observed
Tractor-scraped cubicles	183	747
Automatically-scraped cubicles (1)	108	472
Automatically-scraped cubicles (2)	138	454
Straw yard	61	181
Total	490	1854

Results

The prevalence of digital dermatitis in each house is shown for a representative twelve week period in Figure 1, while Table 2 shows the overall mean prevalence for each group (which includes the data collected for longer than the twelve week period shown in Figure 1).

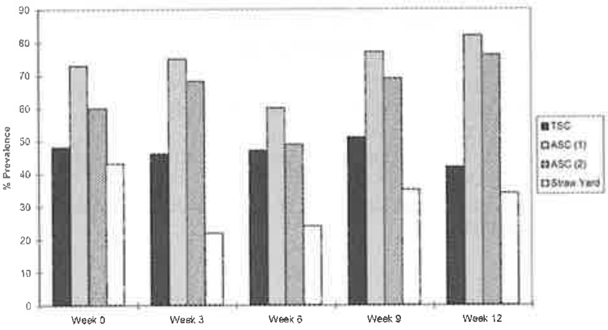

Figure 1: Effect of housing type on the prevalence of digital dermatitis
TSC = Tractor-scraped cubicle. ASC = automatically-scraped cubicles

Table 2: Overall mean prevalence and cumulative prevalence of digital dermatitis

	Mean Prevalence (%)	Cumulative Prevalence (%)
Tractor-scraped	47	67
Automatically-scraped (1)	73	82
Automatically-scraped (2)	64	78
Straw yard	32	

Housing significantly affected the mean and cumulative prevalence of digital dermatitis. Cows on straw yards had a significantly lower prevalence of digital dermatitis than cows in any of the three cubicle yards ($P<0.001$). Cows on the tractor-scraped yard had a significantly lower prevalence of digital dermatitis than cows on automatically scraped cubicles ($P<0.001$). Comparison of the two automatically-scraped cubicles also found a significant difference in prevalence of digital dermatitis. However, the magnitude of this effect was much less ($P=0.03$). There was a significant change with time ($P<0.05$) for all four groups, however there was no discernible pattern to this change.

Mean lesion score

There was no significant change in time of lesion score ($P>0.05$), so data for each house were amalgamated for further analysis. The mean lesion score for the cows in the straw yards (3.1 0.08) was significantly lower than that of any other group of cows ($P<0.01$), while the mean score for the cows on the tractor-scraped cubicles (3.2 0.03) was significantly lower ($P<0.001$) than that for the cows on automatically-scraped cubicles (3.5 0.06 for both yards)

Discussion

As the cows in this study were housed in pre-existing buildings, it is difficult to isolate risk factors and analyse them separately. Nevertheless this study does identify some significant risk factors for digital dermatitis. The main factors identified by this study are housing type (cubicles vs. straw yards) and scraping mode (automatic vs. tractor). Other environmental factors influence digital dermatitis, but this study suggests that their impact is relatively small. This is exemplified by the difference between the two automatically-scraped yards. One was a wooden kennel structure, the other a modern concrete and metal yard, but the effect of these differences was considerably less than the effect of scraping mode.

The cause of the differences in digital dermatitis between houses has not been conclusively identified by this study, but some suggestions can be made. The most likely reasons for the low prevalence on straw are a reduction in exposure to the digital dermatitis agent, and a cleaning effect of the abrasive straw. However, the feet of the cows on the straw yards were of similar cleanliness to those in the cubicles, thus the reduction in exposure is the most likely cause of reduced prevalence.

The causes of the differences in prevalence because of scraping mode are more difficult to assess. The differences between the systems include cubicle design, stocking density, bedding and scraping mode. All three cubicle houses are standard and meet current recommendations and there is no evidence that the design differences had significant behavioural effects. Stocking density was lower in the automatically-scraped cubicles than the tractor-scraped yard, so it is unlikely that this played a significant role in the development of digital dermatitis. The bedding in both of the automatically-scraped cubicles was shavings and lime while that in the tractor-scraped group was straw and lime. Chopped straw may be less prone to combine with slurry and lime than shavings and thus less prone to adhere to the feet. This hypothesis requires further study. However, no differences were observed in the cleanliness of the feet between the groups of cows, thus the major difference was probably the method of scraping. Automatic scraping should clean more thoroughly, but it can result in more slurry around water troughs and connecting passageways, and the collection of pools of mixed slurry in front of the scraper. Although cows tend to avoid these pools, there are times, such as at milking, when the cows may be moved through the slurry, which thus comes into contact with the skin of the foot.

The severity of digital dermatitis was affected by environment in the same order as prevalence was affected. This suggests that severity of digital dermatitis is affected by the same environmental factors as prevalence. The lack of difference in severity between the cows in the two automatically-scraped cubicle yards further confirms that the effects seen here are associated with scraping mode.

Acknowledgements

The authors would like to thank the Milk Development Council for funding this study at ADAS Bridgets.

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INFLUENCE OF FOOTBATHING ON PREVALENCE OF DIGITAL DERMATITIS AFTER INTRODUCTION OF DISEASED ANIMAL INTO HEALTHY DAIRY HERD

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Abstract

On several intensively managed dairy farms, where disinfectant barriers were regularly used, we discovered that after introducing one cow with digital dermatitis, prevalence of digital dermatitis rose enormously. This rise in the number of cases was significantly higher in herds where disinfectant barriers were regularly used.
Bacteriological analyses of samples showed that in all herds we normally found on the heels and interdigital space of the claw Spherophorum necrophorum and Bacteroides nodosus and often also Clostridium perfringens. In occult clinical cases of digital dermatitis we found also Treponema spp., and in most cases two types of spirochetes.
Prevalence of digital dermatitis after introducing a diseased animal was much higher in the herds where disinfectant barriers were regularly used than in herds without footbathing. Where different organic acids or copper sulphate were used incidence was higher than in cases where formaldehyde as disinfectant was used.
Our conclusion is therefore, that maceration of the digital and interdigital skin must be an important factor in establishing digital dermatitis. Furthermore, we think that all infectious elements are not able alone to provoke outbreak of the disease if prior maceration and micro trauma on the skin above claws did not occur.

Introduction

Digital dermatitis was first described by Cheli and Mortellaro in 1974. Espinasse and al described in the Atlas of claw diseases in ruminants also a papillomatous digital dermatitis as a separate disease.For almost 30 years different researchers and scientists try to find reasons for the clinical picture of digital dermatitis. In 1994 Zemljic showed that an important role in the development of the disease was played by different types of Treponemas.Subsequently, groups of researchers found out that digital dermatitis and papillomatous digital dermatitis are histopathologically and immunohistochemically one and the same disease (Zemljic 1996, Read and Walker 1998), which is an important welfare and economic issue in intensively managed dairy production. In the Orlando lameness symposium Zemljic considered that we must reconsider our attitude to the disease and not to try reproduce the disease only with different micro-organisms which were proved as potential reason of the clinical picture of the disease. Although we recognise that digital dermatitis is a multifactorial disease with microbial, environmental and management influence, we did not take into account other reasons and causes as equally important and tried to attribute to the microbacteriological part of the story the greatest importance. There were different mostly unsuccessful trials to reproduce the disease with implantation of specific micro-organisms on the healthy and even damaged skin above claws. Now there is a great need for production of the models enabling studies of virulence factors of digital dermatitis (Edwards et al, 2003), which also failed to consider the wide range of potential factors. In the present paper we try to describe simultaneous action of different factors in the development of digital dermatitis, which not only produced the full picture of the clinical picture but also affected prevalence and severity of the disease in dairy cattle.

Material and methods

In three intensively managed dairy herds with average 40 dairy cows and production of more than 8000 l per standard lactation they introduced new disinfectant barriers with novel formula. Before this change they did not have any obvious problems with digital dermatitis, with prevalence less than 10% of diseased animals in the herd. In The farm B and C digital dermatitis after introduction of the disinfectant barriers spread and caused severe problems in management and production with evident losses in the cost benefit analysis. In the farm A the situation was not changed significantly.
After unsuccessful implementation of the different therapies, we made epidemiological analysis firstly and after discovering of the cause of the spreading of the disease, we implemented measures which diminished severity of the clinical signs in the two herds.
In all three farms(A,B,C)the disinfectant barrier was introduced at much the same time. In two farms (B,C) they purchased two cows each with clinical signs of digital dermatitis. In farm B the new animals were introduced about a fortnight before introducing disinfectant barrier and in farm C new animals were introduced in the herd just few days after introducing barriers. In farm A no new animals were introduced.
After discovering which animals were causing the disease spread we took samples for bacteriological and pathohistological analysis. After obtaining the laboratory results we introduced antibiotic therapy and new disinfectant for foot barriers. After three weeks we again made clinical investigation in all three herds to determine changes in prevalence of digital dermatitis.

Results

We made clinical examination of all animals in the all three herds and discovered that in herd C where they did not purchase any new animals, only 2 of 42 productive animals had slight changes in the typical sites on the heel of the hind legs above the coronet.We isolated Spherophorum necrophorum and Bacteroides nodosus, but we did not succeed in demonstrating any Treponemas. Clinical signs were mild to moderate with some pain in the diseased areas.

In farm B with 40 animals, where they purchased two animals 14 days before introducing the disinfectant barrier, we found in 22 animals a typical picture of digital dermatitis, mostly with severe and extensive areas of damage on the heel above the coronet of hind legs. Lameness was generally severe,as were the signs of pain. One animal tended to lie down most of the time. Production drop in this case was according to farm data mostly 80%. Microbiological samples showed presence of Spherophorum necrophorum, Bacteroides nodosus and Clostridium perfringens with two types of Treponema . Pathohistological samples confirmed a severe maceration of the skin layers with ulceration and no tendency to heal.

In farm C with 43 animals, where new animals were introduced a few days after putting in a new disinfectant barrier we found 24 animals with severe changes in hind feet with mostly moderate to severe lameness,and where diseased animal tended to lie down as soon as possible. The bacteriological and pathohistological samples results were similar to the second farm.

On farms B and C we advised quarantine of diseased animals.This was done on farm C but not on B. We also introduced antibiotic topical therapy with tetracyclines twice daily. For all animals we introduced disinfectant barriers with formaldehyde in 3% concentration before entering the milking parlour.

After three weeks we again visited all the farms.The situation was cured on farm C where all measures were carefully carried out.. On farm B prescribed measures were introduced partly. Therapy with topical tetracyclines was not carried out consistently because the owner was afraid of presence of residues in the milk. They used a formaldehyde disinfectant barrier before milking parlour, but the concentration was not controlled efficiently, so in some cases the concentration was substantially higher as prescribed. In some cows we found severe damage of the skin above the coronet, not typical for digital dermatitis. In one case we found exungulation of one claw, another animal had been euthanasied, and prevalence of the disease was still very high (25%).

After consideration we recommended reintroduction of a new disinfectant method on farm C but not on farm B where we reapplied recommendations from three weeks previously with strict control every third day. After another three weeks we found only two animals with clinical signs of the disease, pain was not severe and only very slight lameness was observed. After a second therapy period we reintroduced the new system of disinfection.

Discussion

In this clinical trial we proved that introduction of new animals is a substantial factor in spread of digital dermatitis into a herd. But new animals alone were not enough to spread and develop full picture of the disease. In two farms it was shown that even disinfectant barriers could help in disease spread. It is necessary to have "new" microbial strains which had bigger potential for spread in the new surroundings than " old" ones. It is necessary to have the opportunity for skin maceration above the coronet of the heel. Such maceration softens the skin, makes this skin more acceptable for microtrauma and in a such way typical bacteria also have more potential entrance sites. We consider that this micro trauma offers bacteria enough substrate for survival and multiplication. Due to such processes those animals are not able to follow healthy animals in the production process which certainly also causes a reduction of the immunological response of such organisms. This could be a field for future investigation about development of digital dermatitis. Incidentally, we also showed that only strict adherence to necessary control measures could lead to the abolition of the acute problems with digital dermatitis in the Slovenian dairy herds.

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FIELD EVALUATION OF PROPHYLACTIC AND THERAPEUTIC EFFECTS OF A VACCINE AGAINST (PAPILLOMATOUS) DIGITAL DERMATITIS OF DAIRY CATTLE IN TWO CALIFORNIA DAIRIES

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(Papillomatous) Digital Dermatitis ((P)DD) is a superficial, painful, and contagious bacterial disease causing ulcerative or proliferative lesions on the skin usually at the palmar surface of rear feet near the interdigital space and heels. Although (P)DD is multifactorial and the precise etiology has not been determined, there is compelling evidence that invasive spirochetes play a major role either as primary or secondary pathogens in (P)DD lesions. (P)DD is a significant cause of lameness in dairy cattle resulting in financial loss due to premature culling, decreased milk yield, poor reproductive performance, weight loss, milk discard due to treatment with antibiotics, and treatment and labor costs. The development of an efficacious and cost-effective vaccine is highly desirable. The objective of this study was to determine whether Treponema bacterin provides prophylactic and/or therapeutic effects for controlling (P)DD in cattle.

A total of 420 and 740 Holstein cows were enrolled from two, commercial California dairies with pre-vaccination (P)DD prevalence of 27% and 29% respectively. All lactating cows from each herd were vaccinated with either Treponema bacterin (Novartis Animal Health, Inc.) or placebo, where each enrolled cow received 3 doses 3 weeks apart (according to label instructions). Investigators were blinded as to which cows received vaccine or placebo. Study cows were grouped according to their disease status prior to treatment (visible lesion, no visible lesion) and treatment received (vaccine, placebo). ANOVA showed no significant differences ($P > 0.05$) between vaccinated and non-vaccinated cows in pre-trial lactation group, milk production, and days in milk, which demonstrated that bias in selecting groups was unlikely. After completion of vaccination, all cows were visually examined for evidence of (P)DD monthly for 6 months. The diagnostic technique used was observation with a bright-light and water-jet test in the parlor during milking. Hoof trimmer records from the dairies were also examined to determine if cows were found with (P)DD during hoof trimming that might have been missed in the parlor examinations. A sub-sample of vaccine and placebo cows with no visible lesions were bled pre-vaccinating and at each of the observation periods for ELISA testing for Treponema antigens.

A series of z-tests comparing the monthly (P)DD proportions among cows without visible pretreatment lesions found no significant prophylactic effect (p-values ranging from 0.105 to 0.847). Therapeutic effects of Treponema bacterin among cows with visible pretreatment lesions were also non-significant (p-values ranging from 0.118 to 0.940). Effects were also analyzed by lactation groups. No significant prophylactic effects were observed for 1st lactation cows and for 2nd or later lactation cows with p-values ranging from 0.060 to 0.979 and 0.178 to 0.977 respectively. Likewise, no consistent significant therapeutic effects were observed for 1st lactation cows and for 2nd or later lactation cows with p-values ranging from 0.016 to 0.966 and 0.299 to 0.993 respectively. In one of the herds, the proportion of (P)DD was significantly different ($p = 0.016$) for 1st lactation vaccinates than for placebos during month 1. In this case, however, the difference was due to a significantly higher proportion of (P)DD observed in vaccinates than in placebos.

Data were further analyzed by lactation groups for only those cows present every month. Although no consistent significant effects were observed for cows with no visible lesions for all lactation cows, the (P)DD proportion on 1st lactation vaccinates was significantly lower than placebos. On the other hand, no consistent significant therapeutic effects (cows with visible lesions) were observed for all lactation cows, 1st lactation cows and for 2nd or later lactation cows. A significant difference for the 1st lactation cows was observed but was due to a significantly higher proportion of (P)DD observed on vaccinates than on placebos.

ELISA for serum antibodies to (P)DD-associated Treponema spp. revealed that approximately half of the cows with no visible (P)DD lesions had titers compatible with infection. We are continuing analysis on the serology and observational results.

We conclude that for the two Northern California dairies studied during 6 months, vaccinating the whole lactating herd did not provide significant prophylactic or therapeutic effects. We speculate that the vaccine might prove more efficacious if used on animals prior to exposure to high infection pressure before joining the milking herd.

INVESTIGATION OF THE EFFICACY OF THE KOVEX-FOAM-SYSTEM IN THE DECREASE OF THE INCIDENCE OF DERMATITIS DIGITALIS; DERMATITIS INTERDIGITALIS AND EROSIO UNGULAE

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Introduction

Dermatitis digitalis is a circumscribed superficial ulceration of the skin bordering the coronary margin, with an epithelial border and chronic dermatitis. On the other hand it looks like a circumscribed proliferative, wartlike lesion at the same location1,2. Dermatitis digitalis is a multifactorial disease with an infectious component3. Dermatitis interdigitalis is characterised by an acute or chronic epidermitis in the interdigital cleft. Usually it affects also other regions, i.e. plantar/palmar and dorsal areas of the skin, respectively. Its location and the existing range of germs do not allow a precise differentiation from Dermatitis digitalis4. Changes of the heel horn in correlation with Erosio ungulae are also mentioned in connection with Dermatitis interdigitalis. Some authors describe these changes as separate diseases, whereas others relate to them as symptoms of the aforementioned disease5,6. The generic term "Digital Skin Disorders Syndrome" was suggested4. For a long time one has tried to control these diseases by the means of foot-baths. Even today there are few investigations showing the efficacy of foot baths. Furthermore the use of most common substances (CuSO4, formalin, antibiotics) to heal, soothe and prevent illnesses is not allowed by the German Drug Law (AMG § 2)7. The use of foam based on tensides and peracetic acid before entering the milking stall should remove dirt from the feet after an immersion time of at least 5 minutes. This compact layer of dirt (faeces, liquid manure, slurry) contains microbial populations, which are involved in the described cases of diseases. Moreover the foam is supposed to be disinfectant, due to its low pH value.

Material and Method

Every 6 months a functional hoof-care is performed by professional hoof trimmers at a farm with 70 animals. The animals which are tied up in a longstall on rubber mats, are milked twice daily in a milking stall next door. Before the treatment was commenced the animals were first examined for signs of Dermatitis digitalis, Dermatitis interdigitalis and Erosio ungulae on the hind extremities. 8 weeks after the use of the Kovex-foam-system the animals were re-examined and a further examination was performed again 12 weeks later. Kovex-foam is a hygiene product consisting of an "activator" containing tensides and a "base" with peracetic acid. It is applied to several square meters of an evenly consolidated ground in front of the milking stall. Therefore a mixing unit is installed near the milking stall to blend and foam the two substances. By means of nozzles the prepared foam is applied to the ground. This procedure can be controlled manually. The animals walk through the relatively thick foam which adheres to their hooves, where it helps to remove the dirt. Fresh foam is repeatedly reapplied under supervision. The cattle enter the milking parlour with foam still on their hooves which will break down as it dissolves the soiling.

Study course

The claws of 55 animals were examined before the use of the Kovex-foam-system. After entering the tied-stall on the left and right side of the feeding table the animals were each allocated to a group. Thus 22 of these animals were included in the test group, the other 33 were included in the control group. The control group at the right side of the feeding table is milked first (without foam), followed by the test group standing at the left side of the table (with foam). Before performing the test the cleansed hind extremities were examined for signs of Dermatitis digitalis, Dermatitis interdigitalis and Erosio ungulae and the results were documented. Subsequently the Chi-square and the Fisher`s exact test were used as the statistical method.

During the first 8 weeks the treatment began with a stabilisation phase, where the foam used twice a day in the first week and then suspended in the following week. The 22 animals of the test group were sent through the foam on their way to the milking parlour. Thereafter a new examination of the hind extremities of all animals in both groups was performed. In the following phase, the phase of maintenance, the animals waded through the foam twice daily on 3 consecutive days, then were not treated for 11 days. This change of treatment was to be maintained permanently. After 12 weeks the final assessment of the hind extremities with regards to Erosio ungulae, Dermatitis interdigitalis and Dermatitis digitalis took place.

Results

1. Examination prior to the start of the investigation
   58% of the control group and 50% of the test group were affected with Dermatitis digitalis. 73% of the animals in the control group and 82% of the test group showed changes of Dermatitis interdigitalis. Erosions of the heel horn were found in approximately 76% of the control group and 45% of the test group.
2. Examination at the end of the stabilization phase
   48% of the control group were still afflicted with Dermatitis digitalis (26 % of the affected animals healed) and 15% of the test group showed Dermatitis digitalis after the stabilization phase (45 % of the animals with signs of Dermatitis digitalis at the first

examination healed). 41% of the control group showed deviations according to Dermatitis interdigitalis. In 25% of the animals within the test group deviations were seen. Now 51% of the control group still showed heel horn erosions, only 30 % of the infected cows had healed. In the test group there were no more affected animals (0 %).

3. Examination after 12 weeks of the phase of maintenance
   Now 73 % of the control group were afflicted with Dermatitis digitalis, 5 of the healed animals (see above) showed lesions again. In the test group there were 55 % afflicted with the disease, 5 of the healed animals (examination after the stabilization phase) had had a relapse, too. 38 % of the control group still showed Dermatitis interdigitalis, 18 % of the test group were laid up with Dermatitis interdigitalis. Erosions of the heel were found in 65 % of the animals in the control group, and in 45 % of the animals within the test group. In each group 5 of the healed cows had a relapse, the others didn't show the characteristic lesions before.

Evaluation after the stabilization phase

1. **Dermatitis digitalis**
   With Dermatitis digitalis there was a significant correlation between the incidence of the disease and the use of the Kovex-foam-system ($P = 0.013$). There was a relative risk of 2.70 for the animals in the control group not to be completely cured (95% confidence interval: 0.9922 -7.3569).
2. **Dermatitis interdigitalis**
   Dermatitis interdigitalis showed no significant effect on the use of the Kovex-foam-system ($P = 0.1465$).
3. **Erosio ungulae**
   The incidence of erosio ungulae was significantly dependent on the use of the Kovex-foam-system ($P < 0.0016$). There was a relative risk of 13.11 for the animals in the control group not to be completely cured (95% confidence interval: 0.8584-200.3933).

Evaluation after the phase of maintenance

After the phase of maintenance there were the following results
There were no more significant differences between the control group and the test group with regard for Dermatitis digitalis, Dermatitis interdigitalis and Erosio ungulae. As to Dermatitis digitalis a trend could be noticed towardsa lower number of animals affected in the test group than in the control group.

Discussion

Due to the significant differences of the hoof condition regarding Dermatitis digitalis, Dermatitits interdigitalis and Erosio ungulae the following can be concluded: The use of dirt-loosening tensides (activator) and the principle to minimize the risk of contamination among the animals by the base (peracetic acid) influences the infectious component3of the mentioned diseases. Especially for combating Dermatitis digitalis and Dermatitis interdigitalis the use of foot-baths is "traditionally" recommended8,9. However, the short reaction time of 30 seconds which obviously can hardly be prolonged with walk-through-baths, does not result in the desired success to stem diseases like Dermatitis interdigitalis10. Compared to the walk-through-baths the contact time of thick foam before the milking process is prolonged and undoubtedly helps to optimise the loosening of the dirt and to reduce the amount of bacteria in the dissolved slurry. The observed restlessness of some animals in the test group during the first week of the therapy with Kovex-foam is likely to be due to a pain reaction. It is assumed that the contact of the foam with a low pH-grade with the lesions is the cause. However, later the animals walked through the carpet of foam without hesitation. Nevertheless the occurrence of the diseases could not totally be eliminated.
Moreover the rate of the diseases increased after the phase of maintenance (3 days with foam twice daily, 11 days without foam). Obviously the regime has to be modified (one week once a day and one week rest or every other day once a day or 4 days once a day every week) to prevent a reccurence of the diseases.
This also emphazises the multifactorial component3. Especially the importance of surface-hygiene on walking- and standing-areas always has to be a main topic. The dissolving of the dirt in the milking parlour for a few minutes cannot alone prevent a new contamination. Dietary factors such as inadequate protein, carbohydrate and raw fibre concentrations as well as a mismanagement and a lack of Cow Comfort also have to be considered.

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TREATMENT OF DIGITAL DERMATITIS WITHOUT USING OF ANTIBIOTICS - A CLINICAL TRIAL

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Abstract

A study was conducted at a high yielding dairy farm to measure the effect of non-antibiotic treatment on digital dermatitis (DD). Functional claw trimming was performed prior to the start of the experiment and 11.8 % of feet (22 of 144) was diagnosed to DD in the herd. Pediline fluid (CID Lines, Belgium), which contains glutaraldehide, copper sulphate, aluminium sulphate, benzalconium chloride, allantoin, non-ionic surfactant, complexing agents and stabilizers, was used for treating the animals. After the first phase of the experiment the occurrence of DD showing clinical symptoms (pain and lameness) was decreased even good epithelisation of the DD lesions were observed. After the second phase all legs were clinically recovered. In conclusion Pediline was strongly effective in the treatment of DD.

Introduction

Lameness in dairy cattle is a major cause of economic losses with approximately 10 to 30% of cows experiencing lameness annually. Lameness was reported as the reason for culling 15% of dairy cows sent to slaughter in the USA in 1996. Lameness is an unacceptable condition which causes severe pain, decreased milk yield, reduced reproductive performance, high culling rates and increased cost of veterinary intervention (Hernandez et al., 2001; Hernandez et al, 2002; Mülling and Lischer, 2002). Average annual losses due to diseases of the bovine digit with clinical symptomps were found US$ 127 per case and 27 US$ per cow present in the herd (Huirne et al, 2002).
Digital dermatitis (DD) is a highly infectious disease that causes painful ulceration to the skin of the foot. The incidence of this disease has increased greatly in recent years. It usually responds to treatment with a range of broad spectrum antibiotics (e.g. lincomycin, oxytetracycline) but commonly recurs within the same lactation (Kofler, 1998; Webster, 2002). Approaches to therapy of DD include surgical excision, foothbaths, topical treatment with various disinfectants or antibiotics, cryosurgery or electrocautery, topical treatment under bandage and systemic antibiotic treatment (Kofler, 1998; Shearer and Amstel, 2002). There are promising studies with using stable-specific vaccines against DD (Szemerédi et al., 2003). Non-antibiotic formulations for treatment of DD may be desirable because of environmental, milk residue and resistance problems of antibiosis.

Materials and methods

The study was conducted in a high yielding dairy farm in Hungary in 2002. The lactational milk yield of the cows was 9000 l on average.
The first sign of DD was the increasing incidence of lame animals in the herd. All the animals were examined and 11.8 % of feet (22 of 144) was diagnosed to DD in the herd. For therapy, Pediline fluid (CID Lines, Belgium) was used. This non-antibiotic formulation contains glutaraldehide, copper sulphate, aluminium sulphate, benzalconium chloride, allantoin, non-ionic surfactant, complexing agents and stabilizers. Pain and size of the lesions were evaluated according to Britt et al. (1999).
In the first phase of the treatment the animals, which had moderate pain and lesion's size < 2.5 cm, were functionally claw-trimmed and walked through a footbath with 5% solution of Pediline two times daily during five days.
One week later, in the second phase of the treatment, the administration of Pediline was repeated but only once a daily footbath was applied during five days.
In serious cases (7 legs, severe pain and lesions > 2.5 cm) individually treatment was applied by using 20% solution of Pediline sprayed onto the sick surface after trimming and cleaning the claws. These feet were treated once daily during five days and treatment was repeated one week later.
The recovery process was evaluated by detecting the pain and size of the lesions, and pictures were taken by digital camera about each legs on day 0, 3 and 5 during the period of treatment.

Results and discussion

After the first phase of the treatment, the occurrence of the clinical symptoms (pain and lameness) was decreased even good epithelisation of the DD lesions and decreasing lesio size were observed. After the second phase of the experiment all the feet were recovered. This good recovery was observable either after footbathing or topical treatment.
Several results of research were published that show positive effects of non-antibiotic treatments of DD. In their study, Laven and Hunt (2002) compared the efficacy of the footbaths with three non-antibiotic product (copper

sulphate, formalin and peracetic acid) and erythromycin. They found that the lesion score decreased significantly in all four groups but there was no difference between the abtibiotic and non-antibiotic treatment. Gradle et al. (2002), in an uncontrolled clinical trial, also reported that a commercial non-anitibiotic mixture was effective, and Blowey (2000) suggested that formalin footbaths could be suitable treatment for DD.

Most published studies have described the use of non-antibiotic products applied as topical application to individual animals. In these trials copper sulphate, peroxide and cationic agents were used as spray or cream (Hernandez et al., 1999; Shearer and Hernandez, 2000; Moore et al, 2001). Pospicher and Kofler (2002) treated the animals topically with a mixture containing organic acids, essential oils and salts of potassium, zinc, copper and aluminium at low pH. These researchers showed the efficacy of the non-antibiotic products in the topical treatment of DD.

In conclusion, Pediline was effective in the treatment of DD either in footbath or local application in the present uncontrolled field trial.

Acknowledgements

Authors acknowledge CID Lines, Belgium and HAT-Agro Ltd, Hungary for the placing at disposal of the Pediline and the support for the study.

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TREATMENT OF DIGITAL DERMATITIS IN AUSTRIAN DAIRY COWS WITH THE NON-ANTIBIOTIC PASTE PROTEXIN™ HOOF CARE

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Introduction

The most common methods of treatment of digital dermatitis worldwide include antibiotic and non-antibiotic formulations applied topically with / without bandage, in footbaths or parenteral antibiotics and surgical removal of digital dermatitis lesions especially from the interdigital skin (1-6, 8, 11-18, 20). The most frequent therapy for digital dermatitis in Austrian dairy cows is the topical oxytetracycline spray or other topical antibiotics (11).
Non-antibiotic formulations for treatment of digital dermatitis may be desirable, as any antibiotic treatment can produce antibiotic residue in milk, meat and the soil and progressive resistance development has been reported (7, 19).

Material & Methods

In this study, 47 cows of 8 different dairy farms with loose housing systems and slatted floors showing acute stages of digital dermatitis located on the plantar/palmar or dorsal skin-horn junction of the claws were randomly assigned to 2 groups. Twenty-six cases of digital dermatitis were assigned to each group. Cows with digital dermatitis lesions on the interdigital skin and on atypical sites, cattle with, other disorders causing lameness, and cattle treated with antibiotics and antiinflammatory drugs for other reasons were excluded from this study. The study group with 26 cases of acute digital dermatitis was topically treated with the non-antibiotic paste Protexin Hoof-Care (Probiotics International Ltd., Stoke sub Hamdon, Somerset, UK; study group). Protexin Hoof-Care contains organic acids (formic-, acetic- and propionic acid), salts of copper, aluminium and zinc and essential oils (peppermint, eucalyptus) and has a low pH.
Twenty-six cases of acute digital dermatitis lesions were in the control group and were topically treated with oxytetracycline spray (Terramycin®Aerosol Spray, Pfizer Austria; control group). Pre-treatment and control examinations were documented on 4 occasions, (day 0, 4, 10 and 28) using a standard protocol and a digital camera.
On day 0 the lameness score at rest (0, 1, 2, 3) and at walk (0, 1, 2, 3, 4) was determined. After functional claw trimming the claws and the digital dermatitis lesions were washed and dried with clean tissues. The pain score (0, 1, 2, 3) was determined by digital pressure. The shape, colour, surface morphology and size of the lesions were documented. Protexin Hoof-Care or oxytetracycline spray was applied to the lesion according to the randomisation table. After topical treatment all cows were kept on a dry surface for 30 minutes before returning to the stable.
Statistical analyses were carried out by means of the SAS 8.02, Windows NT Version. The Mc Nemar test was used for comparison of the distribution of changes in scores (lameness, pain) on day 4, 10 and 28 within each group (examination for agreement; $p < 0.05$). The Wilcoxon-Mann-Whitney Rank Sum test ($p > 0.05$) was applied for the unpaired comparison of the scores of the study group and the control group.

Results

In these 47 cows a total of 52 cases of acute stages of digital dermatitis were diagnosed. Forty-eight cases were in the hindfeet and 4 cases on the forefeet with the typical localisation on the plantar/palmar aspect of the claws at the horn-skin junction. In 5 cows digital dermatitis lesions were found in both hindfeet. The mean lesion size was 3.43 x 2.59 cm in diameter.
All acute lesions were highly (17/16) or moderately (9/10) painful on day 0. On day 4 the lesions were black in colour and had a crusty surface. On day 4 the cows showed less pain in both groups: about half of the lesions in each group (13/14) were painless, the others were slightly painful. On day 10 twenty-one cases of the study group and 23 cases of the control group were painless, and on day 28 only 3 cases of the study group and 2 cases of the control group showed a slight painful reaction on palpation.
On day 10 lameness was no longer seen in any cow of the group treated with Protexin Hoof-Care, and only in one cow of the group treated with the oxytetracycline spray. Good epithelializiation of the digital dermatitis lesion was observed on day 10 in 16 cases of the study group and 14 cases of the control group. On day 28 the lameness score was 0 in both groups (Fig. 1).
Our data indicate that both topical applications of the Protexin Hoof-Care paste and oxytetracycline aerosol spray had an identical efficacy for the treatment of digital dermatitis in these dairy cows. No statistical difference ($p> 0.05$) in the examined scores (lameness, pain) could be found between the study and control groups.

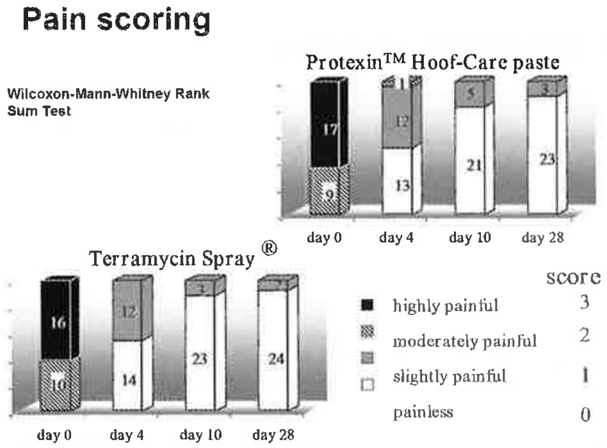

Fig. 1: Development of the pain score of both groups from day 0 to day 28.

Discussion

The convincing results of the non-antibiotic paste Protexin Hoof-Care for the treatment of digital dermatitis, for which spirochaetes are responsible as bacteriologic agents, is caused by the antimicrobial effect of the components of this formulation. These are organic acids and copper- and zinc-sulphate salts which are known for their antiseptic activity, for example in footbaths (1, 5, 12, 15). The low pH of about 2 of this non-antibiotic formulation prevents bacterial growth (5).

The results showed that treatment with either Protexin Hoof-Care paste or oxytetracycline aerosol spray resulted in a significant improvement in digital dermatitis within 4 to 10 days of the first treatment. The examined scores (lameness, pain) showed no statistical difference ($p > 0.05$) between groups.

It has been reported that the site of digital dermatitis lesions can affect the efficacy of topical spray treatment with oxytetracycline in dairy cows (9). Therefore only cows showing lesions on the skin of the heels or the dorsal coronet were selected for this study.

The results in 47 cattle with spontaneous, acute digital dermatitis lesions agree with similar studies from the USA, in which the non-antibiotic formulation Victory® was tested for treatment of digital dermatitis lesions in cattle over a period of 12 to 30 days (1, 13, 17).

The use of a placebo group would have been desirable in this study. Which was carried out under field conditions in private dairy herds and the agreement of the cattle owners for a placebo group could not be obtained. A study on cows of a veterinary teaching hospital herd in Munich showed that the cows with digital dermatitis lesions treated only with the pacebo (water spray) did not show any improvement of the lesion within 10 days (3).

The advantage of Protexin® Hoof-Care paste is that it contains no antibiotics, is not subjected to the medical prescriptions and therefore has no withdrawal time, and cannot leave any antibiotic residue in milk or meat. Formic acid, copper and zinc sulphate are indicated in annex II of the official regulation (EWG) No. 2377/90 of the Council and are permissible in food animals (21).

The results of this study show that Protexin Hoof-Care paste can be recommended to veterinarians and claw trimmers as an interesting and efficient alternative to topical antibiotic treatment for digital dermatitis in dairy cattle.

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21BMGF Bundesministerium für Gesundheit und Frauen (2003):www.bmgf.gv.at; access: 17.09.2003

ANIMAL WELFARE AND CLAW DISEASES

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It might seem to be a strange metamorphosis for a group that primarily discusses the classification, etiology, pathophysiology, and management of digital disease to turn to the welfare problem represented by the occurrence of lameness in ruminants. I feel otherwise. To eventually take our knowledge about these diseases back to the farm and consider a holistic vision of lameness seems a natural progression. As we understand more about the causes and cures for lameness we turn to examining the meaning of lame animals to the health and well being of the herd or flock as well as the economic well being of the producer. The veterinary profession is usually responsible for the care of sick or injured animals, either directly or via surrogates such as farm managers. An oath sworn by veterinarians in the USA upon completion of their studies includes these phrases, "...I swear to use my scientific knowledge and skills for the benefit of society through the protection of animal health, the relief of animal suffering, the conservation of livestock resources..." These words clearly support the idea that, as useful information is available, veterinarians are obligated to use this information to work for the welfare of the animals in their or their clients' care. Lameness, despite the veterinarian's claim of expertise and responsibility, is more often treated by the farmer or hoof trimmer. Perhaps there are more important roles for veterinarians and other experts in lameness than in therapy for the individuals that the farmer asks to be treated. Outreach and education of those closest to the herds might enhance animal welfare more than improving our own techniques. Clearly, there are many unanswered questions about the conditions causing lameness in cattle and naturally some disagreement about the proper steps to prevent or treat lame individuals. However, it is within our mandate to consider the welfare ramifications of our knowledge. How well are we doing at transmitting the collective wisdom about lameness to our peers and clients? Are the practical results of our research widely available to aid in the welfare of our patients? What are the unanswered questions?

There are 3 areas of concern regarding the welfare of domestic ruminants and claw diseases. These are the prompt treatment of affected cattle to promote healing of the disease, the relief of pain due to the disease, and the prevention of cases of lameness by whatever means are appropriate. Each concern will be addressed individually with the understanding that they are interrelated. I will use cattle for most of my examples throughout the paper with the understanding that other ruminants merit similar considerations.

Detection and Treatment of Lameness

Prompt treatment of lame individuals requires several steps. First the detection of the disease or lameness in the affected cow must take place. Herd managers carry the responsibility for establishing systems for detecting lame cattle. In small herds and flocks the responsibility often is solely held by the owner but in larger units the complexities of labor management and delegation of duties often place herd managers or other workers in charge of detection of lame cows. For most herds and flocks the detection of lameness is done by observing the animals walk. For dairy cattle observation may be done at milking time for parlor systems or by moving through the pens or corrals of cattle at other times when the cattle are active. For tied cattle observation may occur during exercise periods. If there are no exercise periods it is sometimes more difficult to detect mild or subtle cases of lameness. For beef cattle on pasture stockmen generally move through the cattle at some interval to check for general health conditions including lameness. Confined beef cattle are usually inspected intensely by cowboys on horseback or other caretakers at least daily. All these methods of detection rely on the judgment of an individual who accepts the responsibility of identifying lame animals. With millions of such people in this role in the world there is certainly a wide variation in the definition of lameness and the sensitivity of the test employed. What this means from the animals perspective is a variable degree of lameness or duration of disease before an intervention is chosen. We must realize that this variability in detection sensitivity and timing represent welfare problems for lame animals. Certainly there are owners and managers who are very astute in identifying lame members of their herds and flocks. However, surveys done in both the United Kingdom (Mill 1994, Whay 2002b) and the United States (Wells 1993) have pointed out the apparent insensitivity of farmers to the degree of lameness that could be documented in their dairy herds. Too often the ordinary which includes lame individuals is judged to be normal and the lame individuals are allowed to suffer without human assistance. In my opinion, the covenant we have with our domestic stock is to help heal their diseases when possible and to provide euthanasia when we can not.

The choice of intervention once the animal has been declared lame includes as much variation as the reliability of the detection method. Treatment for lameness is done by everyone including farmers, their employees, hoof trimmers, veterinarians in general practice, and veterinarians who specialize in the affected species or who are hospital-based clinicians with specialized resources. Furthermore, farmers often receive advice about treatments from neighbors, truck drivers, other farmers, and now via the internet. A great part of this resulting variability comes from a lack of knowledge of the diseases causing lameness and in less developed regions lack of resources for restraint or therapy. It is of grave concern that treatments are sometimes inappropriate and may in fact create more pain or tissue damage than the original disease. These inappropriate treatments are in no way confined to the less developed regions of the world.

Technological means of detecting lameness are used by some farmers. The system employed most commonly is used in dairy cattle and consists of either pedometers worn on a limb at the fetlock joint or activity monitors worn around the neck. These systems are primarily employed to detect estrus via increases in steps or head bobs compared to an individuals rolling average. They have the secondary capacity to detect decreases in activity associated with lameness. Companies marketing these systems have not stressed nor developed their software to promote lameness detection. In fact, the sensitivity of the detectors to motion sometimes results in higher activity scores for cows that limp badly because they generate more exaggerated movements of the sensor. To the author's knowledge no systematic evaluation of movement detectors has been made with regard to lameness detection. Herds with activity monitors almost always have automatic milk recording. These data allow the cows who have a significant drop in milk production compared to their rolling average to be identified and examined. If the problem with the cow is lameness, it is usually identified during the general examination that is standard procedure in many herds with automatic milk recording, even if activity monitoring is not part of their systems.

An alternative technology for detection of lameness is under development for commercial application by Bou-Matic company of Madison, Wisconsin, USA. Some research about this system has been presented in previous meetings of this organization. The detection of lameness relies on measuring signals from a load sensing platform that is placed in the return alley from the milking parlor. Computer manipulation of signals from the sensors can estimate load placed on each leg and stride characteristics. Deviations from normalized data are used to identify cows with lameness. To the authors knowledge this system has not been deployed on commercial farms. Other systems not even dreamed of yet by engineers will probably be developed in the near future. One possibility is for video recognition software such as is used in security monitoring to identify individual cows within a herd and further characterize their gait in some fashion that would permit identification of lameness. The author has had discussions with a software company that indicate the feasibility of this method.

There clearly is a role for veterinarians, livestock specialists, and extension personnel to help educate farmers and their employees about lameness recognition. This notion seems so simplistic but the performance of many current herd managers in lameness detection is so poor that a need clearly exists for more effort. Secondly, appropriate treatments should also be taught so that relief of pain and suffering might be as prompt and effective as possible. Since treatments are delivered by veterinarians, farmers, and hoof trimmers, all these groups should be included in efforts to deliver more knowledgeable interventions. There would certainly be disagreement about the final details of appropriate therapy for even many common conditions among the attendees at this meeting. Perhaps we should encourage more clinical research in the areas of therapy for the most common conditions.

Pain Management for Lame Stock

The science of pain management has made many advances in human medicine in recent years and includes books and journals devoted to the subject. There have been some symposia in veterinary medicine devoted to pain management but their focus has been companion animals not ruminants. A very recent textbook on pain management for animals is quite comprehensive but offers little for ruminants (Gaynor 2003). The methods of pain management for lameness problems involve both the specific therapy for the primary problem, may involve pain avoidance by methods such as hoof blocks and housing in more comfortable or protected locations, and specific pharmaceutical agents for amelioration of pain (O'Callaghan 2002). Dr. Becky Whay provided a concise review of the pathophysiology and treatment of pain at the 12th International Symposium on Lameness in Ruminants (Whay 2002a). The first options should be part of the general philosophy of lameness treatment. The intervention should relieve the pain of the primary condition whenever possible. Changing the housing to a hospital pen or other environment where competition for food, water, and a comfortable place to lie are minimized should also be a standard part of lameness therapy. We are aided in our understanding of the physiology of pain in lame cattle and the usefulness of pharmacological intervention by the work of the group in Liverpool (Whay 1998)Specific drug therapy for pain management depends on drug availability which varies greatly around the world. In the United States the only drugs available for pain management are 2 corticosteroids, dexamethasone and isoflupredone, flunixin meglumine, and aspirin. Only aspirin is specifically labeled for pain control and thus veterinary prescription is required for use of the other drugs in pain management with associated withholding of meat and milk from market channels. Registration requirements for animals that produce food products are very restrictive in the United States and apparently somewhat less so in other countries. Flunixin meglumine may be used in special circumstances in dairy cattle in the United States if a set of regulatory criteria are met. The enthusiasm with which animal owners and their veterinarians employ such drugs are often linked to their cost and their requirements for withholding milk or meat from the market. In my opinion, such drugs are not routinely used for lame cattle due to the reluctance to advocate their use by veterinarians who are sensitized to the owners concerns about treatment costs and subsequent drug-dependent restrictions on marketing options.

Veterinarians treating lameness in the United States are, in general, probably not advocating treatments aimed at pain relief. Why? First, there is no educational effort in the general veterinary curricula of the colleges devoted to the topic of pain in food animals or in its management. Second, cost of therapy is always on the mind of veterinarians who perhaps have an overriding concern for treatment costs. We have been investigating the cost of lameness in dairy cattle for several years. The average cost of treatment including all labor by farm workers is $26 per case which is about 10% of the total cost. Typical drug cost and associated milk discard for 3 days of therapy with flunixin meglumine would add $61 to the treatment cost. The cow would have to make more milk than otherwise to repay the investment in the drug treatment and this would be about 800 lb of milk at current prices in the USA. We do not have any data on the improvement in recovery from lameness with treatment for pain with drugs such as flunixin meglumine. Veterinarians and farmers alike in our dairy culture would hope to justify the treatment of pain on economic grounds. Is this ethical? Should pain relief be attempted regardless of economic consequences? In reality, if veterinarians prescribed expensive treatments for lameness they would likely not be asked to examine or treat future cases unless the treatments were clearly superior to historic treatments that did not treat pain. In the system of drug control in the United States aspirin may be used at the discretion of the farmer. If aspirin could have similarly beneficial effects to flunixin it might be more acceptable since a 3 day course of treatment would cost $30 and only require 400 lb of extra milk to balance that cost. Corticosteroids require no milk withhold in our regulations. Thus, if their use would be beneficial and not harmful to pregnancy, the cost would be only $3 for 3 days treatment which almost any farmer would tolerate. I am unfamiliar with drug use practices or regulations in other countries so can not discuss pain management in those jurisdictions.

Animal Welfare via Disease Prevention

Prevention of diseases causing lameness can hugely contribute to animal welfare. Changes in management or housing or outside environmental management are the primary tools for lameness prevention. It is beyond the scope of this paper to discuss all the known risk factors in detail. Feeding management is considered by many to have a central role in preventing ruminal acidosis and subsequent laminitis/coriosis. Housing design and maintenance have many influences on animal health. Walking surfaces and resting places are very influential in predisposing to claw horn diseases. Manure management and foot bathing control infectious digital diseases. Outside the barn the conditions of corrals, animal walkways or lanes, and around feeders or waters can influence both claw horn and infectious diseases. Good designs and management practices have the ability to greatly minimize lameness problems at the herd level. Achieving a low incidence of lameness should be a prominent concern in establishing design criteria for new or renewed facilities. Once facilities are in place the management of them is equally important for successful control of lameness. Once again, the role of veterinarians and livestock management advisors can be to educate producers to establish management regimes that reduce the known risk factors for digital diseases. Comfortable lying areas or stalls, minimizing time standing on concrete, clean and dry alleys, diets that do not cause ruminal acidosis, regularly scheduled hoof trimming, non-injurious cow lanes and control of mud around feeding and drinking places for cattle kept outside will all contribute to producing a lower incidence of lameness. Many producers have operations that violate more than one of these guidelines for lameness prevention. Since many of these same producers seem immune to welfare concerns perhaps economic arguments explaining the cost of lameness will help persuade them to adopt more cow-friendly practices and create fewer lame cattle.

Remaining Questions

In light of the apparent inability of farmers to adequately recognize lame cattle in their herds, perhaps the most pressing question is, why not? Furthermore, what can the informal group of enthusiasts attending this conference or reading these proceedings do to improve the detection and treatment of the lame individuals which inevitably populate all herds? Can we define best management practices for treatment of individual cases of lameness? Will pharmaceutical companies invest in registration for effective drugs to treat the pain of lameness? Can we convince farmers of the ethical imperative to acknowledge that lameness means pain and without pain management we are not fulfilling our commitment to our domestic animals?

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LAMECOW 2002-2006 A MULTIDISCIPLINARY APPROACH TO THE REDUCTION IN LAMENESS AND IMPROVEMENT IN DAIRY COW WELFARE IN THE EUROPEAN COMMUNITY

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Introduction

Lameness in dairy cows is well recognised globally to the extent that it is now the subject of this dedicated international bi-annual conference. Lameness is the major farm animal welfare problem in Europe. It is an unacceptable condition which causes severe pain, decreased milk yield, reduced reproductive performance, high culling rates and increased cost of veterinary intervention. There is general consensus that lameness has multi-factorial causes including management, housing, genetics, breeding, nutrition and physiological state, all of which can influence the incidence of hoof related lameness in dairy cattle. Some commercial dairy farmers are far more successful in maintaining claw health than others. This is exemplified by a recent report which surveyed 340 UK dairy farms and showed that the best quartile of farms had a lameness incidence of 5.8% compared with 50.3% in the worst. While cubicle housing was shown to produce a mean incidence of 25.5% compared with 17.7% in straw yards, lameness occurred with an incidence of 6.6% in the best quartile of cubicle houses compared with the much larger value of 34.4% of the worst quartile for straw yards Whitaker et al 2000). The combination of reasons for these critically major differences are not known. However what is clear is that their identification and application by the poorer three quartiles has potential to radically improve foot health of the dairy cow. A major aim of this project is to identify best practice criteria and test responses to the application of such information.

Intensification of dairy production systems has led to a disproportionate increase in the incidence of non-infectious lameness in the last three decades. Little attention has been given to genetic traits associated with foot health and to the fundamental biological and physiological characteristics which may predispose the hoof unit to a lesser or greater resistance to lesion formation. Our current limited knowledge indicates that lameness is a multifactorial disease caused by factors which are extrinsic and environmental and intrinsic and animal-related in origin. It is therefore necessary to combine applied farm-based approaches with those of cutting edge laboratory investigation to more clearly evaluate the role of both animal and environment in the aetiology of the disease and to formulate solutions to its reduction.

It is against this background that research scientist from across Europe have come together to form the research consortium funded by the EU Framework 5 programme and known as LAMECOW which has as its major ?? aim the improvement of our understanding of the multifactorial causes of lameness in dairy cows. The project involves partners in the UK (University of Aberdeen, University of Warwick), Sweden (Swedish University of Agricultural Science), Germany (Freie Universität Berlin, Klauenpfleger Meister Rene Pijl), Austria (University of Veterinary Medicine Vienna) and Poland (August Cieszkowski University of Agriculture). In addition, contribution to the project will be made by the following of subcontractors with expertise in specific areas; these are IPC Oenkerk, VitroTec Development Berlin Germany, Austrian Institute of Polymer Science, Roger Blowey and Alberto Brizzi.

Objectives

The aim of this project is to reduce the incidence lameness of infectious and non-infectious origin in dairy cows through analysis of 'best practice' in dairy enterprises in member states of the EU and to understand the biological mechanisms by which non-infectious lameness is caused and may be minimised.

Our specific objectives are:

1. Assessment of husbandry systems: To evaluate existing husbandry systems in relation to the incidence, nature and severity of lameness and its impact on animal wel fare in current and future member states of the EU. This will be achieved through on-farm monitoring, data recording and identification of interactions between foot health and different biological conditions in heifers and dairy cows and to formulate "best practice" husbandry systems to reduce the incidence of lameness and improve animal welfare. (Work packages 1, 2, 3, 4, 7)

2. Morphology and bio-mechanics: To elucidate the mechanisms by which husbandry systems and

physiological state interact with hoof tissue using an integrated approach employing morphological and bio-mechanical methods. (Work packages 5 and 6)

3. Model systems: To establish and use ex-vivo and in vitro model systems such as isolated limb perfusion, tissue explants and keratinocyte culture to study the effects of inflammation and metabolic stress factors on the vascular system in the dermis of the hoof and the cellular and molecular mechanisms by which dermal and epidermal tissues interact and respond to bioactive mediators. (Work packages 8, 9 and 10)

4. Guidelines and training packages: To recommend standards to improve animal welfare, provide guide lines to develop minimal lameness husbandry systems and to modify existing husbandry systems, and to use modern information technologies to prepare multilingual, multimedia training packages for stock people, farmers, hoof trimmers and veterinarians in practice and at the Universities in the different member states of the EU. (Work package 11)

Expected Achievements

The main outputs of the research programme will be:

- Characterisation of "Best Practice" husbandry systems with the potential to reduce lameness and improve productivity and welfare and which will establish the basis for European guidelines for the improvement of existing, and development of new integrated, hus bandry systems.
- Understanding of interactions between foot health/dis orders and different biological conditions (prepartum, pregnancy; parturition and lactation) in heifers and dairy cows
- Understanding of mechanisms by which husbandry sys tems and physiological state interact with hoof tissue.
- Ex-vivo and in vitro model systems available for future studies on hoof biology.
- Multilingual, multimedia training packages for stock people, farmers, hoof trimmers and veterinarians.
- Guidelines for the reduction in the incidence of lame ness and the implementation of 'best practice' hus bandry systems in Europe.

Materials and Methods: Structure of the Project

To achieve our objectives the project is divided into twelve work packages (WPs). WP1 will standardise the methods and measurements to be used on-farm to record the incidence and nature of lameness as part of WP2. The data will be collected on selected farms in four different European locations. The data collected will be categorised and analysed by simple and multivariate techniques to identify factors associated with lameness in WP3. This data will be used to formulate 'best practice' husbandry systems which will be implemented on the participating farms and the impact of the revised husbandry systems evaluated in WP4. Histology and pathology of samples collected on-farm in WPs 2 and 4 and in WPs 7, 8, 9 and 10 will be assessed as part of WP5. The bio-mechanical properties of hoof samples collected on-farm in WPs 2 and 4 and in WPs 7, and 10 as well as samples collected specifically for WP6 will be determined and used to develop computer models in WP6. WP7 will examine the interaction between flooring properties and hoof biology using model systems, full-scale experimental trials and commercial farms. Model systems to examine the mechanisms by with hoof lesion may arise will be developed and applied in studies using cell culture (WP8), tissue explants (WP9) and whole limb perfusion (WP10). In WP11, the data resulting from these studies will be formulated into European guidelines and recommendations for the application of 'best practice' husbandry systems and will be used to produce multilingual training packages which will be tested on participating farms prior to distribution to end user groups with Europe. The co-ordination, and preparation and submission of reports will be carried out as part of WP12.

Results

The results of work carried out so far as part of LAMECOW are presented in separate papers presented at this conference and reported in abstracts by Wüstenberg et al, Frohberg-Wang et al, Mülling et al, Nebel et al, .Hepburn et al (2), Amory et al (2), Manske et al, Telezhenko et al, and Pijl et al.

Discussion

The opportunity to develop collaboration between practitioners and research scientists on a pan-European scale can only be achieved under the umbrella of EU funding. LAMECOW is the first major multinational and multidisciplinary research programme to tackle the problem of lameness in dairy cows. Papers presented at this conference represent the early stages of development of research methods in cell biology, limb perfusion, flooring and locomotion studies. These will be used in combination with the approaches described to collect and analyse farm and animal factors, aided by the development of an electronic data recording system, to improve our understanding of the aetiology of lameness problems and to suggest ways to minimise their occurrence.

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This work is funded by the EU LAMECOW project OLRT-2001-00969.

LAMENESS PREVALENCE AND BEHAVIOURAL TRAITS IN CUBICLE HOUSED DAIRY HERDS - A FIELD STUDY

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Abstract

In the present study, the relationship between the standing behaviour and the prevalence of lameness in commercial dairy herds has been addressed. In total 970 cows out of 17 herds in northwestern Germany housed in standard cubicle barns were locomotion scored and behaviour was individually recorded for 3 h.
The average lameness prevalence was 45% (25-58%). Lame animals spent significantly less time feeding and stood longer with the front feet and with four feet in the stalls, respectively. Behaviour of healthy cows in terms of time spent at the feed bunk and standing with four feet in the stalls was significantly correlated with the proportion of moderately/severely lame cows (rs=-0.72 and rs=0.56, respectively).
In conclusion, our on-farm data did not confirm a relationship of standing with two feet in the stalls with lameness prevalence under the housing conditions investigated.

Introduction

The role of behaviour in the causation of lameness has recently been addressed in a number of studies. This work provided evidence that the way how and where cows spend their time standing influences claw health (e.g. Berry et al. 1998, Galindo et al. 2000). Therefore, the aim of the present study was to investigate the relationship between the standing behaviour and the prevalence of lameness in commercial dairy herds.

Animals, material and methods

17 herds in northwestern Germany (Holstein Friesian) housed in standard cubicle barns were included in the study. Average herd size was 57 cows and the average milk yield was 8700 kg per cow and year. All cows (n=970) were individually marked and their locomotion was scored using a 5-point system (Winckler & Willen 2001). Cows with a locomotion score $\geq$ 3 were considered clinically lame. In all farms, behaviour (lying, standing at the feed bunk, standing in the alleys, standing with the front feet in the stalls, standing with four feet in the stalls) was individually recorded using scan sampling at 15 min intervals during a 3h period between 12:00 am and 3:00 pm. In a previous study, this time slot had been proved representative with regard to the time budget during the day.
The influence of lameness status on activity patterns was analyzed using GLM taking the effect of the farm into account and the relationship between behaviour and lameness prevalence was determined using Spearman rank correlations.

Results

The average lameness prevalence was 45% (25-58%) and the proportion of moderately/severely lame animals (locomotion score 4 and 5) ranged between 2 and 23% (fig. 1).
When all animals cows were classified as either being lame (locomotion score 3, 4 and 5) or not lame (locomotion score 1 and 2), lame animals spent significantly less time feeding and stood longer with the front feet and with four feet in the stalls, respectively.
In order to investigate wether the behaviours shown actually relate to lameness prevalence, only behavioural data from healthy cows were taken into account for this analysis. When these healthy cows spent less time at the feed bunk or stood longer with four feet in the stalls, higher proportions of moderately/severely lame cows (rs=-0.72 and rs=0.56, respectively; n=17) were observed. However, there were no significant relationships with the total time spent standing/lying when the overall lameness prevalence (including mildly lame cases) was considered.

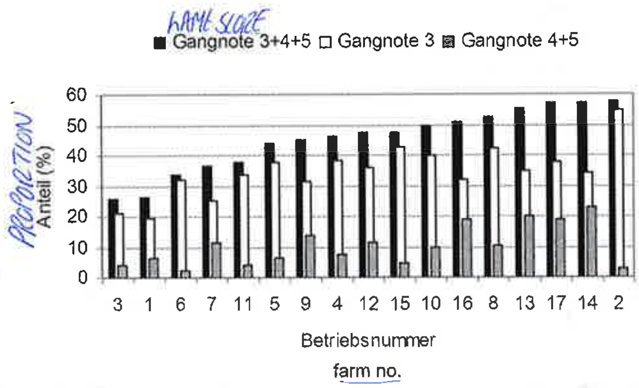

Figure 1: Total lameness prevalence and proportions of mildly and moderately/severely lame cows in the farms investigated

Discussion

In conclusion, our on-farm data confirm changes in time budgets for lame cows which may reduce availability of resources for these animals. However, standing with two feet in the stalls did not seem to be related with the lameness prevalence under the housing conditions investigated.

Acknowledgements

The State of Lower Saxony and the European Union (ProLand Niedersachen) is gratefully acknowledged for financial support. We also wish to thank the farmers participating in the project for the possibility to carry out the investigations and their support during the study.

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A COMPARISON OF DAIRY COW BEHAVIOR IN SAND AND MATTRESS FREE STALL BARNS IN RELATION TO LAMENESS

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Introduction

Cook (2003) found a significantly lower prevalence of lameness in a group of Wisconsin dairy herds using sand stalls compared to those using other types of stall surface, which included rubber mats and rubber crumb filled mattresses, in both free stalls and tie stalls. Although sand has been suggested as an ideal stall surface for dairy cows (Bickert, 2000), there exists little data to support improvements in cattle health related to its use.

The objective of this study was to identify behavioral differences between cows housed in free stalls bedded with deep sand (SAND) and cows housed in free stalls with rubber crumb mattresses (MAT), which may explain differences in lameness prevalence observed between the two types of barn.

Materials and Methods

Twelve Wisconsin dairy herds were selected to include six sand stall herds (SAND) and six mattress stall herds (MAT). At a single milking, all lactating cows, including those in the sick pen, were locomotion scored and the prevalence of clinical lameness was calculated for each herd. For the duration of one 24 hour period, during which no other farm management practices were performed, the mature cow high group pen on each farm was video filmed. Ten cows per farm were randomly selected after the morning milking and color marked with spray paint with a distinctive pattern so that cows could be individually tracked. Each cow was locomotion scored. Location in the pen (alley or stall), activity (standing, lying, feeding, drinking) and time spent performing each activity (to the nearest minute) was recorded for each marked cow.

The data were analyzed using the mixed procedure of SAS. One way ANOVA was used to compare cow and herd level data and a mixed effect model was created to investigate differences in cow behavior between SAND cows and MAT cows.

Results

Mean (SE) lameness prevalence was significantly higher in MAT herds (24.0%, 2.1) than in SAND herds (11.1%, 1.3), ($P<0.001$).

There were no significant differences in parity, days in milk at the day of visit, last DHIA recorded daily milk yield and last DHIA recorded ME305 milk yield between cows selected for filming in SAND herds and those selected in MAT herds. Normal sound cows behaved similarly in both types of barn with respect to time spent milking, eating, socializing and lying down. Although lying time was similar, the manner in which it was obtained showed interesting differences. Cows in SAND herds had a significantly greater proportion (SE) of lying bouts which were greater than 60 min (0.61, 0.03) than cows in MAT herds (0.49, 0.03), ($P=0.03$).

Time spent standing in the stall with all four feet on the platform or perching with two feet on the stall platform and the rear feet in the alley was significantly different between the two groups. Normal cows in MAT herds stood in stalls for 2.4h/d compared to cows in SAND herds that stood for 1.7h/d. Time up in stall for slightly lame cows in MAT herds was 4.4h/d compared to 2.1h/d in SAND herds ($P<0.0001$) and for moderately lame cows in MAT herds it was 6.1h/d compared to 1.8h/d in SAND herds ($P=0.0183$). Moderately lame cows in MAT herds took 46% fewer lying sessions per day and lay down for only 10.0h/d.

Table 1. Effect of Locomotion Score and Stall Base Type (SAND v MAT) on Daily Activity Time Budgets

Mean (SD) Activity h/d	Locomotion Score					
	1 (normal)		2 (slight lame)		3 (moderate lame)	
Stall base type	Mat	Sand	Mat	Sand	Mat	Sand
Lying time	12.0 (2.1)	12.0 (1.6)	11.7 (2.3)	12.0 (2.8)	10.0 (4.9)	12.8 (0.2)
Standing in Stall (including perching)	2.4 (1.4)	1.7 (0.8)	4.4 (2.2)	2.1 (1.9)	6.1 (3.9)	1.8 (1.6)
Time standing in alley (including drinking)	2.8 (1.7)	2.3 (1.2)	1.6 (0.5)	2.2 (1.4)	1.4 (0.7)	1.8 (0.1)
Time Up Feeding	4.3 (1.0)	4.7 (1.0)	3.8 (1.1)	4.6 (1.3)	3.5 (1.1)	5.1 (0.4)
Time Up Milking	2.5 (0.7)	3.3 (1.2)	2.6 (0.6)	3.2 (0.8)	3.0 (0.9)	2.7 (1.3)

Discussion

Mean daily lying time for cows housed in sand free stalls was higher than previously reported (Manninen et al., 2002; Tucker et al., 2003) at 12h/d. The surface cushion provided by sand appears to result in a greater proportion of long duration (> 60 min) lying bouts compared to a mattress surface.

Few studies have accurately documented the behavior of lame cows in free stall barns. Singh et al. (1993) compared behavioral differences in free stall housing. Lame cows lay down for longer (8.3 h/d) than normal cows (6.8 h/d) and spent more time standing in the stalls, the differences however were not statistically significant. Margerison et al. (2002) documented behavior of cows by locomotion score. They noted that stall occupancy (both standing and lying) increased with increasing locomotion score, but failed to examine stall standing and lying behavior separately.

Altered daily activity time budgets were identified in lame cows in this study in cows on mattress free stalls. Increased time spent standing in the stall compressed the time available for other activities such as socializing in the alley, feeding and ultimately lying down - due to a reduction in the number of lying sessions per day. This alteration in activity was not observed in lame cows on sand free stalls.

We speculate that the surface traction provided by sand allows lame cows to rise and lie down more easily, without fear of slipping, thereby maintaining normal lying session behavior in cows with sore feet. The pain and fear associated with rising and lying in lame cows on a mattress stall surface leads to extended bouts of standing in the stall during a lying session. Extended time spent standing in the stall may be detrimental to claw health, increasing the duration of lameness and explaining the higher prevalence of lameness observed in MAT herds.

Acknowledgements

We would like to thank Drs Hunter Lang, David Rhoda, Mike Bradford, Robert Steiner and Jerome Gaska, and Karl Burgi, Jeff Monson and James Bailey for their assistance in selecting the herds, and the farmers for their cooperation with the study.

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USING INDICES OF COW COMFORT TO PREDICT STALL USE AND LAMENESS

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Introduction

Several indices of cow comfort are used on dairy farms including the Cow Comfort Index (CCI) and the Proportion Eligible Lying (PEL) or Stall Usage Index. Targets of >85% for the CCI and >75% for the PEL taken at 1h after return of the cows from the morning milking have been suggested (Overton et al., 2003), but these benchmarks have been derived from very few herds.

The objective of this study was to measure several indices of comfort for one 24 hour period in the high group pens of a selection of Wisconsin dairy herds using either sand or rubber crumb mattress surface free stalls, and to determine the most appropriate time of day for measurement in terms of their ability to predict cow behavior and lameness prevalence.

Materials and Methods

Twelve Wisconsin dairy herds were selected to include six sand freestall herds and six mattress freestall herds. For the duration of one 24 hour period, the mature cow high group pen on each farm was video filmed. For each herd, the film was analyzed every hour in order to determine the number of cows in the pen that were lying down, standing in the stall with all four feet and perching in the stall with the rear two feet in the alley, standing in the alley, drinking and feeding. From these data, for each hour, three cow comfort indices were calculated namely; the Cow Comfort Index (CCI: proportion of cows in stalls that are lying down), the Stall Standing Index (SSI: proportion of cows in stalls that are standing ie. 1-CCI) and the Proportion Eligible Lying or Stall Usage Index (PEL: proportion of cows not eating that are lying down). Daily times spent lying down in the stall and times spent standing in the stall were obtained for 10 cows in each herd. All cows in the high group pen were locomotion scored using the four point system described by Cook (2003).

The start time of the morning milking was used as a reference point to align the hourly data for each farm. Differences in the indices of cow comfort between sand and mattress herds were examined using repeated measures in the mixed procedure of SAS. $P<0.05$ was used to determine significance. The association between the indices of cow comfort recorded at each hour for each herd and the mean herd daily lying and stall standing times derived from the 10 individually tracked cows in each herd and the mean pen lameness prevalence was examined using PROC REG and PROC GLM in SAS. The optimal hourly relationship between the indices and the outcome variables was selected based on an optimal combination of adjusted R2 and P value. A P value of <0.002 was used to determine significance in order to reduce the chances of making an erroneous conclusion due to the multiple comparisons being made.

Results

There was a significant effect of base on CCI and SSI ($P=0.002$) and on PEL ($P=0.003$). The hourly effect was also significant for each variable ($P<0.001$) and figure 1 shows the differences observed by hour for each stall base for the CCI as an example. More variability was observed in the CCI in mattress herds compared to sand herds, and the average over the 24h period was 14% lower.

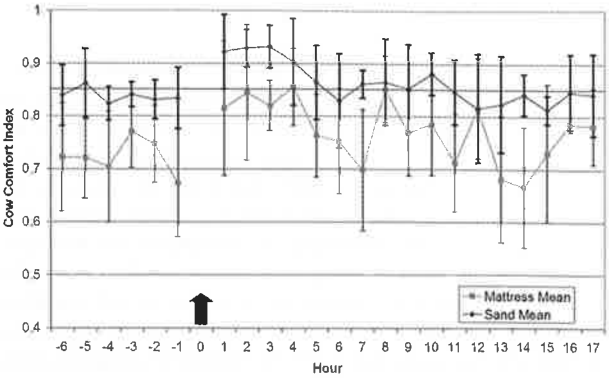

Figure 1. Mean (with one-standard deviation error bars) Cow Comfort Index (%) by Hour for Sand and Mattress herds. The bold arrow represents the time of departure for the morning milking.

All of the indices were poor predictors of mean daily lying time at all hours of the day. There was a significant relationship between CCI/SSI and mean daily standing time in the stall at 5h and 2h before the morning milking, with the reading at 2h having the higher adjusted R2 (0.83) and P value (0.0003) combination. Base was not significant in the GLM at this time ($P=0.24$).

At 2h before the morning milking, there was a significant relationship between CCI/SSI and lameness prevalence in the pen (R2 0.89, $P=0.0005$), but there was also a significant effect of base ($P=0.0008$), which complicated the relationship. SSI greater than 24% was uniformly associated with pen lameness prevalence rates greater than 20%.

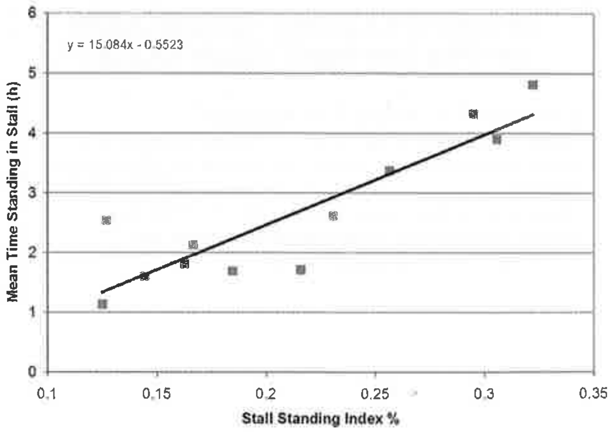

Figure 2. Relationship between Stall Standing Index (%) measured 2 hours before the morning milking, and mean daily time standing in the stall (h/d) for 12 herds

Discussion

Traditional indices of cow comfort do not predict mean daily lying times of individuals within the pen, the CCI or SSI do however predict time spent standing in the stall. Cook et al., (these proceedings) have documented the increased time spent standing in the stall in lame cows in mattress facilities and the lack of this behavior change in lame cows in sand barns. This suggests that these indices are actually monitoring the stall standing behavior of lame cows within the pen.

If we are to use any of these indices, we must re-define our perception of 'cow comfort' as they do not appear to predict mean daily lying time. We suggest that an absence of change in stall use behavior in lame cows could be one definition. Stall standing behavior is an important behavior modification associated with increased rates of lameness in compromised environments. We therefore propose the use of the SSI, taken 2h before the morning milking as a predictor of standing behavior. An SSI greater than 24% taken at this time appears to be associated with lameness prevalence rates in excess of 20% and could be used to trigger locomotion scoring of the herd and a more detailed investigation of lameness and free stall design.

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SWEDISH HOLSTEINS' LOCOMOTION ON FIVE DIFFERENT SOLID FLOORS

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Introduction

The most important part of the dairy housing in its influence on cattle locomotion is flooring (Stefanowska et al., 1998). Concrete is used for most walkways in cattle houses. To decrease risk of slipping on concrete different methods are used like cutting grooves or tamping different patterns on concrete (Albutt et al., 1990). Some new flooring materials have been introduced such as mastic asphalt and rubber mats. The choice of flooring is much discussed, but there is a little empirical basis for objective assessment of animal locomotion on different floors.
The objective of the present study was to compare the locomotion of dairy cows on 5 different solid floors.

Materials and methods

The locomotion of 42 Swedish Holsteins from a research farm (Swedish University of Agricultural Sciences, Alnarp) was studied. The cows were kept in free stalls with slatted concrete passageways. The 30 meter alley (with concrete slatted floor) from the free stalls to the milking parlour was used for the locomotion studies. Cow locomotion was assessed on the following solid floors: smooth concrete, concrete with grooved diamond patterns, tamped concrete with hexagon patterns, acid resistant mastic asphalt and soft rubber mats KSL® (Gummiwerk Kraiburg, Germany). Each floor type was inserted on a 10 m length of the alley three weeks before the locomotion measurements started; thus the cows were accustomed to walk on the different floorings. Concrete slatted floor was used as a control for each floor tested.
Floor measurements. The friction of the different floor materials was measured by a testing machine with a test body made of polyethylene (95 shore hardness) and in a shape of a claw. The test body was vertically loaded with a 200kg weight and was pulled horizontally along the floor by a hydraulic plunger. A load cell placed between the test body and the plunger measured the force needed to pull the body along the floor. The coefficient of friction - static and dynamic - was calculated as the quotient between the horizontal force and the vertical force. During the tests the floor was covered with a thin layer of slurry similar to the preparation of the surfaces for locomotion measurements.

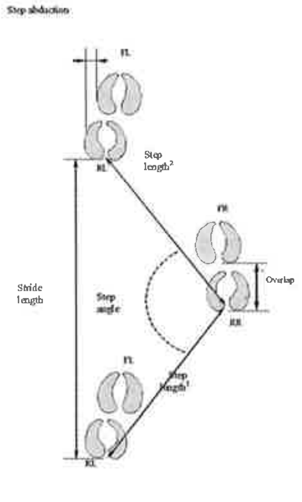

Figure 1. Trackway measurements
Hoofimprints: F=front, R=rear, L=left, R=right

Locomotion measurements. A trackway measurement system (Telezhenko et al., 2002) was used to evaluate cow locomotion on different floors. The walkway was prepared with a small amount of lime powder that was mixed with slurry and spread on the surface in a thin layer.
The cows walked voluntarily through the control and test floor, making tracks on the surfaces. The following parameters from four consecutive strides on each floor were measured with a ruler and an angle-meter (Figure 1):

- stride length - the distance between the two consecutive imprints of the same rear foot;
- step length - the distance between two consecutive imprints of rear feet;
- step asymmetry - absolute value of difference between two consecutive steps (step length1 and step length2);
- step angle - the angle between the lines connecting three consecutive imprints of the rear feet;
- step abduction - the side distance between the lateral edges of one front hoof imprint and the next imprint of the same side's hind hoof. The value is positive if the

rear foot is placed lateral to the front foot;

- overlap - the longitudinal distance between the front hoof imprint and the next imprint of the same side's hind hoof. The trait has a positive value if the rear foot is placed ahead of the front foot.

The time of passing each test course was measured to estimate the walking speed of the cow.

Statistical analysis. In within-subject design the differences between measurements on the tested floors and baseline measurements were analysed with an Analysis of Variance (GLM procedure).

Results

The results of floor friction testing showed minor differences in friction between concrete floorings (Table 1).

Table 1. The coefficients of static and dynamic friction (μ) for the floorings tested, means ± std

Floor	Static friction	Dynamic friction
Slatted concrete	0.404 ± 0.013 a,b	0.298 ± 0.008 a
Smooth concrete	0.410 ± 0.008 a,b	0.308 ± 0.007 a,b
Grooved concrete	0.397 ± 0.009 a	0.322 ± 0.029 b
Tamped concrete	0.415 ± 0.011 b	0.318 ± 0.010 b
Mastic asphalt	0.479 ± 0.009 c	0.405 ± 0.006 c
Rubber mats KSL®	0.602 ± 0.015 d	0.533 ± 0.015 d

Different superscripts indicate values within columns that differ significantly ($P<0.05$)

Mastic asphalt surface demonstrated significantly higher static and dynamic friction than concrete floors, and elastic rubber mats revealed the highest friction properties.

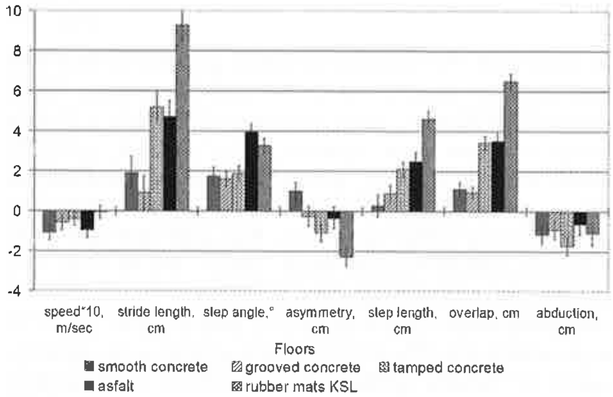

Figure 2. Differences in locomotion between tested solid floors and control floor (slatted concrete)

There was no significant floor effect on walking speed, which made it easier to compare locomotion traits on different floors (Figure 2). The rubber mats had greatest effect on increasing stride and step length. Steps were also less asymmetrical on the rubber mats. Strides and steps on asphalt and tamped concrete with hexagon patterns differed more from slatted concrete floor then smooth and grooved concrete but less then rubber mats. Concrete floors without patterns and with grooved diamond patterns were more similar to the slatted concrete than other floors. Step asymmetry was expressed more on the smooth concrete floor. There was no evidence of flooring influence on step abduction.

Discussion

The floor friction measurements did not reveal any effect of grooves and patterns in solid concrete floor. However, it was demonstrated that coefficient of friction was not an accurate predictor for the slipping risk in livestock (McKee and Dumelow, 1995). Very soft rubber mats KSL®, which are usually used for floors on the lying places, produced rather high friction, but the constant vertical force of testing device did not reproduce the real hoof loading while walking (Phillips, 1993).

Increased step length is a positive sign of cow locomotion comfort. It was shown that on slippery floors cows walk with frequent, short steps (Phillips and Morris, 2001). Positive influence of elastic rubber mats on the step length increase was presented by Benz et al. (2002). Increased overlap on the rubber mats suggested that stride length was increased by greater protraction of the rear limbs. However, the step angle was not increased on the rubber mats to such an extent as the stride did. This suggested that cows kept the posture base quite wide while walking on the rubber mats. It might be influenced by the low friction of the rubber mats in the initial stage of stride. Asphalt and tamped concrete with hexagon patterns had similar characteristics in relation to slatted concrete floor; although the difference in step angle was greater for asphalt floor, which could be explained by higher friction on this floor. Lengthening of strides and steps on the tamped concrete confirm that tamped concrete is more slip resistant then grooved concrete (Albutt et al., 1990). Walking irregularity, described as step asymmetry, was highest on the smooth concrete, which could be indirect indication of higher slipping risk on that floor.

Acknowledgements

This work was carried out as part of the "LAMECOW" project funded through the EC Framework V programme.

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INFLUENCE OF THE HOUSING SYSTEM ON LAMENESS PREVALENCE IN ORGANIC DAIRY FARMING

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Abstract

It was the aim of this study to assess the health status regarding lameness in organic dairy farming in Germany. For this purpose, a representative cross-sectional sample of 53 commercial organic dairy herds was investigated using locomotion scoring for lameness prevalence. The average lameness prevalence was 18% (min: 2%, max: 53%) thus underlining its importance also in organic farming. Cubicle housed herds showed a significantly higher lameness prevalence than herds housed in straw yard systems. Swellings of the carpal and tarsal joints were positively correlated with lameness prevalence. However, within cubicle housed herds, there was no significant effect of the stall surface on lameness.

Introduction

In organic production systems, the housing conditions according to EU legislation and guidelines set by organic farming associations are supposed to be welfare friendly. However, there is evidence that the adaptability of farm animals is often overstressed in organic farming (Sundrum, 2001).
Lameness is regarded as a serious problem in dairy farming due to its welfare relevance and economic importance (Kossaibati & Esslemont, 1997). Whereas lameness prevalence under conventional husbandry conditions has been investigated in several studies, there is little information available about the situation in organic dairy farms. Studies in different eEuropean countries on the most important diseases revealed similar incidences compared with conventional farming (Vaarst et al. 1998, Reksen et al. 1999, Weller & Bowling 2000). More comprehensive analyses taking also environmental parameters such as the housing system into account were carried out in Switzerland (Busato et al. 2000) and Norway (Hardeng & Edge, 2001). However, farm structures and housing systems investigated are not comparable with the German situation.
Therefore the aim of the present study was to determine the lameness prevalence in German organic dairy farming and to investigate possible relationships with the specific housing conditions.

Animals, material and methods

For this purpose, a cross-sectional study was carried out in a representative sample of 53 organic dairy herds during the winter housing period 2002/2003. The housing systems were either cubicle (n=43) or straw yard systems (n=10). Holstein-Friesian was the prevalent breed (n=32), other breeds were Simmental (n=12) and Brown Swiss (n=9). Milk yield did not differ significantly between cubicle housed herds (6,491 kg/a, sd 1,148) and herds housed in straw yard systems (6,055 kg/a, sd 1,798).
In each farm, the locomotion of a random sample of cows, which was adjusted to the herd size, was scored using a 5-point system (Winckler & Willen, 2001). Cows with a locomotion score ? 3 were considered clinically lame. Additionally, leg injuries were recorded using a scheme which took size and severity of the lesions into account. Milk production parameters were obtained from the performance recording systems. Housing design criteria (e.g. dimensions, available space, softness of the stall surface etc.) and management parameters (e.g. health and feeding routines) were recorded using checklists and questionnaires.

Results

At herd level, the average lameness prevalence was 18% and ranged from 2 to 53%. Cubicle housed herds showed significantly more lameness (20%, sd 11.7) than herds in straw yard systems (10%; sd 5,4; $p<0.05$, Mann-Whitney-U, df=1) thus supporting previous studies (Somers et al., 2003). However, the proportion of moderately/severely lame cows was low and did not differ between cubicle housed herds (mean 3%; 0-11%) and straw yard systems (mean 0.4%; 0-2%).
Moderate/severe swellings of the carpal (17%; sd 18.6) and tarsal joints (7%; sd 10.1) were significantly more frequent in cubicle systems than in straw yard systems (carpal joints: 0%; tarsal joints: 0%). The frequency of swollen tarsal joints was significantly correlated with lameness prevalence within cubicle housed herds ($r_s=0.48$; $p<0.01$, n=43).
When cubicle houses were classified according to the softness of the stall surface, there was no difference in total lameness prevalence between highly and less comfortable farms.

Discussion

The results of this study demonstrate that lameness plays a considerable role in German organic dairy farming. However, lameness prevalences are lower than those recorded in conventional dairy farming using the same scoring system (Winckler & Brill 2003). Whereas straw yard systems reportedly have beneficial effects (see also Somers et al. 2003), the importance of other (management) factors seems to be even more pronounced also in (comfortable) cubicle systems. This will be subject to further data analysis.

Acknowledgements

The Federal Agency for Agriculture and Food (BLE), Bonn, is gratefully acknowledged for financial support within the Federal Organic Farming Scheme. We also wish to thank the organic farming associations for help with organizing the field study and the farmers for allowing us to carry out the on-farm investigations and for their great hospitality.

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LAMENESS HERD HEALTH IN A VETERINARY PRACTICE DURING 7 YEARS

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Abstract

Of a mixed practice of 5 veterinarians, of which 4 mainly working as bovine practitioner, the data of the invoice system, which refer to lameness in cattle, are analysed.
Variables are: year, season, month and treating veterinarian.

Main conclusions:

- **Claw lameness and (peri)arthritis occur the most in winter and spring: second half of the housing period and the beginning of the pasture season.**
- **Delivering of medicals, related to use for infectious lameness causes, take place also the most in winter and springtime: chlortetracycline spray and lin comycine spray for topical treatment of dermatitis dig italis and dermatitis interdigitalis and oxytetracycline-HCl powder and lincomycine (+ spectinomycine) powder for application in footbaths.**
- **Cowslips are applied the most in spring and summer.**
- **Over the years from 1993 up to 2000 the quantity of treatments of the veterinary practice were on a constant level, what means a consolidated practice in he field of lameness herd health.**

NUTRITION AND THE BOVINE CLAW METABOLIC CONTROL OF KERATIN FORMATION

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Nutritional management continues to be a major focal point in the attempt to reduce lameness in dairy cattle (Nocek, 1997). Lameness is a multifactorial disease resulting from an array of factors inherent to dairy operations (Lischer and Ossent, 1994). Factors affecting lameness and locomotion include nutrition, feeding strategies, wetness, abrasive or slippery floor surfaces and health events causing production of poor quality horn (fever, age, off-feed, metabolic disturbances, toxins/mycotoxins). A considerable body of evidence is available for the impact of protein, carbohydrates, non-forage fiber, fiber length, and various other macro nutritional management factors pertaining to ruminal function and performance of the dairy cow during the transition period. However, for a long time less emphasis has been placed on the role of hormones, vitamins, minerals, and trace elements and the roles they play in development of quality claw horn and keratin formation.

The objective of this paper is to summarize the processes involved in formation of quality claw horn. Special emphasis is placed on the nutritional and hormonal factors that affect claw keratin formation during the periparturient period and their potential role in production of inferior horn tissue resulting in increased incidence of lameness.

Transition period challenges

Many physiological changes occur in late gestation and early lactation of the dairy cow which affect nutrient uptake and flow. Despite the tremendous quantity of research conducted on nutrition and physiology of transition cows, the transition period remains a problematic area on many commercial dairy farms, and metabolic disorders continue to occur at economically important rates (Burhans et al., 2003). Godden et al. (2003) reported that approximately 25% of cows that left dairy herds in Minnesota from 1996 to 2001 did so during the first 60 DIM, with an uncertain percentage leaving by the end of the lactation as an end result of difficulty during the transition period. These findings are supported by summary data from California indicating approximately 30% of cull cows were leaving dairy herds by 60 DIM (Overton, 2003). Many of these cows suffer from claw abnormalities which occur in early lactation (Green et al., 2002) and may be partly due to the result of nutritional deficiencies or hormonal changes occurring during the periparturient period.

Hormonal Control of Horn Growth. An interesting area of developing research relates to the hormonal control of horn protein production and how changes at parturition may affect potential for future lameness. One of the primary physiological adaptations of transition cows is the need to synthesize and direct glucose to the mammary gland. The cow accomplishes this by concurrently increasing hepatic gluconeogenesis (Reynolds et al., 2003) and decreasing oxidation of glucose by peripheral tissues (Bennick et al., 1972). Vermunt and Greenough (1994) suggested that overfeeding during the dry period, which gives rise to hyperinsulinemia and hyperglycemia (two classic signs of insulin resistance) in early lactation, appeared to predispose cows to laminitis. Green et al. (2002) reported that incidence of first lameness was highest three months after calving, suggesting that factors affecting horn growth during the dry period and in early lactation result in production of inferior horn and subsequent lameness in early lactation.

In research to investigate keratinization control, Hendry et al. (1999) demonstrated that insulin binding was detected in both the epidermal and the dermal layers of explanted bovine hoof tissue. In the early lactating dairy cow, there is a decrease in insulin sensitivity (Cowie et al., 1980) and an inverse relationship between circulating insulin and animal productivity (Hart et al., 1978). Therefore, a decrease in insulin sensitivity, and or concentration, in early lactation could compromise production of claw horn keratin due to depressed uptake of glucose and amino acids (Hendry et al., 1999). It is conceivable that this could be exacerbated if the living (horn producing) epidermis in the claw shares the post-receptor insulin resistance shown by other tissues during the periparturient period (Vernon, 1988).

Epidermal growth factor. Epidermal growth factor (EGF) was reported to have potent mitogenic and anti-differentiative effects in epithelial tissues other than the claw (alimentary and uterine tracts), yet was bound more locally than insulin, being found only in the differentiating epidermal layer (Hendry et al., 1999). During the process of keratinization, epidermal cells rely upon the dermal layers for supply of nutrients. This supply must be provided entirely via diffusion from blood vessels in the underlying dermis because the epidermis is an avascular tissue

(Mülling et al., 1999).

Hendry et al. (1999) reported that EGF may impact keratin formation and result in formation of inferior horn production. EGF stimulated protein synthesis in bovine hoof tissue explants (Hendry et al., 1999), while EGF was reported to decrease keratin expression in healthy equine tissue (Grosenbaugh et al., 1991). Steroid hormones elevated in pregnancy down-regulate local production of EGF in a number of tissues (Plaut, 1993). If this also occurs in the claw the result would be an inhibition of keratin synthesis (Hendry et al., 1999).

Prolactin. Another hormone of particular interest during the periparturient period is prolactin. The major lactogenic hormone prolactin may also influence EGF-dependant keratin deposition (Cowie et al., 1980). Hendry et al. (1999) found hoof explant culture stimulation of protein synthesis by EGF was antagonized to a modest degree by prolactin. Although prolactin itself did not influence hoof protein synthesis, its ability to decrease EGF-stimulated protein synthesis in hoof tissue cultures may be another factor in reducing keratin synthesis during lactation (Hendry et al., 1999).

Glucocorticoids. Goff and Horst (1997) reported that periparturient dairy cows are often subjected to stress with a subsequent increase in cortisol. Glucocorticoids are thought to have an impact on maturation of keratinocytes through regulation of protein synthesis as cortisol affects the metabolism of glucose, protein, and fats (Goff and Horst, 1997). Hendry et al. (1999) found that hydrocortisone inhibited keratin protein synthesis in bovine hoof tissue explants. Epidemiologists have yet to identify a causative relationship between systemic glucocorticoid concentration and laminitis in dairy cows. Yet, it is notable that highly productive herds, which have a greater incidence of laminitis (Nocek, 1997) also have higher glucocorticoid levels (Johnson and Vanjonack, 1976). Milne (1985) reported that steroid treatment of horses exacerbates laminitis. Stress and subsequent elevation of cortisol during the periparturient period and during lactation (Goff and Horst, 1997) may predispose dairy cows to claw disorders resulting from production of inferior claw horn.

Required Nutrients for Keratinization

Amino Acids. The amino acids Cys, His and Met play key roles in establishing the structural integrity of the keratinocyte (Ekfalck, 1990; Ekfalck et al., 1990). Fraser and MacRae (1980) reported that the formation of disulfide bonds between Cys residues was an integral step in the final stage of keratinization and in cornification and establishment of the cellular envelope providing cell wall rigidity and high resistance against a variety of proteolytic enzymes (Elias, 1981). Grosenbaugh and Hood (1993) reported that cultured explants preferentially incorporated 35S-Cys into partially keratinized epidermal lamina as opposed to the uptake of 35S-Met, thus supporting the requirement for Cys in formation of the keratin rich cornified hoof wall.

Amino acid requirements of dairy cattle are not known with much certainty (NRC, 2001). However, the NRC (2001) does suggest that high producing dairy cows may not be able to produce adequate quantities of metabolizable protein to meet the demands of milk production, especially in early lactation when dry matter intake is depressed (Marquardt et al., 1977). This lack of metabolizable protein in early lactation could contribute to insufficient protein synthesis by developing keratinocytes and thus result in production of inferior horn.

Minerals. The onset of lactation places such a large demand on mechanisms of calcium homeostasis that most cows develop some degree of hypocalcemia at calving (Goff and Horst, 1997). This is important in that calcium plays an integral role in the keratinization and cornification process. Calcium is needed for activation of epidermal transglutaminase (TG), which is active in cross-linkage of the cell envelope keratin fibers and in addition is involved in initiation and regulation of the terminal differentiation of the epidermal cells. This enzyme helps activate the final step in the production of the mature squame (i.e. fully cornified keratinocyte) by linking of cell envelope proteins on the cytoplasmic side of the cell wall via glutamyl-lysine bonds to form a cellular envelope of high proteolytic resitancy (Mülling et al., 1999).

Insufficient Ca provided to the maturing keratinocyte due to inadequate vascular supply (Nocek, 1997) or Ca availability due to hypocalcemia may lead to depressed TG activity and formation of dyskeratotic horn. Mülling et al. (1999) reported that differentiating epidermal cells were very sensitive to changes in plasma Ca levels. They suggested inconsistent levels of Ca around parturition, in particular with the onset of lactation, will certainly influence the metabolism in differentiating epidermal cells. This may provide an explanation for the horn rings consistently observed associated with pregnancy in cows. Horst (1986) reported that between 5 and 10% of all milking cows suffer with hypocalcemia during or shortly after calving. Therefore, it may be probable that some of the laminitic insults seen in high producing dairy cows (typically moderately hypocalcemic) and those that have suffered from hypocalcemia may be in part related to impaired TG activity and its impact on terminal differentiation control and formation of the cellular envelope.

Zinc. Zinc has been identified as a key mineral in the processes of keratinization (Smart and Cymbaluk, 1997; Mülling et al., 1999; Mülling, 2000). The ubiquitous distribution of Zn among cells, coupled with Zn being the most abundant intracellular trace element, points to very basic functions. While Zn is a component of over 200 enzyme systems, it has a role in three key functions in the keratinization process: catalytic, structural and regulatory (Cousins, 1996). Catalytic roles are found in enzymes such as RNA nucleotide transferases, RNA polymerase, alkaline phosphatase, carboxypeptidase, alcohol dehydrogenase and the carbonic anhydrases (Cousins, 1996; NRC, 2001). As indicated earlier, the presence of ribonucleic and deoxyribonucleic acid, ascorbic acid, free aldehyde groups and alkaline phosphatase in keratinizing cells serves as a positive indicator of intense cellular activity (Frazer and MacRae, 1980; Hendry et al., 1997). These catalytic enzymes are Zn metalloenzymes and as such are dependant upon Zn as an activator and thus an integral component in the differentiation of keratinocytes.

Zinc also plays a key role in formation of the structural proteins during the keratinization process. Zn-finger proteins are involved in functions requiring protein to protein interactions, most of which are thought to affect cellular differentiation or proliferation (Cousins 1996). Two examples are the transcription factors of retinoic acid and calcitriol (1,25-dihydroxycholecalciferol) receptors (Cousins, 1996),

The third key role of Zn in differentiating cells including differentiating keratinocytes is regulatory. Zinc regulates calmodulin, protein kinase C, thyroid hormone binding and inositol phosphate synthesis (NRC, 2001). Calmodulin is responsible for binding $Ca2+$ and carrying $Ca2+$ into the cytosol of the cell when activated. This is important in the final step of the developing keratinocyte because as noted earlier, calcium activates epidermal transglutaminase. Protein kinase C (which is also calcium dependant) is responsible for phosphorylation of proteins, thus adding available energy to the differentiation process. Thyroid hormone acts to regulate the action of calmodulin and protein kinase C. Inositol phosphate acts to increase $Ca2+$ by mobilizing the ion from intracellular stores, primarily from the endoplasmic reticulum.

Zinc is also required for activation of the cytosolic enzyme Cu/Zn superoxide dismutase (Cu/Zn SOD). In Cu/Zn SOD, Cu functions at the catalytic site, while Zn has a role in the 3-D structure of the enzyme (Cousins, 1996). Cu/Zn SOD is responsible for prevention of lipid peroxidation. Protection of the intercellular cementing substance is critical in maintenance of the structural integrity and biological function of the claw (horn) (Mülling et al., 1998, 1999). Mülling (2000) reported that organic Zn like other trace elements minerals and vitamins is involved in numerous biochemical pathways during keratinisation including formation of keratin protein.,

British workers (Baggott et al., 1988) reported findings of lower Zn concentration in claws of lame cows than those with no history of lameness. Claws of lame cows were also softer than the non-lame individuals. This suggests an insufficiency of Zn or lack of adequate vascular supply to the developing keratinocytes. On dairies with high incidents of foot problems, cows fed 2 to 3 g/d of ZnSO4 for 70 d had fewer claw problems than cows not receiving supplemental Zn (Weaver et al., 1978). In contrast, sheep fed rations supplemented with ZnSO4 for up to 6 months did not show a reduction in claw problems (Cross and Parker, 1981). Inconsistent responses to feeding Zn in the form of ZnSO4 can be attributed to antagonists present in the diet affecting the bioavailability of the Zn (NRC, 2001). Organic sources of Zn, such as zinc methionine, have proven to be more bioavailable than Zn from inorganic sources (Wedekind et al., 1992).

Several studies have shown that complexed Zn improves claw integrity. In a year long study conducted at Illinois State University, cows fed an additional 200 mg/d of Zn from Zn Met had fewer cases of foot rot, heel cracks, interdigital dermatitis and laminitis than cows not fed Zn Met (Moore et al., 1989). Observations on ulcers and white line disease (indications of dyskeratotic structurally altered horn tissue) trended towards improvement. Of beef cattle receiving 540 mg/d of Zn from complexed Zn, 2.45% had foot rot while 5.38% of cattle not receiving complexed Zn had foot rot (Brazle, 1993). These studies indicate that feeding organic Zn complexed to a single amino acid has a beneficial influence on keratinizing tissues, thus improving hoof horn and skin integrity, resulting in improved animal well-being and performance. Zinc requirements for dairy cows vary by stage of lactation (NRC 2001). Milk production creates a significant drain on zinc stores, thus zinc requirements are highest in early lactation (NRC 2001). Insufficient supplies of bioavailable zinc, during the periparturient period and during lactation, may predispose cows to production of inferior horn tissue with a concomitant increase in lameness.

Copper. Much like Zn, Cu is instrumental in activation of enzymes. Copper is needed for activation of cytochrome oxidase enzyme involved in aerobic respiration, lysyl and thiol oxidases for structural integrity of cells, ceruloplasmin, which is essential for absorption and transport of iron for hemoglobin synthesis and superoxide dismutase which works with Zn in reducing the toxic effects of oxygen metabolites (NRC, 2001). Of greatest importance in the keratinizing horn cell is the activity of thiol oxidase (O'Dell, 1990). Copper activates thiol oxidase enzyme, which is responsible for formation of the disulfide bonds between Cys residues of keratin filaments (O'Dell, 1990). This process is essential for structural strength on the cellular level giving rigidity to the keratinized cell matrix.

Cattle suffering from a subclinical Cu deficiency are more susceptible to heel cracks, foot rot and sole abscesses (Puls, 1984). This response may be the result of insufficient cytochrome-c oxidase activity resulting in reduced respiration and oxidative phosphorylation and thus deficient energy supplies for differentiating keratinocytes (Linder, 1996). Heel cracks and abscesses may also be the result of insufficient Cu availability for activation of Cu/Zn SOD resulting in increased fragility of cell membranes. Unsaturated lipids in the cell periphery are particularly vulnerable to oxidative damage (Linder, 1996). The intercellular lipids are an integral part of the cementing substance responsible for cell-to-cell adhesion (Mülling and Budras, 1998). Therefore, any nutrient deficiency that leads to production of inferior intercellular cementing substance or predisposes it to excessive oxidative damage may potentially lead to production of dyskeratotic horn tissue with increased susceptibility to cracking and wear.

Selenium. Selenium is a constituent of the enzyme glutathione peroxidase. Glutathione peroxidase is responsible for reduction of H2O2 and free O2 to H2O (NRC 2001). By acting much like Cu/Zn SOD, glutathione peroxidase plays a role in protecting both the intra and extra-cellular lipid membranes against oxidative damage. This way Se may contribute to protection and maintenance of physiological function of the lipid rich intercellular cementing substance.

Excessive supplementation of Se may be damaging to developing keratinocytes. Selenium in the form of selenomethionine (SeMet) is readily absorbed by the same mechanism as Met (Combs, 2000). Inorganic Se absorption does not appear to be regulated and is quite high (>50%, NRC, 2001). Bodily storage of inorganic Se from selenite or selenate occurs as seleno-amino acids SeCys and SeMet. Combs (2000) indicated that the most biologically efficacious of these is the SeMet form, yet SeCys is also very active. Combs (2000) reported that Se and/or seleno-amino acids may be preferentially incorporated into sulfur requiring AA sites during protein production and thus change the integrity of the protein structure.

Larson et al. (1980) reported that dairy cows supplemented with 50 mg of injectable Se (over 6.6 x NRC requirement) during the dry period suffered severe claw problems in the postpartum period. They indicated that between 48 and 69% of cows receiving the supplemental Se injection had increased lameness, sore feet, deformed claws and loss of hair from the tail versus 28 to 30% claw problems in non-supplemented cows. It is very likely that the excessive Se supplement was incorporated into keratin fibers of the maturing keratinocytes with the key Cys and Met sites replaced by SeCys or SeMet. During this process critical disulfide bridge formation may have been reduced or inhibited during the cornification process creating inferior hoof horn lacking structural rigidity with poor integrity. The recommended level of Se in dairy diets is 0.3 mg/kg DM and should be closely monitored to insure over supplementation does not occur, especially during the dry and early lactating periods (NRC, 2001).

Manganese. Manganese plays an indirect role in the keratinization process. Manganese helps minimize feet problems by maintaining proper leg formation (Miller et al., 1988). Manganese is needed for activation of galactotransferase and glycosyltransferase enzymes, which are needed for the synthesis of chondroitin sulfate side chains of proteoglycan molecules (Keen and Zidenberg-Cherr, 1996; NRC, 2001). Proteoglycans are essential building blocks in formation of normal cartilage and bone. Animals suffering from a Mn deficiency will exhibit skeletal abnormalities, crooked legs and shortening of tendons as noted by knuckling over of feet (NRC, 2001).

Manganese also plays a role in activation of other critical enzyme systems, such as pyruvate carboxylase, an enzyme that catalyzes the first step of carbohydrate synthesis. This process is responsible for gluconeogenesis and production of cellular energy an essential component in production of quality horn tissue (Keen and Zidenberg-Cherr, 1996). Similar to Cu/Zn SOD, Mn plays a role in activation of Mn superoxide dismutase (Mn SOD) and the removal of superoxide free radicals. Therefore, Mn SOD may play a protective role for the lipids involved in cementing together mature keratinocytes.

Combinations of trace minerals. There are significant interactions between trace minerals and hence it is imperative that nutritionists formulate rations to maintain an appropriate balance of trace minerals in order to maximize animal performance. Research has demonstrated that supplying a combination of complexed trace minerals is more beneficial to claw integrity than supplying a sole complexed trace mineral because of synergistic effects. A two year study conducted on five commercial dairy herds in Central New York indicated that cows fed 360 mg complexed Zn, 200 mg complexed Mn, 125 mg complexed Cu and 25 mg complexed Co, in combination with inorganic trace minerals, resulted in better claw integrity than cows fed only 360 mg of complexed Zn or no complexed trace minerals (Nocek et al., 2000). Supplementation of the diet with a combination of complexed trace minerals reduced the incidents of double soles, white line separation, digital dermatitis, sole hemorrhages and ultimately, sole ulcerations (Nocek et al., 2000). In addition, three hundred cows on a large commercial dairy in Florida were fed a combination of complexed Zn, Mn, Cu and Co to evaluate claw health (Ballantine et al., 2002). Cows fed complexed trace minerals tended to have fewer incidents ($P<0.15$) of claw disorders than cows fed inorganic trace minerals at 75 days postpartum (23.6 vs. 34.1%) and numerically lower incidents at 250 days postpartum (10.0 vs. 17.7%). Feeding complexed trace minerals reduced incidents ($P<0.15$) of white line disease at 75 (9.5 vs. 14.6%) and 250 days postpartum (4.9 vs. 8.8%). Feeding complexed trace minerals during the late dry period and during early lactation improved ($P<0.05$) claw lesion scores. These results indicate that if cows fed complexed trace minerals did develop a claw lesion, the lesion was less severe, as measured by size and painfulness of the lesion, as compared to control cows that developed a claw lesion.

Role of vitamins in horn production/formation

Vitamin A. Vitamins also play an integral role in developing the structure and quality of keratinized horn tissue. Vitamin A is needed for cell differentiation (Olson, 1996). Differentiating cells have specific binding sites for vitamin A and once bound can both stimulate or inhibit gene expression. Vitamin A is needed for normal growth and development and for maintenance of skeletal and epithelial tissues (NRC, 2001). The role of vitamin A in keratinizing cells is tied to its action in gene expression (NRC, 2001).

Vitamin D. One of the most important biological regulators of calcium metabolism is vitamin D (synonym calciferol) (NRC 2001). Derived from cholesterol, a Mn dependant process, vitamin D is responsible for minute-by-minute calcium and mineral homeostasis. In its biologically active form 1,25(OH)2D3, vitamin D is required for control of Ca2+ re-absorption, absorption and mobilization/accretion from bones (Norman, 1996). Because the body can endogenously produce vitamin D3 and because it is retained for long periods of time in vertebrate tissues, it is not likely that dairy animals would be deficient in vitamin D. However, with increased confinement and reduced exposure to direct sunlight, dairy animals lacking sufficient supplementation could succumb to minor vitamin D deficiencies. Therefore, any lack of vitamin D will certainly impact calcium metabolism and thus affect the keratinization process.

Vitamin E. The best understood role of vitamin E is as a lipid-soluble cellular antioxidant (NRC, 2001). Via this function and possibly others, vitamin E is involved in maintenance of cellular membranes. This function may be important to the integrity of keratinized tissues as the intercellular cementing substance is composed of lipid rich material (Mülling et al., 1999). A deficiency of vitamin E at the cellular level is generally accompanied by an increase in lipid peroxidation of cellular membranes (Sokol, 1996). This may lead to deceased energy production by mitochondria, oxidation and mutation of DNA and alteration of normal transport processes of the plasma membrane (Sokol, 1996). Therefore, cells exposed to oxidative stress (i.e. a laminitic insult) will show more rapid injury and necrosis when rendered vitamin E deficient (Sokol, 1996). This may also help explain why transition dairy cows fed low levels of vitamin E and subjected to undue stress at parturition incur higher than normal levels of lameness and production of poor horn tissue (Nocek, 1997).

Biotin. A water-soluble "B" vitamin, biotin is possibly the vitamin of greatest importance to the keratinization process. Biotin is essential for the formation and integrity of the keratinized tissues (skin, hair, claws and footpads) in mammals and birds (Maynard et al., 1979). Biotin is a cofactor for enzymes used in a diverse array of metabolic pathways. Amino acid metabolism, cellular respiration, gluconeogenesis and lipogenesis involve enzymes that require biotin (Mock, 1996). Four biotin-containing enzymes are found in mammalian cells: acetyl-CoA carboxylase, B-methylcrotonyl-CoA carboxylase, propionyl-CoA carboxylase and pyruvate carboxylase. All four enzymes require biotin to become activated (Weiss and Zimmerly, 2000).

With regard to keratin formation, biotin-dependent enzymes are directly involved in synthesis of lipids and glucose with particular importance placed on synthesis of long-chain fatty acids (Meyer et al., 1998; Weiss and Zimmerly, 2000). Mülling et al. (1999) demonstrated that biotin was essential for formation of the complex lipid molecules in the intercellular cementing substance. He also demonstrated, in biotin deficient calves, that biotin deficiency affected keratinizing epidermal cells as well as composition of the intercellular cement (Mülling et al., 1997). Research in pigs and horses has shown that biotin positively influenced the integrity of the hoof horn (Geyer, 1998).

Functioning ruminants are able to produce biotin in the rumen. However, high grain (> 50% of DM) rations reduce ruminal synthesis of biotin in-vitro (DaCosta-Gomez et al., 1998). This response may be due to an insufficient conversion of lactate to pyruvate. Mock (1996) reported that biotin deficiency was tied to insufficient pyruvate carboxylase activity resulting in cellular lactic acidosis. It may be possible that ruminants receiving proportionately high grain diets lack sufficient biotin in their rumen to convert lactic acid to pyruvate and then oxaloacetate, thus predisposing them to lactic acidosis. Nocek (1997) reported lactic acidosis as one of the possible contributing factors in lameness of dairy cows. Recent works (Fitzgerald et al., 2000; Hedges et al., 2001; Weis and Zimmerly, 2000) indicate that dairy cows respond favorably (improved claw integrity and reduced lameness) when provided supplemental biotin (20 mg/hd/d) for a period of greater than 6 mo. In a study of five dairies with a total of 900 cattle, Pötzsch et al. (2003) reported biotin supplemented at 20 mg/d for longer than 6 mo reduced white line disease in multiparous cows by 45% to 8.5 cases per 100 cow years. However, the effect of biotin in primiparous cows was not significant. These studies indicate that biotin reduced the incidents of white line abnormalities in particular and other claw diseases such as sole hemorrhage, sole ulcers, digital dermatitis, and heel erosion.

Mülling et al. (1999) proposed the analogy of building a brick wall to the effect of supplements such as biotin on hoof keratin formation. Zinc is needed for activation of the enzyme systems needed for formation of sound cellular structure, while biotin is needed for production of the intercellular cementing substance. The two together allow the keratinizing squamous cells to generate stronger horn with greater integrity that will better withstand environmental stresses. It is this ability to withstand environmental stress that ultimately determines the productivity and potential profitability of the animal.

Cconclusions

Formation of keratin proteins is an essential/crucial part of a systematic process of cellular changes that transform living, highly metabolic active living epidermal cells into dead structural horn cells with no metabolic activity. This differentiation of epidermal cells is very complex and very sensitive to hormonal controls, nutrient flow and environment. It is the process of nutrient flow, as impacted by hormonal controls that plays an important role in determining the quality and integrity of keratinized tissues of the horn. When nutrient supply to keratin forming cells is compromised or completely altered, inferior keratinized tissue is produced. Inferior tissue increases the potential for development of claw disease and may ultimately lead to lameness. Calcium, Zn, Cu, Mn, vitamins A, D & E and biotin all play important roles in production and maintenance of healthy keratinized tissues. Increasing the bioavailability of trace minerals, especially Zn, Cu and Mn improves their utilization and thus contributes to improved integrity of keratinized tissues such as skin and claw. Integrity of claw horn is one prerequisite for claw health which in turn is the prerequisite for overall animal well-being, productivity and potential profitability.

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EFFECT OF AN INCIDENT OF OVER FEEDING OF CONCENTRATE ON CLAW HORN LESION DEVELOPMENT IN FIRST LACTATION DAIRY HEIFERS

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Introduction

Ruminal acidosis is widely considered to be a primary risk factor for the development of claw horn lesions in dairy cattle. It is hypothesised that vasoactive substances resulting from disturbed rumen function lead to vascular reactivity, laminitis, hoof deformation and finally and abnormal horn synthesis thereby compromising hoof health.

Materials and methods

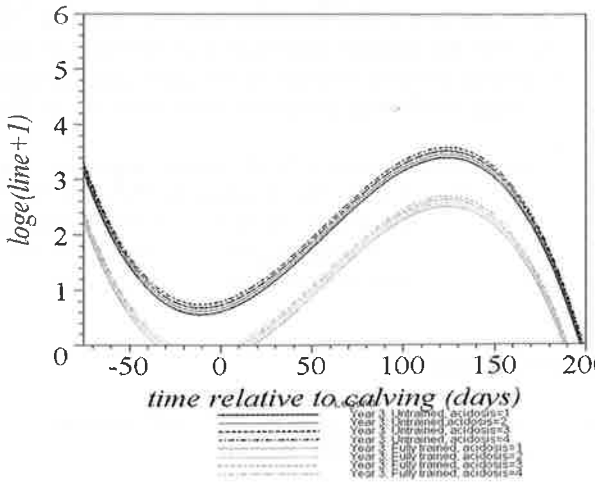
Claw horn lesion development was observed in a group of 22 spring calving first lactation Holstein Friesian heifers. These animals were part of an observational lameness study looking at the effect of training heifers to use cubicles before calving on hoof health after calving (Logue et al., 2004). All but 2 were housed in a lactating group when they were accidentally overfed concentrates within a TMR (almost twice ration) early in their first lactation (on average 11 weeks post calving, range 4 - 15 weeks). The exact amount of concentrate given to the animals could not be identified but the error occurred because the normal farm mixer wagon (with limited sensitivity of + or - 60 kg) was used to weigh 90kg of barley and 65 kg of blended concentrate for a group of 20 animals. The animals showed signs of ruminal acidosis, some more severely than others. Severely affected animals were moved to a straw yard and given veterinary attention. Rumination ceased in the majority of animals (14/22), seven of the 22 were given Vitatrace (Vetoquinol UK Ltd); five became recumbent and required intravenous fluids. All animals recovered and were returned to the cubicle house within 2 - 4 days. Since animals were already part of a lameness study detailed claw horn lesion data existed before the animals went sick. Routine examinations continued throughout lactation and again at housing (October) and 3 months post housing (January) for any long-term effects of the feeding incident.
After the feeding incident the cows were treated and for analysis purposes were split into four groups by severity of clinical signs and (treatments given) as given in Table 2.

Table 2 Definition of 'illness score' according to severity and numbers of animals affected.

Illness score	Definition	N (trained, untrained)
1	Still ruminating	6 (2,4)
2	Not ruminating	5 (4,1)
3	Requiring Vitatrace, not ruminating	4 (3,1)
4	Recumbent, requiring IV fluids and Vitatrace.	5 (3,2)

Results

The initial models applied to the data are detailed in Logue et al. (2004). For this study acidosis score was included in the model and so lesion data were modelled as a function of training regime, days relative to calving, the time of year, optionally, days an straw, and severity of the acidosis.
There was significant benefit from fully training for sole lesions (P=0.006, and P=0.012, for the models excluding and including days on straw, respectively). For line lesions it was significant for the model excluding days on straw (P=0.030) and marginally insignificant when including days on straw (P=0.091). Inclusion of the acidosis score to the models reduced the respective significance of the treatment effects to P=0.013, and P=0.015 for sole lesions and P=0.060, and P=0.144, for white line lesions.
The inclusion of acidosis score in the models slightly reduces the between cow variance component for sole lesions, but for line lesions it is increased slightly. In all cases the coefficient was positive (for line and for sole for the models excluding days on straw and for line and for sole for the models including days on straw) indicating a very marginal increase of line and sole lesions with increasing acidosis score (see figure 1). Acidosis score, however, was not statistically significant for any model considered.



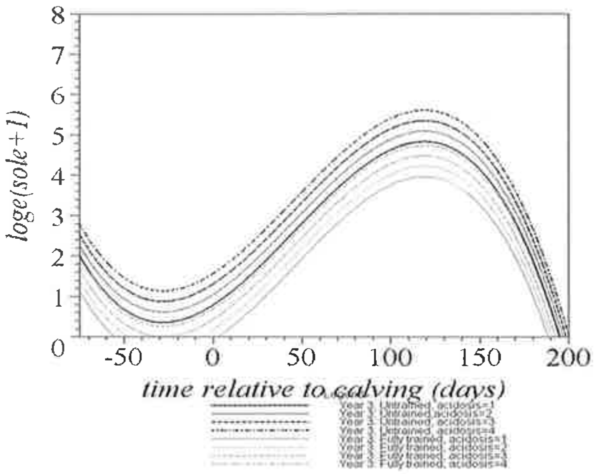


Figure 1. Effect of acidosis score on white line and sole lesions with time relative to calving

Discussion

The lesion scores observed in this group of heifers were much lower than expected given the acute response to overfeeding and the commonly held association between this and laminitis. True laminitis (acute lameness, increased blood flow and heat in the feet) was not observed in any of the affected animals despite the acute symptoms. However, treatment was very prompt and the overfeeding occurred only once and not over a prolonged period. While this final year had other confounding factors eg. a later calving period (due to breeding delays caused by the FMD outbreak) the general level of lesions observed was quite similar to previous years (Logue et al., 2004) .
The feet of this group of animals were examined again at autumn housing and again three months later to see if the feeding incident had had any effect on the long term ability of the hoof capsule to withstand the challenge of housing on concrete from grass. However lesion scores at housing (when animals were in late lactation) were generally low. Three months later very few even superficial lesions were seen suggesting that there was no long term damage to the feet resulting from the acidosis. It may be that the prompt treatment and removal of the more severely affected animals to the straw yard limited any damage which may otherwise have occurred to the feet.
This finding raises some difficult issues regarding the importance of acidosis in the development of claw horn lesions in dairy cows. It also demonstrates the tolerance of the cow's feet to poor feeding practice given good husbandry and ideal environmental conditions.

Acknowledgements

We gratefully acknowledge farm staff. SAC receives financial support from SEERAD

Reference:

Logue, D.N., Offer J. E. Brocklehurst, S. (2004) Proc Int Conf Lameness in Domestic Ruminants pp

THE INFLUENCE OF AN ORGANIC TRACE MINERAL SUPPLEMENT ON THE FREQUENCY OF CLAW LESIONS IN HOLSTEIN DAIRY COWS

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Introduction

Lameness continues to be a major area of concern for dairy managers. While nutrition is often blamed for laminitic insults, it may also have a positive impact on lameness management. Previous workers have indicated that supplementing dairy cows with organically complexed trace minerals will help improve bioavailability and aid in the reduction and alleviate the severity of claw disorders in dairy cattle (Moore et al., 1989; Nocek et al., 2000; Ballantine et al., 2002). The objective of this study was to evaluate the effects of supplementing metal specific amino acid complexes on the incidents and severity of claw disorders in dry and lactating dairy cattle.

Materials and Methods

Animals. A total of 157 pregnant Holstein cows were blocked by parity, milk production and season of calving and randomly assigned within block to one of two dietary treatments: daily supplementation with 443 mg of Zn, 444 mg of Mn, 261 mg Cu, and 25 mg of Co as inorganic salts (control) or a combination of inorganic salts and complexed trace minerals (treatment, 14 g of 4-Plex /day, containing 360 mg Zn from zinc methionine, 200 mg Mn from manganese methionine, 125 mg Cu from copper lysine, and 25 mg Co from cobalt glucoheptonate, Eden Prairie MN, USA). Trace mineral supplementation was equal between treatments. Diets were fed from -60 d from projected calving date through 250 d postcalving. Dry cow diet composition on a % of dry matter basis: corn silage (32.1%), grass hay (30.3%), mixed haylage (5%), and a grain mix containing the treatment supplement (32.6%). Lactating cow diets on a % of dry matter basis contained; corn silage (32.2%), mixed haylage (12.8%), dry cow grain treatment supplement (11.8%), corn grain (7.9%), citrus pulp (8.1%), whole cottonseed (5.7%), soyhulls (5.9%), protein/mineral mixture (15.6%). To be included in the study, cows had to be on the precalving ration for at least 60 days prior to calving.

Housing and Management. During both the pre and post-calving periods, cows were housed in cubicles (free stalls) lined with a surface of chopped rubber covered with a nylon mesh sheet bedded with sawdust. Cows were milked 2 X/day. All feed ingredients were weighed and mixed together into a TMR and fed once a day prior to and after calving. Mineral supplements were combined with corn grain and pelletized. Diets were formulated to NRC (2001) standards for energy and protein for late gestation, nonlactating cows and lactating cows producing 30 or 40 kg of milk, depending on stage of pregnancy and lactation.

Claw Examination. Examination of claws and soft tissues surrounding the claws was made by a trained clinician at -60 d, 30 d, and 250 d relative to calving. After cleaning, claws and soft tissues around the claw and interdigital area were examined for lesions. Lesions were classified according to Toussaint Raven (1989) based on macroscopic examination of claw and tissues. Lesions were classified as involving the keratinous tissue or soft tissue. The keratinous lesions were classified as follows: dorsal wall ridges, erosion of the heel bulb, abaxial wall lesions, double sole, white line separation, sole abscess, sole hemorrhage, sole ulceration or sole erosion. The lesions of the soft tissues surrounding claws were classified as: digital dermatitis, pododermatitis of the digit or interdigital area, interdigital fibroma, or hairy heel warts. Lesions were mapped for location on the claw and surrounding soft tissues.

Statistical analysis. Claw lesions were analysed as repeated observations with PROC GENMOD (SAS, Raleigh NC, USA) and subject = cow (claw) treated as the nested term for the covariance matrix. Claw and tissue were analyzed using the logistic link function. Multiple lesions were analyzed using the cumulative logistic link function and the multinomial distribution.

Pearson's correlation coefficient was used to examine the correlation between lesions on the same claw and lesions on the same cow between claws and feet.

Results

A total of 157 cows were assigned to the study and began the experiment. Due to early calving, defined as less than 60 days on precalving diets, or health complications (abortion, mastitis, etc.) only 138 cows were included in the final analysis (73 cows on control, 65 cows on treatment diet). At thirty days postcalving, two cows were removed for health reasons, resulting in 136 cows at 30 d postcalving (73 cows on control, 63 cows on treatement diet). At 250 days postcalving, 9 cows on the control diet and 6 cows on the treatment diet were removed prior to sampling, resulting in 121 cows for sampling at 250 d postcalving.

Heel erosions were the most frequent claw lesion across all treatments and time periods (45%). The second most common lesion were abaxial wall lesions (26.7%).
Only 18 (4.6%) of cow examinations across all periods had no lesions. Erosions were distributed uniformly between front and rear feet and medial and lateral claws (Table 1). Dermatitis of the soft tissue was more often observed in the rear feet (Table 1).

Many cows had multiple lesions. A total of 3160 claw examinations were made. Claw lesions were classified as involving keratinous tissues (% of claw lesions observed) including heel erosions (44.4%), abaxial wall lesions (28.3%), sole erosion (7.9%), white line separation (3.4%), double sole (2.75%), dorsal wall ridges (1.59%), solar abscess (1.2%), sole hemorrhage (0.1%), sole ulceration (1.1%). Lesions of the soft tissues surrounding claws included digital dermatitis (12.4%), interdigital dermatitis (6.2%), pododermatitis (0.03%), hairy heel warts (0.2%) and interdigital fibroma (0%). Sixty eight percent of the claws were identified to have one or more lesions. Observations identified 39.6% of claws had one lesion, 19.2% had two lesions, 6.5% had three lesions, 2.2% had four lesions, 0.5% had five lesions, and 0.2% of claws had six or more lesions.

Overall, supplementation with complexed trace minerals did not affect claw or foot health ($P<0.31$), yet supplementation had an effect on reducing solar lesions and had a tendency to reduce claw lesions. The incidence of claw lesions (lesions involving the keratinous tissues) tended to be lower for the complexed trace mineral supplemented group (61.5% versus 65.2%, $P<.08$). This was largely due to a reduction in solar lesions at 30 days postcalving in supplemented cows. Significant factors influencing claw lesions included lactation number ($P=0.002$), phase ($P<0.001$; time relative to calving), season ($P<0.001$), and the interactions of treatment*phase ($P=0.03$) and treatment*phase*lactation ($P<0.001$). In addition, lactation*season ($P<.002$) and phase*season ($P<.0001$) were significant factors describing claw lesions.

Results of this study indicate that supplementation of dairy cattle, beginning in the dry period, may help reduce the number of claw lesions in the subsequent lactation. However, lactation number, stage of lactation and season had a greater influence on the number and severity of claw lesions.

Table 1. Lesions by claw and foot of cows that had at least one claw or foot with the condition from a total of 395 cow examinations from 138 cows made over three periods at -60d , 30 d and 250 d relative to calving.

Claw/foot	Heel and sole erosion	Claw lesion associated with laminitisa	Dorsal wall ridge, abaxial wall lesion and fissure	Normal claw or footb	Infectious tissue lesionc
LF inside	184	17	56	0	
LF outside	153	24	44	153	
LF foot					21
RF inside	182	16	45	151	
RF outside	148	24	40	158	
RF foot					26
LR inside	168	16	55	135	
LR outside	102	40	23	74	
LR foot					177
RR inside	165	18	56	137	
RR out-	173	33	22	73	
RR foot					173

a Double sole, White line separation, Solar abscess, Sole hemorrhage, and Sole ulceration
b No claw or foot lesions on any claw
c Digital dermatitis, Interdigital dermatitis, Pododermatitis, Hairy Heel Warts, and Fibroma

References available upon request.

EFFECT OF FEEDING COMPLEXED TRACE MINERALS TO HEIFERS FROM 12 MONTHS OF AGE TO ONE MONTH PREPARTUM ON RISK OF DEVELOPING CLAW LESIONS DURING LACTATION AND LACTATION PERFORMANCE

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Introduction

Research has shown that lameness contributes to reduced milk production (Guard, 1997; Robinson et al., 2003) and reproductive failure (Sprecher et al., 1997; Hernandez et al., 2000: Melendez et al., 2002). Furthermore, research indicates that previously lame cattle are more prone to future reoccurrences (Nocek, 1997). Therefore, preventing animals from becoming lame must be a key management objective. Supplementing cows with a combination of complexed zinc, manganese, copper and cobalt has been shown to reduce incidence (Nocek et al., 2000; Ballantine et al., 2002) and severity of claw lesions (Ballantine et al., 2002). However, the effect of complexed trace mineral supplementation during the rearing phase on claw lesion incidence has not been examined. The objective of this study was to determine the effect of feeding heifers a combination of cobalt glucoheptonate and zinc, manganese and copper amino acid complexes (Availa®4) on incidence of claw lesions both during the rearing phase and in the first lactation.

Materials and methods

Five hundred seventy-two heifers at a commercial heifer rearing facility were alternatively assigned to one of two dietary treatments: control diet (DM basis, 66 ppm Zn, 66 ppm Mn, 18 ppm Cu and 0.52 ppm Co) or control diet plus daily 360 mg zinc, 200 mg manganese and 125 mg copper from amino acid complexes and 12 mg cobalt from cobalt glucoheptonate (CTM; Availa-4, Zinpro Corporation, Eden Prairie, MN). Treatments were fed from 12 months of age until one month prepartum. Heifers originated from one of four source dairies and were returned to the source dairy at one month prepartum

Heifers were housed in open earthen-mound lots without overhead protection and fed a total mixed ration (TMR) consisting of 98.1% forage and 1.9% concentrate. Diets were formulated to contain (DM basis) 15% CP, 30% ADF, 40% NDF, 64% TDN, 1.0% Ca, 0.36% P, 0.28% Mg, 0.22% S and 145 ppm Fe. Pen feed intakes were monitored daily. Diets were reformulated weekly based on DMI changes, and bimonthly based upon changes in diet ingredients.

Claws evaluations were conducted prior to initiation of treatment, at approximately one month prepartum and at two months postpartum by one claw trimmer using a clean, light grind. The claw trimmer was a graduate of the Dairyland Hoof Care Institute (Baraboo, WI) and was not informed of the heifers' treatment assignment. Lesions were noted in the seven zones of the claw and each lesion was scored for severity on a scale of 1 to 3 (1=minor, 2=moderate, 3=severe). To assess both incidence and severity of claw lesions, a claw lesion incidence and severity (CLIS) index was calculated. This index was the average number of zones affected per cow multiplied by the average severity score multiplied by 10. After returning to the source dairy, heifers were housed in naturally ventilated free stall barns, fed similar TMR with respect to the source dairy and milked 3X/d. Results of first lactation performance were collected via production record systems on each dairy by the researchers as heifers completed or neared completion of their first lactation. Due to removal of some of the trial ID tags of heifers at one dairy after completing the claw evaluation at 2 months postpartum, lactation performance data were available from only 421 heifers.

Production data and CLIS index measurements were analysed using the MIXED procedure of SAS V8.1 (1999) with the effects of dietary treatments and source dairies as discrete, class variables, and CLIS index in phase one and its interaction with dietary treatments as continuous variables.

Result and discussion

Feeding CTM increased ($P < 0.05$) the CLIS index for sole hemorrhages and tended to increase ($P < 0.15$) the CLIS index for claw lesions and heel erosion at one month prepartum (Table 1).

Table 1. Effect of feeding complexed trace minerals[a] to growing heifers on incidence and severity of claw lesions as measured by the claw lesion incidence and severity index[b] during the rearing phase and subsequent lactation.

Claw Disorder	1 Month Prepartum		2 Months Postpartum	
	Control	Complexes	Control	Complexes
Overall claw lesions	3.8^{w}	4.8^{x}	6.6^{w}	5.5^{x}
Dorsal wall ridges	0.00	0.01	0.6	1.2
Heel erosion	29.4^{w}	34.5^{x}	36.8	31.5
Abaxial wall fissures	0.7	0.8	2.6	2.6
Double soles	0.2	0.3	0.3	0.0
White line separationc	2.8	3.0	7.5^{y}	4.9^{z}
Sole hemorrhages	9.7^{y}	16.7^{z}	29.2	24.1
Sole Ulcers	--	--	0.5^{w}	0.1^{x}
Digital dermatitis	1.5	0.0	1.6	0.0
Interdigital dermatitis	0.2	0.2	0.2	0.2

a Availa®4 provided daily 360 mg Zn, 200 mg Mn, 125 mg Cu and 12 mg Co

b Calculated using the following formula (number of zones affected per cow x average severity score x 10); Severity score ranged from 1 (minor) to 3 (severe)

c Slope of P3 on P1 for control heifers was greater than 0 ($P < 0.05$) indicating a positive relationship between the CLIS index at 12 months of age and the CLIS index at 2 months postpartum

wx Within phase, within row, means with uncommon superscripts differ ($P < 0.15$)

yz Within phase, within row, means with uncommon superscripts differ ($P < 0.05$)

However feeding CTM from 12 months of age to one month prepartum reduced ($P < 0.05$) the CLIS index for white line separation and tended to reduce ($P < 0.15$) the CLIS index for claw lesions and sole ulcers two months postpartum.
For heifers fed the control diet, there was a positive relationship ($P < 0.05$) between the CLIS index one month prepartum with the CLIS index two months postpartum for heel erosion, abaxial wall fissures and digital dermatitis (Table 2).

Table 2. Effect of feeding complexed trace mineralsa to heifers during the rearing phase on the regression coefficients of the relation between claw lesion incidence and severity indexb at 1 month prepartum and at 2 months postpartum.

Claw Disorder	Control	Complexes
Overall claw lesions	0.067	0.150
Dorsal wall ridges	---	-0.008
Heel erosion	0.164z	0.040
Abaxial wall fissures	1.311z	0.268
Double soles	-0.073	0.010
White line separation	-0.003	-0.000
Sole hemorrhages	0.234	0.368z
Digital dermatitis	0.164z	0.209z
Interdigital dermatitis	-0.001	-0.013

a Availa®4 provided daily 360 mg Zn, 200 mg Mn, 125 mg Cu and 12 mg Co

b Calculated using the following formula (number of zones affected per cow x average severity score x 10); Severity score ranged from 1, minor to 3, severe

z Differs from zero ($P < 0.05$)

These results indicate that if control heifers had heel erosion, abaxial wall fissures or digital dermatitis one month prior to calving, they were more likely to have heel erosion, abaxial wall fissures and digital dermatitis two months post calving. For heifers fed CTM, there was a positive relationship ($P < 0.05$) between the CLIS index one month prepartum and the CLIS index two months postpartum for sole hemorrhages and digital dermatitis.
The regression coefficient for the relationship between presence of claw lesions during the rearing phase and milk yield in the first 60 to 90 days of lactation was greater than zero ($P < 0.15$) for CTM heifers, indicating that if heifers had a claw lesion during the rearing phase, feeding CTM tended to increase production in the first 60 to 90 days of lactation (Table 3).

Table 3. Effect of feeding complexed trace mineralsa during the rearing phase on the regression coefficients of the relationship between claw lesions and lactation performance

Item	Control	Complexes
Milk yield, first 60 to 90 days of lactation	-10	20z
Milk yield, 305 Mature Equivalents	-5503z	-1901
Fat yield, 305 Mature Equivalents	-19.65	-38.80
Protein yield, 305 Mature Equivalents	-147.55	57.34

a Availa®4 provided daily 360 mg Zn, 200 mg Mn, 125 mg Cu and 12 mg Co

z Differs from zero ($P < 0.15$)

The presence of a claw lesion two months postpartum tended to ($P < 0.15$) reduce the 305 mature equivalent milk yields of heifers fed the control diet during the rearing phase.
Results of this study indicate that feeding CTM to heifers did not reduce incidence of claw lesions during the rearing phase. However, feeding CTM to heifers during the rearing phase did reduce incidence of claw lesions at two months postpartum and helped alleviate the effects of claw lesions on lactation performance.

Acknowledegements

The authors would like to acknowledgement Emerald Lane Farms, Miltrim Farms, Quella Farms, Bredl Farms, Badgerland Holsteins, Harlan Tripp, Denise Tripp and Ryan Wernberg for their diligence in carrying out the trial protocol

REFERENCES AVAILABLE UPON REQUEST

INFLUENCE OF BIOTIN SUPPLEMENTATION ON HOOF CHEMICAL COMPOSITION AND RATES OF WEAR AND GROWTH IN LONG FED F1 WAGYU/BLACK ANGUS STEERS

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Introduction

Daily biotin supplementation of dairy cows has frequently improved hoof integrity (Fitzgerald et al 2000). However, relatively little is known about changes in growth rates of hooves in intensively long fed beef steers. Such animals are fed high grain based diets for long periods (>300 days), exhibit high live weights (>700kg) and contend with periodic muddy conditions. Biotin deficiency has been associated with feeding grain-based diets to cattle by compromising ruminal biotin synthesis (Fitzgerald et al 2000). An experiment was conducted to determine the effects of biotin supplementation at three levels (0, 10, 20mg/hd/d) on hoof chemical composition and rates of wear and growth in F1 Wagyu/Black Angus beef steers fed long term on a dry rolled wheat based ration.

Material and Methods

The experiment, conducted in Queensland, Australia began in March 2001 and consisted of twelve pens of nine F1 Wagyu/Black Angus steers per pen (live weight 410.5kg, SD 24.4). Pens were allocated to one of three biotin treatments, including nil biotin (Cont), 10mg/hd/d (B10) and 20mg/hd/d (B20) in a randomised complete block design with four replicates. Biotin was administered to pens within a mineral supplement mixed into a wheat-based ration at a rate of 250g/hd/d.

A subset of 36 steers was selected for hoof assessment. Steers were weighed and hoofs assessed for growth and wear (mm) on seven measurement periods (MP), including days 0-55 (MP1), 55-111 (MP2), 111-167 (MP3), 167-188 (MP4), 188-251 (MP5), 251-301 (MP6) and 0-301 (MP7), using method described by Tranter and Morris (1992).

Steers were despatched for slaughter after approximately 430 days on feed (live weight of 844.3kg, SD 54.2). At slaughter, all front hooves were collected for determination of white line width (lateral and medial), lesion frequency (sole and white line) and fatty acid composition (fat pad and white line). Results were analysed by analysis of variance for a randomised complete block design.

Results

Biotin supplementation had an effect on hoof growth (P=0.09) and wear (P=0.12) for MP6. B10 had a lower growth (7.0mm) compared to both B20 (20.8mm) and Cont (16.0mm). B10 tended to have a lower wear (11.2mm) when compared to B20 (29.6mm), with no difference to Cont (21.4mm). Total hoof wear (MP7) also tended to be lower (P=0.06) for B10 (52.6mm) compared to B20 (75.7mm), with no difference to Cont (64.8mm). Net growth (growth - wear) for MP7 approximated 0.5, 4.5 and -13.1mm for Cont, B10 and B20 respectively.

Irrespective of biotin treatment, rates of hoof growth and wear were not constant over MP as described in Table 1. No relationship existed between steer live weight and rate of hoof growth or wear. However, rates of hoof growth and wear coincided with rainfall events, particularly for MP5. Hoof growth rate increased by 6.4mm and wear rate increased by 15.5mm for this period. Rate of wear (18.9 and 22.0mm) exceeded rate of growth (14.5 and 14.3mm) for MP5 and 6 respectively.

Table 1: Effect of rainfall (mm) and steer live weight (kg) on hoof growth and wear rates for MP.

	MP					
	1	2	3	4	5	6
Rain	0.0	26.8	10.8	10.8	158.1	28.0
LW	474	545	606	638	694	749
Growth						
Overall	10.1^{a}	12.3^{a}	6.5^{b}	8.1^{ab}	14.5^{c}	14.3^{c}
s.e.m	0.8	1.3	1.4	1.0	1.1	2.3
Wear						
Overall	3.8^{a}	10.3^{a}	6.7^{a}	3.4^{a}	18.9^{b}	22.0^{b}
s.e.m	0.6	1.3	0.9	0.5	1.6	3.3

Means within each row with different superscripts differ (P<0.05).

Biotin supplementation had no effect on white line width of the lateral claw. Whereas B10 tended to increase white line width of the medial claw (P=0.12). B10 had a greater white line width (40.2mm) when compared to Cont (36.1mm) and B20 (38.2mm). The lateral and medial claws showed evidence of lesions (sole and white line), however lesion distribution was not different between biotin treatments. Biotin supplementation failed to effect fatty acid profile (C14, C16, C18, C18-1, C18-2 and C18-3) of both white line and fat pad. During the study (MP6), three steers from Cont became clinically lame, requiring treatment.

Discussion

In this study, environmental conditions (rainfall) were observed to effect hoof growth characteristics. Wet pen conditions increased both hoof growth and wear, with wear rate exceeding growth. Cattle maintained under constant wet conditions, have shown an increased incidence of sole lesions and hoof wear (Fitzgerald et al 2000). The B10 treatment reduced hoof growth, wear and total wear. The B10 treatment also provided the highest net growth level. In contrast, the B20 treatment appeared to have a negative effect on hoof integrity. Previous research has shown that supplementing dairy cattle with 20mg/hd/d of biotin significantly improved hoof integrity and milk production (Fitzgerald et al 2000). This suggests that the biotin requirements of long-fed beef cattle are lower than dairy cattle.

An increased incidence of lameness would be expected from steers suffering from high hoof wear levels (Hoblet and Weiss 2000). However, incidence of lameness was isolated to Cont treatment. Biotin supplementation has shown to improve structural integrity of hoof tissue and therefore provide greater resistance to wear and damage (Mulling et al 1999). The existence of lesions on soles and white line of hooves, indicate that all hooves of steers were challenged. However, the low hoof wear level of the B10 treatment indicates an improvement in hoof structural integrity and greater resistance to these environmental challenges.

Reduced hoof wear was not related to differences in fatty acid composition of white line or fat pad. Seymour (1998) reported that linoleic acid (C18:2) was required for synthesis of semi-waterproof hoof wall.

The white line width of the medial claw was increased with B10 treatment. The white line provides the junction between the horn forming tissues of the hoof wall and sole (Hoblet and Weiss 2000). In the front limbs of cattle, the highest pressure exists on the medial claw (Tol et al 2002). Therefore, a wide white line indicates a hoof that is more resistant.

Supplementing 10mg/hd/d of biotin to long fed beef cattle may contribute to improved hoof integrity by reducing wear and increasing white line width in wet pen conditions.

Acknowledgments

The authors gratefully acknowledge the support and co-operation of Roche Vitamins, Australia, the Harris family, Queensland Department of Primary Industries, University of Queensland, Cameron Livestock, Dr Mac Kimura and Oakey Abattoir.

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EFFECT OF METHIONINE CONCENTRATION ON EPIDERMAL CELL VIABILITY IN THE BOVINE CLAW

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Introduction

Sulphur amino acids have an important role in the structure and function of integumental tissues. Previous studies (Galbraith et al, 2002) have described the concentration-dependent importance of methionine in regulating apparent total protein and DNA synthesis in claw tissues by explant technique. The aim of the present study was to investigate the effects of (a) inadequate and (b) approximately optimal concentrations of methionine on indices of cell proliferation under similar conditions in vitro.

Materials and Methods

Claw tissue for culture was obtained from right hind lateral claws of crossbred female cattle (age 18-20 months) post mortem at a commercial abattoir. Explants were prepared from the solear tissue, each weighing 30-50mg, and were cultured in an atmosphere of air/CO_2 (95:5, v/v) at 37 °C in DMEM/F12, deficient in methionine, with methionine added at 1 or 30 μM. Each well of 6-well plates contained ca 300mg tissue and 2.5ml culture medium. Explants were removed from the media at 24, 48 and 72 h. Another set of explants were treated at time 0 with a pulse of 0.05 mM BrdU for 6 hours then fresh media was replaced and explants removed at 24, 48 and 72 h. Explant tissue slices were transferred into OCT embedding compound and frozen in liquid nitrogen. Cryosections were cut at 10 μm using a steel knife on a cryostat (Reichert-Jung Frigocut 2800E) and air dried onto Vectabond coated slides.

For immunohistochemistry, cryostat sections were fixed in 4% paraformaldehyde for 10 min then immersed in a 1 mg/ml trypsin solution. Endogenous peroxidase was blocked by incubating the tissue section in 3% hydrogen peroxide in methanol for 5 min. The sections were rinsed in buffer prior to incubation with dilute serum to block non-specific binding. The primary antibodies were added (monoclonal mouse anti-PCNA, 1:3000 dilution, Sigma; polyclonal rabbit anti-Bcl-2, 1:50 dilution, Oncogene; monoclonal mouse anti-BrdU, 1:1000 dilution, Sigma) and left overnight at 4 °C. The sections were then washed in tris-buffered saline and blocked with serum as before. The secondary antibody Vectastain Universal Elite ABC kit (Vector Laboratories) was used to achieve detection. Sections were then dehydrated through an ethanol series finishing with histoclear for 5 min and mounting in Styrolite (BDH) under a glass coverslip. Apoptotic cells at the basement membrane and measured following the manufacturer's method were stained with digoxigenin-dUTP antibodies (Apoptag; Oncor Inc.). Cryostat sections for Feulgen staining were fixed and immersed in glacial acetic acid: ethanol (1:3) for 2 hours and then hydrated through 75% ethanol for 10 min followed by 50% ethanol for 10 mins. Sections were then washed and immersed in 1 M HCl for 6 min at 60 °C. Following hydrolysis, sections were immersed in Schiffs Reagent (Sigma) for 20 min. Sections were then washed and mounted in glycerol under a glass coverslide and stored at 4 °C until examination.

Slides were viewed using a Polyvar microscope (Reichert-Jung) that was attached to a digital camera (TK-C1381, JVC). Images were captured using specialised analysis software (Image Pro-Plus Version 4.5.0.19 for Windows 98/NT/2000). The positive epidermal cells along the basement membrane of papillae were counted and compared as a function of its length (1 unit basement membrane length = 401 μm) rather than of total cells, for 18 papillae (3 sections x 3 explants x 2 animals). Data were analysed by Minitab (Release 13.3, General linear model).

Results

Epidermal cell proliferation measured by the PCNA and Feulgen techniques was greater ($P<0.05$) at the higher (30µM) compared with the lower (1 µM) concentration of methionine after 72 hours with non-significant trends evident at 48 hours (Table 1; Figure 1). A significantly greater number of cells expressed a positive signal for BrdU at 48 h with similar trends evident at 24 and 48h, in response to the higher methionine concentration. There was also evidence of a significantly greater expression of the apoptosis inhibitor Bcl-2 (30μM vs 1 μM, 72h). However, tests for apoptosis by TUNEL gave time-dependant increases in positive signal but also a greater apoptotic response to the higher methionine concentration.

Table 1. Cells staining positive for various proliferation and apoptosis markers with two concentrations of methionine. Results are shown as mean ± SEM ($P<0.05$).

Positive cells / length basement membrane	Methionine concentration and time removed				
	0 h (n=18)	1 µM at 48 h (n=18)	30 µM at 48 h (n=18)	1 µM at 72 h (n=18)	30 µM at 72 h (n=18)
PCNA	105 ± 4.79^{a}	132 ± 11.1^{a}	153 ± 7.65^{a}	122 ± 8.89^{a}	231 ± 11.5^{b}
Feulgen	118 ± 6.14^{a}	119 ± 7.15^{a}	126 ± 7.19^{a}	89.5 ± 5.13^{b}	101 ± 4.23^{a}
Bcl-2	127 ± 6.30^{a}	133 ± 12.4^{a}	122 ± 8.71^{a}	87.5 ± 4.79^{b}	130 ± 4.68^{a}
Apoptosis	54.2 ± 7.48^{a}	90.5 ± 5.40^{b}	116 ± 8.18^{bc}	105 ± 5.12^{b}	146 ± 14.7^{c}

Mean values with different superscripts within rows are significantly different ($P<0.05$).

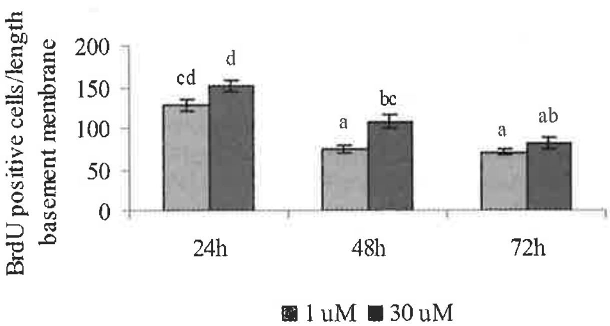

Figure 1. Number of cells staining positive for BrdU, after a 0 to 6 h pulse. Results are shown as mean ± SEM for n=18 papillae ($P<0.05$).

Discussion

The data produced evidence of the importance of methionine supply in supporting proliferative activities in basal epidermal cell keratinocytes. This result was particularly evident with increasing time of incubation and likely depletion of endogenous concentrations up to 72h. Responses were associated with the maintenance of the signal for the apoptosis inhibitor Bcl-2 at the higher compared with the lower methionine concentration. In contrast, the signal for apoptosis by TUNEL analysis suggested a greater number of apoptotic cells at the higher methionine concentration. This result, along with that for proliferation, may indicate a stimulation of epidermal cell turnover. The reasons for this are not clear but may relate to changes in cell cycle behaviour in response to alterations in nutrient supply. It is concluded that the work confirms the importance of methionine supply on proliferative behaviour and identifies an apparent apoptotic response. It also suggests a future requirement to clarify effects on the cell cycle and the mechanism of sensing nutrient sufficiency by epidermal horn-producing cells.

Acknowledgements

This work was funded by BBSRC and the Scottish Executive Environment and Rural Affairs Department. Technical assistance provided by Mr J Struthers and Mr M Birnie is gratefully acknowledged.

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SITE-SPECIFIC SULPHUR AMINO ACID UPTAKE, INCORPORATION AND PROTEIN AND DNA SYNTHESIS IN BOVINE CLAW TISSUE CULTURED IN VITRO

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Introduction

The claw is a specialised structure of the bovine integument with function dependent on interaction between epidermis and dermis at a number of sites. These sites vary in the composition of horn-produced (Galbraith et al., 2002a) and contribution to suspensory function. Relatively little is known about the dynamics of nutrient uptake and protein and DNA synthesis at these functionally important locations. The sulphur containing amino acids are among important nutrients that support successful horn production. The dermis is known to supply nutrients and chemical signals to support proliferation and differentiation of horn-forming epidermal keratinocytes and expression of proteins involved in cytoskeleton and intercellular adhesion. The cytoskeletal intermediate filament proteins and associated proteins contain variably high concentrations of cysteine, which, in certain tissues may be maintained by methionine via the transulphuration pathway. Methionine also has a central role in polyamine synthesis, methylation and protein synthesis both structurally and in translation. Utilisation of methionine by tissues is governed by extracellular and cellular uptake and incorporation during synthesis and metabolism.

The aim of this study was therefore to investigate at functionally important sites, (a) the uptake of methionine and its incorporation into protein in cells and (b) DNA synthesis by thymidine incorporation under conditions standardised and established previously (Galbraith et al., 2002b).

Materials and Methods

Tissue explants (ca 50 mg wet weight) from the sole, heel, mid-laminar and coronary region were prepared post mortem from hind right lateral claws of female cattle (n=6, age 18-20 months). Explants (4 per well) were incubated in DMEM/F12 culture medium and an atmosphere of air/CO2 (95:5, v/v) at 37°C. After 21 h incubation, fresh media were added supplemented with either L- [35S] methionine (1μCi/ml) or U- [14C] sucrose (0.5μCi/ml) for 30 min followed by washing in fresh media, blotting and incubation in the presence of TCA (10%, w/v) for 16 h. Radioactivity in the TCA soluble fraction was counted and intracellular uptake of L- [35S] methionine calculated as released radiolabel, applying corrections for uptake in extracellular space (U- [14C] sucrose uptake) (Shennan and McNeillie, 1994). Also after 21 h, a series of explants were incubated in fresh media supplemented with L- [35S] methionine (6μCi/ml) and [6-3H] thymidine (2.5μCi/ml) for 3 h to measure their incorporation into the TCA-insoluble, formate-soluble fractions as measures of protein and DNA synthesis by methionine and thymidine incorporation, respectively. Data were analysed by Minitab (Release 13.3, General linear model).

Results

Measurement of intracellular accumulation of methionine as a measure of cellular uptake in tissue explants was significantly higher in the mid-laminar region compared to the sole and heel but not different from the coronary region (Table 1). In contrast, methionine (35S) incorporation in the cellular protein fraction, was significantly higher in the sole compared to the mid-laminar but not different from the heel or coronary region (Table 1). There was no difference in the incorporation of thymidine into the TCA insoluble, formic acid soluble fraction, as a measure of DNA synthesis, between the various regions (Table 2).

Table 1. Uptake and incorporation of methionine in four regions of the claw. Results are shown as mean ± SEM for n=6 animals.

	Region of hoof measured:			
	Sole	Heel	Mid laminar	Coronary region
Methionine uptake (mmol/kg intracellular water/30 min)	29.4 ± 3.54^{a}	31.6 ± 2.81^{a}	41.5 ± 3.18^{b}	38.7 ± 3.39^{ab}
Extracellular space (F-value)	0.30 ± 0.02^{a}	0.51 ± 0.01^{a}	0.20 ± 0.02^{b}	0.21 ± 0.02^{b}
Methionine incorporation (nmol/kg intracellular water /3h)	3.52 ± 0.35^{b}	2.59 ± 0.31^{ab}	1.59 ± 0.31^{a}	2.63 ± 0.31^{ab}

Mean values with different superscripts within rows are significantly different ($P<0.05$).

Table 2. Incorporation of thymidine in four regions of the claw. Results are shown as mean ± SEM for n=6 animals.

	Region of hoof measured:			
	Sole	Heel	Mid laminar	Coronary region
Thymidine Incorporation (pmol/kg intracellular water /3h)	46.6 ± 6.46	38.8 ± 3.60	49.5 ± 7.15	52.8 ± 6.21

Discussion

The data for the proportion of extracellular space indicate significantly greater values for sole and heel compared with the coronary and mid-laminar region. These results may suggest a larger ratio of extracellular matrix to cells in the former than the latter explant systems studied. Results for methionine uptake and incorporation and thymidine incorporation were of a similar order to those recorded previously for solear explants (Galbraith et al., 2002b). The data for cellular uptake suggest a greater apparent transport of methionine into cells of the mid-laminar region than the sole during the time period of study. Reasons for this are not known but may relate to differences in the epidermal-dermal structures at these sites which may effect nutrient supply to the predominantly epidermal cells. In contrast, the incorporation of the 35-S label into the intracellular protein fraction suggests a greater rate of protein synthesis in cells of the solear compared with the mid-laminar region. Trends were also noted which gave some evidence suggesting apparently greater cellular protein synthetic activity in cells of heel and coronary compared with that of the cells of the mid-laminar region. The absence of significant differences in apparent DNA synthesis in cells suggest a similarity in proliferation rates at the four sites. It is concluded that the characterisation of the in vitro explant system provides the first comparative description of proliferation and protein synthetic potential at the functionally important sites investigated. Future work will refine the systems to permit investigation of the regulation of these processes and how they may be influenced to produce the lesions characteristic of lameness in affected claws.

Acknowledgements

This work was funded by the EU Lamecow Project QLRT-2001-00969. The technical assistance of MJ Birnie and MA Brown is gratefully acknowledged.

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CONTENTS AND COMPOSITION OF CERAMIDES IN THE BOVINE HOOF HORN FROM NORMAL COWS AND COWS WITH SUBCLINICAL LAMINITIS

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Introduction

Hoof disease is one of the most serious problems in the dairy industry. It leads to reductions in milk yield and fertility and to shortening of productive life. Hoof disease can be caused by various factors, including housing, behavior, genetics, infective agents and nutrition. A strong and intact horn shoe (stratum corneum) is required for the protection of the claw's soft tissues and for maintenance of the biomechanical function of the claw. Lipids are a component of cell membranes and are present in the intracellular space of the claw epidermis in cattle. It is known that these lipids are important for both water retention and barrier function in the stratum corneum of human skin. Water content of the claw horn was found to be closely related to the horn hardness. Hardness is an important factor for the protection of soft tissue and basal membrane cell layer inside the claw against physical loads, especially body weight. Thus, water regulation mechanism is important for maintenance of the proper function of the claw horn. Ceramides are composed of sphingosine and fatty acid, and have been shown to be predominantly associated with water retention and barrier function. It has been reported that seven types of ceramide were detected in porcine and human skin. These ceramides play crucial roles in physiological function of skin, and a decrease in ceramide contents of the strutum corneum is related to atopic lesions of skin. However, ceramides in the bovine hoof horn have not been analysed in detail. In this communication, we present results of analysis of the quantity and pattern of the hoof horn ceramides from normal cows and cows with subclinical laminitis.

Materials and methods

Hoof horn samples of Holstein-Friesian cows were obtained from slaughterhouse. Cows were clinically evaluated to determine hoof health, and were identified as having sound hooves (n=13) or suffering from subclinical laminitis (n=8). The diagnosis of subclinical laminitis was made on the basis of findings of extensive yellow sole horn consisting of extensive yellow discoloration and haemorrhage. Four zones of sole horn based on the recommendation of the Sixth Symposium of Disease of the Ruminant Digit were used in our study. Two zones of wall horn were newly shown in this study (Fig. 1). Lipid extraction from hoof horn samples was performed according to the methods of Scaife et al. (2000) and Imokawa et al. (1991). Briefly, samples of bovine hoof horn were reduced to fine shavings using a rasp. Lipid extraction was performed at room temperature for 1 h in a 15-fold excess of chloroform : methanol (2:1). Ceramides of hoof horn samples were separated by thin-layer chromatography and were developed twice with chloroform : methanol : acetic acid (190:9:1) . After solvent development, the chromatograms were air-dried, sprayed with 10% $CuSO_3$-8%H_3PO_4 aqueous solution, and charred on a 180° hot plate. The charred lipids were quantitated using NIH (National Institute of Health) Image software. Ceramides were measured by determining the amount on ceramides on a TLC chart of appropriate commercial standards and expressed as µg ceramide / g hoof horn. Ceramide type 1 and type 2 were used as standards for ceramides I, IIa, IIIb, III, IV, V+VI and VII respectively. Reproducibility of this method was confirmed using triplicate samples from the same animal, and deviation of values was within 5%. The level of significance of the difference was calculated by Student's ttest.

Results and discussion

Ceramides I, IIa, IIIb, III, IV, V+VI and VII were clearly detected in sole horn samples, but ceramides I, III, IV and VII were not detected in wall horn samples. Total ceramide content of the sole horn samples was 1346.4±221.3 (µg/g) and was significantly ($p<0.01$) higher than that of wall horn samples, which was 638.6±82.6 (µg/g) (Table 1). This is the first report on quantity and types of ceramide from the bovine hoof horn. It has been reported that seven types of ceramide were detected in the stratum corneum of human skin. In this study, the patterns of chromatograms of hoof horn ceramides from sole horn samples were similar to those of human skin reported by Imokawa et al (1991). The reason for the differences in quantity of ceramide from sole and wall horn was not clarified in our study.

The contents of total ceramide and ceramide type IIa, IIb, III and VII of hoof horn from cows with subclinical laminitis were significantly ($p<0.05$) lower than those in claw horn samples from normal cows. Among the 6 types of ceramide fraction, ceramide 1 was found to be most significantly reduced in atopic lesional skin of humans. It has been reported that keratinocytes in the basement membrane cell layer are important for the formation of claw horn, and that cell metabolism was markedly inhibited in cow with laminitis. From our findings, we speculated that the decreased quantity of ceramide of hoof horn in cows with laminitis was caused by the inhibition of ceramide synthesis by keratinocytes in the condition of subclinical laminitis. Determination of the types of ceramide in the bovine claw horn may be important for clarification of the mechanism of bovine claw disease.

HEALTHY FEET REQUIRES COW COMFORT 24 HOURS

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Introduction

Claw and leg injuries resulting in lameness are of major concern to the dairy industry. The importance of the problem increases with more intensive production, larger units, and more automatized operations like the milking robot. There are many straggling risk factors involved in the development of lameness, such as genetics, nutrition, management, foot trimming and housing. Among the housing factors the physical as well as the biological environment are of concern. The animals' general and immunological resistance and behaviour predicts the development and severity of the lesions. Especially flooring and, indirectly, lying conditions and the time of exposure of these specific areas are crucial.

Claw conformation and development of claw lesions

Cows´ feet have evolved to resist the most varying situations, from heavy precipitation to drought and from hard rocks to soft swamps. In nature, cows live on soft yielding surfaces that are most often clean from manure, non-slippery, and self-maintaining. The animals can move and behave naturally and there is enough space for the cow to escape or to avoid critical areas. It is also possible for her to choose a lying place that is comfortable and gives enough support and space for rising and laying down, reducing the risk for chafing. However, in confined dairy operations, the floors are most often made of non-yielding concrete that gets slippery with time, and manure contamination of the feet is a rule rather than an exception. In most species with hooves or claws, the wall is the main supporting structure that should take most of the initial loading. In contrast to smaller species with cloven hooves and similar to the horse, cattle has developed a thicker sole to protect the sensitive structures beneath. Despite this, the majority of the most important claw lesions occur in the sole of the outer rear claws. Regular and functional claw trimming corrects asymmetric claws, equalizes weight distribution and reduces the risk of claw lesions and lameness by early detection (Manske et al. 2002a).

Difference in growth and wear of the claw horn is to a great extent a result of the environmental conditions and is reflected in the shape of the claws. Many studies have paid attention to growth and wear of the claws but mainly of the wall and not of the sole. Tranter and Morris (1992), however, studied growth and wear of both the wall and the sole and related the results to environment, parity and lactation-stage. Already when bringing the cow from pasture to the milking area, where the walkways and the collecting area were paved concrete, the natural conditions were altered and resulted in changes of the claw conformation. During lactation, both growth and wear of the wall and the sole increased in the outer claws of the rear feet in comparison to the inner claws. The sole shape of the inner claw remained concave while the sole of the outer rear claw became flat and sometimes convex. When the animals were dried off and kept on grass alone, the concave shape of the outer claw was somewhat restored. Thus, it seems like asymmetry between outer and inner rear claws is associated with change of environment at calving.

Claw horn lesions

When the outer claw in the rear feet increases in size and the sole is flattened, the counter pressure between the rear part of the claw bone and the ground may cause traumatic damage to the sole corium. A yielding surface will give less counter-pressure and reduce the risk of lesions.
Furthermore, metabolic changes related to laminitis, and hormonal changes at calving (Tarlton et al. 2002) weaken the bone-to-claw-capsule adhesion and could be contributing factors in the development of traumatic sole lesions. Primarily, a contusion results in capillary damage and as a consequence sole haemorrhages can be observed at the sole surface about 3 months after the initial injury. If the damage is consistent and large enough, a sole ulcer may develop. Swiss pathological studies (Lischer et al. 2002) revealed that cows with sole ulcers had a significant dislocation of the claw bone and that the fatty containing, chock absorbing claw cushions were affected. Whether the claw cushions were affected before the development of the ulcer, or if the damage of the cushions was an effect of the ulcer was not determined. However, it can be presumed that long-term exposure of the feet to hard floors most likely wears and tears the elastic structures and the claw cushions. White line disease often starts with a sole haemorrhage and a fissure of the soft white line and an abscess can develop beneath the sole. The corium in connection with the white line lesion is damaged and hemorrhagic, and can progress to necrosis and deeper infections just like at the typical sole ulcer site. It is likely that different types of sole lesions are of the same origin (Manske et al. 2002b) and differences in their appearance are related to the interaction between each cow's anatomy, behaviour and different aspects of

her housing conditions.

Hygiene related claw diseases

The claw horn needs moisture to maintain elasticity while in arid conditions the wall may crack. This is rarely a problem in intensive dairy production because the cows are walking more or less permanently in dirt. However, the manure also includes chemicals that can damage horn and it is a reservoir for microorganisms that can infect the skin. On grass, the feet are often cleansed mechanically. Thus pasture normally causes less of a problem for the feet. When cows are confined, there is an increased risk for consistent moisture of the claws if there are few dry areas to which they have access and can dry off. Moist conditions that resolve the protecting skin is presumed to an important risk factor for the development of digital dermatitis (Rodriguez Lainz et al. 1996). Moreover, the rear feet are more prone to infectious diseases such as digital and interdigital dermatitis, verrucose dermatitis, interdigital hyperplasia, interdigital phlemon and heel horn erosion (Bergsten et al. 1998) because the cow is often standing with the rear feet in the dirt while the fore feet are standing drier, e.g. tie stall or half into the cubicles.

Walking comfort

The flooring of the dairy facility is of outstanding importance for claw health. Concrete is a cheap, strong material for constructions and easy to clean, but does it give the optimal conditions for the cows´ feet and legs? Manske (2002) found that lameness and claw horn lesions (sole ulcers, double soles, white line lesions, dermatitis and heel horn erosion) were significantly associated with loose housing systems, concrete floors and poor hygiene. The prerequisite for good claw health is simple to understand: a floor, which is resilient, hygienic and keeps a balance between growth and wear. Such an optimal floor is not yet invented but a "movement" has started.

Soft flooring

We have more than twenty year's positive experience from rubber mats in tie stalls. Although the traditional, old type of rubber mats were rather hard, they resulted in an improved claw health (less sole ulcers and less sole haemorrhages), when compared to concrete stalls (Bergsten 1994; Bergsten and Frank 1996). It is not always obvious whether this is a direct effect from standing on softer flooring or an indirect effect from standing less time due to more comfortable bedding. Technical difficulties in maintaining soft floors and associated investments are, however, hard to carry out in loose housing systems. Several systems to improve softness and friction for both slatted and solid floors are under development, using natural or synthetic rubber, PVC, elastic epoxy or mastic asphalt. A German study in a cubicle stall, has shown that installation of soft slatted rubber mats significantly improve the animals´ ability to perform natural behaviour and claw health in comparison cows kept on crude concrete slatted floors, although the activity, and thus also the loading of the feet, was increased in the cows on the slatted rubber mats (Benz 2002). The study also showed that both claw growth and wear were reduced on the rubber and the net growth was not dramatically increased. Even if concrete may not be the best for cows´ feet, we must remember that cows can adapt to different environments. Spring calving heifers with a four months pre-calving accommodation period to concrete had significantly lower post-calving sole lesion scores than autumn calving heifers that came directly from pasture to concrete stalls prior to calving (Bergsten and Frank 1996).

Daisy's choice

Does the cow know what is best for her? In the ongoing EU-project LAMECOW, two preference tests have been made. Firstly, three hundred cows in a commercial dairy, in groups of more than 40 cows, were video-recorded in the holding pen before entering the 2x12 parallel milking parlor. During milking the number of animals in the holding pen was reduced by 12 at a time and the position of the animals was assessed after a few minutes, when the positions were stabilized. A new group of animals were introduced after the pen was completely emptied. The cows had two floor-alternatives at each testing; "soft rubber" mats versus "very-soft rubber" mats, "concrete" versus "soft rubber" mats or "concrete" versus "very-soft rubber" mats. Each alternative was tested during 4 days and repeated in reversed position of the mats (i.e. left and right side) to correct for side preferences. Initially when the pen was full there was no preference. However, as the number of animals in the holding pen decreased cows could more easily change positions. The largest significant difference in preference was found between any of the rubber mats (75%) and the concrete flooring (25%) when there were 4 square meters or more available per cow in the pen. A preference was also seen for the very soft rubber mat (65%) versus the soft rubber mat (35%) when there was more than 12 square meters available per cow in the pen, but the difference was not significant. In a second experiment, the preference for different floors on the walk-way after milking was tested. One hundred fifty cows in a commercial dairy could chose to walk on either of two different floors when they were voluntarily leaving the milking parlor; "slatted concrete" flooring versus "slatted rubber" flooring, "slatted concrete" flooring versus "solid rubber" flooring, and "slatted rubber" flooring versus "solid rubber" flooring. The animals had earlier been accommodated to the different floors. The preference increased successively during the four days observational period. At day four 80% of the cows selected slatted or solid rubber flooring to slatted concrete flooring (20%). A slight preference for solid rubber versus slatted rubber flooring was also noticed but the preference was less consistent over time.

Locomotion comfort

In the LAMECOW study, we have also analyzed the locomotion comfort on different surfaces/floors by analysing the cow track ways (measurements of foot prints along a 10 m lane) as an expression of the locomotion pattern (Telezhenko et al. 2002a; Telezhenko et al. 2002b). We found that when a cow walked on 5 different surfaces one after another, she walked most natural on firm sand, and least good on slippery slatted concrete. When slatted or solid rubber mats were applied on top of the concrete floors, the locomotion was significantly improved. Moreover, healthy cows were compared to cows with mild or moderate lameness according to Sprecher and Hostetler (1997). Mild to moderate lameness resulted in bigger step asymmetry, a shortened stride and step length compared to healthy cows (Telezhenko and Bergsten 2003). Interactions were seen between lameness and floor type. Decreased step angle (wider posture) and asymmetry in lame cows were most pronounced on slatted concrete and least on sand. Rubber mats on solid and slatted concrete floorings improved the gait parameters relatively more for lame cows than for healthy ones.

Slipperiness

Slippery flooring causes accidental traumatic injuries and downer-cows and is an animal welfare problem. Slipperiness also reduces natural and reproductive behaviour and makes heat detection more difficult. Concrete is sensitive for acids and together with the use of steel scrapers the superficial structure changes and gets slippery with time. Therefore solid concrete floors are often grooved for better footing. Acid resistant mastic asphalt, as an alternative, can give a good friction and has long durability. Rubber floors are slippery when wet but with more resilient rubber the claws still get acceptable footing (Benz 2002). In another LAMECOW trial, Telezhenko et al. (2004) tested a group of dairy cows monthly on different floors after a three weeks adaptation period. Three concrete floors with different surface texture (smooth, hexagone stamped and diamond cut pattern), mastic asphalt and rubber mats (Kraiburg KSL) were tested. Slatted concrete was used as reference flooring and the cows walked relatively "more natural" (longer strides) on rubber mats than they did on any of the other surfaces compared to the concrete slats. The surface of concrete floors and mastic asphalt is of most importance for friction and abrasiveness. A negative consequence, when friction is required on a concrete or asphalt floor, might be excessive abrasiveness. If animals are too much exposed, the wear will be larger than the growth and the corium can be traumatically injured. This is a very common problem when introducing animals to a newly constructed concrete floor, no matter if it is solid or slatted. A common recommendation is to treat the fresh concrete with cold asphalt and saw dust as a softener until the sharp layer wears off. If mastic asphalt gets too abrasive it must be grinded or remodelled because it will not be less abrasive by wear. Experiments to study the long-term effects and time budget of different floors on claws´ conformation and health are under progress in the LAMECOW project.

Lying comfort

Cow comfort includes lying comfort as well as comfort when the cow is standing and walking. Lying comfort is very important for the foot health, because if animals are not lying down their feet will be exposed to environmental challenges, which may result in claw lesions and lameness. Leonard et al. (1994) showed that with uncomfortable cubicles (reduced space and concrete surface) first calving heifers lay down less around calving than heifers in more comfortable stalls (larger size and equipped with rubber mats). The animals that lay down less time developed significantly more sole lesions, as recorded one month after calving. Leonard et al. (1996) also showed that over-crowding in a cubicle stall and competing for a place at the feeding area led to significantly more sole lesions in animals that lay down less time.

Space limitations

The lying comfort can be divided into three important features: space for lying down and rising, comfort of the bed, and hygiene. In state of nature, all these features are likely to be satisfactory. Even if the lying base is not always very soft in nature, the animal may easy change her position to reduce repeated pressure on sensitive body parts as rising and lying down has no obstructions. In some loose housing systems like deep-litter bedding (straw pack), the space and bedding may be satisfactory. But in tie stalls (stanchions) and in loose housing (cubicles, free stalls) the freedom is more or less restricted. In short tie stalls the cow must have her head over the manger while resting, and rising and lying down is often impaired due to the constructions around her. A short stall corresponds in size to the lying area (~1.75 m) of cubicle (free stall). The Swedish animal welfare legislation states a minimum total length of 2.30 m of the cubicle provided that there is a lunge space to the side. But as lunge space to the sides feels less convenient for the cow it can result in a cow lying diagonal and defecating in the stall. The lunge space needs to be about one meter extra in length from the normal lying position. The neck-rail is probably the most difficult part to adjust correctly. Unfortunately, the neck-rail is often used to make the cow lie down further back in the cubicle. If the neck-rail is installed too low or too much backwards in the stall, there is an increased risk that animals refuse the cubicles or stand half in. This may result in increased loading and manure contamination of the rear feet, leading to claw lesions and lameness. Instead, the neck-rail must allow the cow to stand completely in the cubicle, but without defecating there. The lying down and rising behaviour are thus facilitated. A specially designed neck-rail with a bow for the neck can direct the cow to the middle of the stall. The brisket board or a rail in front of the head of the cow is used to keep the lying cow in a position to prevent dirt in the stall. It is not possible to have individually tailored stalls, and thus it is appropriate to keep a herd with animals that do not differ too much in size. Mixing Jerseys with Holsteins may therefore not be a very good idea.

Cosy bedding

Not too long time ago, most animals were lying on concrete with small amounts of bedding. Cermac (1988) showed that the lying time was shortest (7 hours) on crude concrete and was prolonged on traditional rubber mats (10 hours) and longest on mattresses (14 hours), that is well comparable to pasture resting time. Today, we have been more aware of the impact on health and well being of soft bedding, and many different materials are developed and promoted. Still, the best bedding comfort, with least hock injuries, is obtained with deep-bedding. Straw, saw dust, shavings and dried recycled manure are common organic materials but are hygienically sensitive. Sand is considered to be the best bedding also from a hygienic point of view and is popular in North America, where Cook (2003) also found it beneficial for the herd claw health. In all deep-bedding systems, management is considered difficult due to cost and labour. Today there is an increasing interest in different kinds of soft mats and mattresses. The soft mat or mattress has two components, the resilient part and the surface layer. Rubber and synthetic materials are most commonly used but also waterbeds are popular. Many new materials have problems because they cause abrasions of the hock or other protruding parts of the body (Livesey et al. 2002). The lesions can not only be blamed on the mat/mattress but also imperfections in the space for the cow in the cubicle and on too much humidity. A layer of litter aids to absorb moisture and to reduce abrasions. For hygienic and bio security reasons the cubicles must be able to clean and disinfect. Cleaning and disinfection can be facilitated through avoiding open seems between mats etc.

Hygiene of stall and floors

Poor hygiene in dairy operations does not affect only the claws but also udder health, milk quality and it has consequences on the environmental emission of ammonia. There are possible improvements in all respects by improving the general hygiene and to cleane the manure from the animals' environment as quickly and effectively as possible.

Filthy floors

The hygiene of the floors is an important task, especially in large herds with intensive production. Traditionally, solid floors are scraped with tractor when animals are milked (twice or three times daily). With much manure produced this does not give acceptable cleanliness. Flushing can result in very clean floors, but using recycled water can recycle microorganisms to cows´ feet. With long alleys and unfrequent scraping much manure is moved in front of the scraper, and if animals are present, the dirt will flood the feet and impair their cleanliness. In Europe, mechanic scrapers are more common and automatic scraping can be made more frequent, even continuously. More efficient scraping would be achieved with a two-direction scraper and with a gutter in each end of the alley. Another feature of high importance is that the urine is drained from the floor between scrapings, usually towards a canal in the middle of the alley. If enzymes from the faeces are mixed with urine, emission of ammonia is highly increased. Sloped alleys which separate urine reduce ammonium emission considerably. Scrapers should be equipped with a resistant rubber, both to clean more efficiently and also to reduce wear of both the floor and of the blade.

Slatted (draining) floors are very popular in Europe although they require a larger investment. Many types of slatted floors exist with different pattern and different percentage of openings. With larger slots or holes there is a risk for damage of the claws, which in severe cases could lead to exungulation or fracturing of the claw. With too small openings, low cow traffic, and use of litter in the cubicles, the draining capacity could easily be reduced. The recent Swedish regulation states that the maximum opening for adult cattle should be reduced from the earlier allowed 40 mm to 35 mm. Still you can find openings of 45 mm or more in many herds. Own experiments showed that drainage capacity could be sufficient with slot openings reduced to 30 mm if the slat width simultaneously is reduced to 100 mm from 125 mm. With too large openings, the cows' locomotion will be affected as the animal is anxious to walk over openings. Also the walking comfort was improved in present trial with the 30/100 mm slatted floor in comparison to the control floor of 40/125 mm slats. The slatted floor needs management to remove dirt from areas where there are not enough cow traffic (behind cubicles) and today scrapers on top of the slatted floors are getting more popular. From preliminary results in the LAMECOW project we found that scraped slatted floors resulted in significantly cleaner floors, cleaner stalls and less lameness in comparison to non-scraped slats.

Bed hygiene affects feet and udder health

The cow must be able to stand and lie down in the stall correctly to prevent manure in the stall. Besides direct effects on claw health, poor hygiene of the lying surface is critical for the growth of pathogenic bacteria that may affect udder health. Moreover, manure is a carrier of spores that affect milk quality. Also, the dirt can be kept out from the cubicle in a loose housing stall by keeping the walk-ways clean. If the feet are dirty from dirty floors the feet can bring dirt into the cubicle where the udder is contaminated. Hygiene is a big problem in tie stalls as the cow is both eating and lying in the same stall and the claws are often standing in the manure. In many countries, electric cow-trainers are used as an aid to promote the cow to defecate in the gutter instead of in the stall. Cow-trainers were used in a study to demonstrate the importance of stall hygiene on the incidence of infectious claw diseases i.e. heel horn erosions and interdigital dermatitis (Bergsten and Pettersson 1992). By using electrical cow-trainers, in one of two rows of animals, different hygienic conditions were created in each of two groups in a tie stall barn, where the cows previously had suffered from severe heel horn erosion. During the grazing season the lesions healed spontaneously and all animals were healthy when housed at the start of the study. All the claws were trimmed and scored for lesions and cleanliness of environment and animals were appreciated. After four months exposure, the claws were trimmed and recorded again. It was found that despite frequent cleansing of the "non-trainer side" the "trainer-side" was much cleaner and all cows had healthy claws while all cows on the dirty side had severe heel horn erosions. When cow trainers were installed over half the numbers of the affected animals the erosions recovered while the other affected animals developed even more lesions. Law in Sweden now forbids electric cow trainers. To improve the hygienic conditions in tie stalls a new a rubber slatted flooring system has been developed. The rear solid part of the stall is replaced by rubber slats above a manure canal. The solid part (1.50 m) of the stall is covered by a traditional rubber mat. When comparing claw lesions, the control group (tied on traditional solid rubber flooring) developed significantly more interdigital dermatitis and heel horn erosions than those tied on rubber slatted floors (Hultgren and Bergsten 2001). The animals on rubber slatted flooring also had a cleaner stall environment, cleaner body parts and significantly better claw hygiene. Today rubber slatted flooring is also installed in the rear part of cubicles not least to improve cleanliness of the udder. When the EU-study LIFE Ammonia project was started in the same university dairy, it was completely rebuilt with short stalls and rubber slatted flooring at the rear 0.30 m. The diets were changed in protein content to reduce ammonium in the manure. As a result from the different actions, the ammonium in the air inside the barn was reduced from 10 ppm to 3 ppm, and the feet were extremely clean and free from claw lesions (Gustavsson et al. 2003).

Specially designed feed stalls

Feed stalls are made in elevated stalls with dividers and rubber mats to secure optimal eating comfort. This type of stalls (~0.8 x 1.6 m) have been used for more than ten ears in Swedish dairies with good experiences. Feed stalls allow the cow better integrity when feeding and alleys can be scraped without disturbing the animals. The rubber mat reduces exposure to concrete and thus less risk for traumatic claw lesions and the curb makes the cow to stand out of dirt and reduces risk for infectious claw diseases. The animals' behaviour was video recorded in the LAMECOW study. It was found that significantly more cows (70%) were feeding after milking and in the feed stalls and there were significantly less cow displacements caused by other cows and/or scrapers in the feed stalls, than at the traditional concrete slatted feeding alley. Results from claw observations with different eating comfort are not yet ready.

Conclusions

Claw lesions result to a great extent from excessive exposure of a flooring which may be too hard, too abrasive, and unhygienic and if the animals feet are not accustomed to it. Similarly, leg lesions, most often hock and carpal inflammation, are results from too much exposure of hard, abrasive floors of the bed, which the animal has not been accustomed to and with a space limited stall construction that inhibit natural rising and laying down. With more high-producing cows these facts can not be compromised and in new dairy operations it has to be planned for Cow Comfort 24 hours.

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EFFECT OF TRAINING DAIRY HEIFERS TO USE CUBICLES BEFORE FIRST CALVING ON SUBSEQUENT BEHAVIOUR AND HOOF HEALTH

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Introduction

Putting untrained heifers into cubicles after calving is not an uncommon management practice on many farms. Field experience suggests that untrained heifers take some time to become accustomed to using cubicles and so tend to lay down less when they first enter the main milking herd. It is postulated that this may contribute to subsequent lameness. This study investigated the effect of heifers experiencing cubicle housing or not during the winter before first calving on claw horn lesion development after calving and housing in the next year.

Materials and methods

The study was carried out over three years. All available unserved heifers each year were randomly allocated to treatment group during the late summer at approximately 12 months of age (a year before they were due to enter the main milking herd). They were either housed in a cubicle shed (trained) or in a straw yard (untrained) during the following winter when all animals were served. All animals were grazed as a group together during the summer prior to calving and were housed as a single group on straw yards before first calving. Heifers remained on the straw yards until batches of four or more animals had calved and were ready to enter the main milking herd in the cubicle shed. (This was to facilitate behavioural observations of heifers on entry to the cubicle shed, the results from which will be published subsequently). Number of days after calving for which animals remained on straw ranged from 1 to 26 days. Numbers of animals calving for each treatment group for each year of the study are given in Table 1.

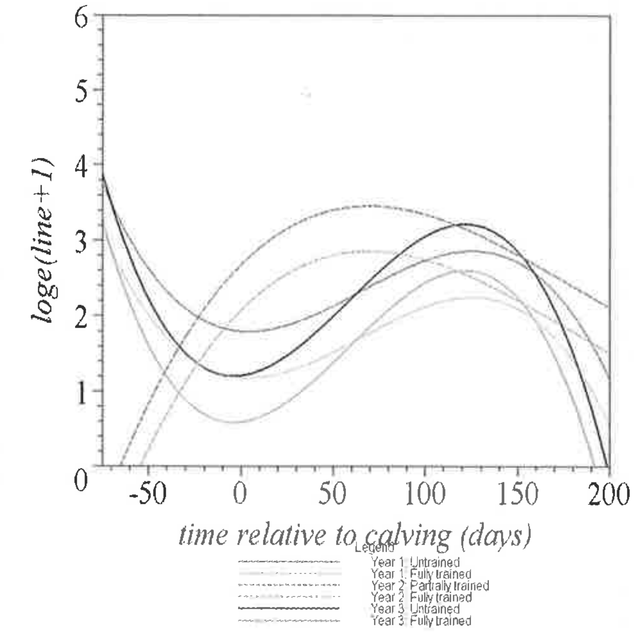
Table 1. Numbers of animals for each year of the study

Year	Calving dates	Untrained	48 hours training	Cubicle trained
1	06/11/99 - 02/02/00	8		8
2	20/08/00 - 23/10/00		14	16
3	28/12/01 - 13/04/02	11		7

After the first year of the study it was noted that one of the early calving, completely untrained animals did not lie down for the first 24 hours after introduction to the cubicle house. Since allocation of animals for the next year was taking place it was therefore decided, on welfare grounds, to compare the effect of partially training to cubicles with full cubicle training. Partial training consisted of allowing the group of heifers to experience cubicles for 48 hours during the winter before autumn calving. Initial analysis of the data showed there to be a less severe effect of not training than was originally thought and indeed some very preliminary analysis showed little or no benefit from partial training. Thus, for the final year of the study treatments reverted to those in year 1 ie. untrained vs. fully trained. In year three animals calved during the spring due to a delay in breeding caused by FMD (Table 1). These animals were turned out to grass when they were four months post calving. This group of animals were accidentally over fed concentrates when animals were on average 2 months post calving (May) as described by Offer et al. (2004). Some became acutely acidotic and were removed immediately to a straw yard until they had recovered sufficiently to return to the cubicle house (2 - 4 days).

In all cases claw horn lesions were monitored at regular intervals throughout first lactation using image analysis techniques (Leach et al 1997). Observations were made approximately 1 month prior to calving, within a week of calving and at 2, 4 and 6 months after calving. All post-calving observations were made while animals were housed apart from the 6 month observation in year 3. In this year animals were housed only until the 4 month post calving examination. In the final year two additional hoof examinations was also carried out, one the day after cows were housed permanently for the autumn (late lactation) and again 3 months later (mid January).

Lesion scores (severity x proportion of the foot affected) were first modelled (using REML) as a function of training regime, year of study, days relative to calving and the time of year and then the number of days for which the animals were retained on straw yards after calving was also included in the model.



Results

Claw horn lesion development after calving followed a similar pattern (see figure 1) to that observed in various studies (Leach et al., 1997, Offer et al, 2000) regardless of training regime. The exact pattern with respect to days relative to calving was, however, affected by year ($P<0.001$, and $P=0.009$ for line and sole lesions, respectively). There was a more marginal effect of time of year (see figure 2) which was insignificant for line lesions and effected by time relative to calving ($P=0.013$) for sole lesions. For line lesions there was a significant effect of training ($P=0.013$), with benefit for fully trained over untrained and partially trained. For sole lesions (see figure 1) there is significant interaction between treatment and year ($P=0.045$), due to fully training in year 1 slightly increasing lesions. However, when days on straw is included in the model ($P=0.043$) this interaction is no longer significant and there is a significant effect of training overall on sole lesions ($P=0.007$), with benefit for fully trained over untrained and partially trained. Thus, the apparent difference between treatment effects seen for sole lesions for the different years could be just due differences in days on straw. Estimated coefficients and P-values for deviance tests for the treatment effects for the models including days on straw are shown in table 2. The treatment effect on line lesions when similarly adjusted for the effect of days on straw ($P=0.086$) is still marginally significant ($P=0.041$) with benefit from full training over no training.

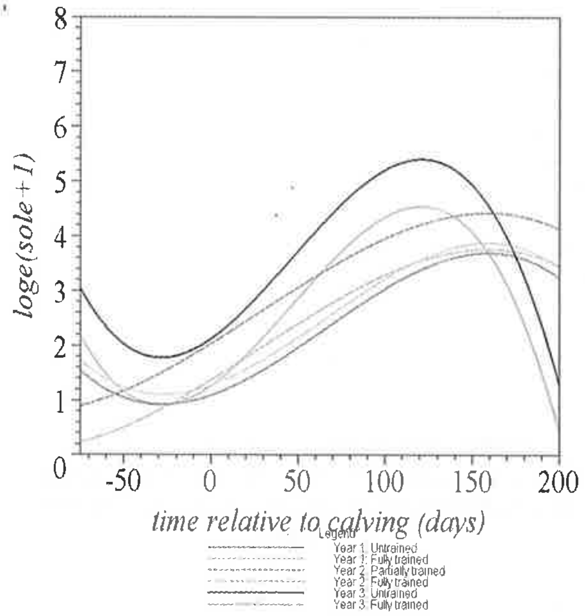
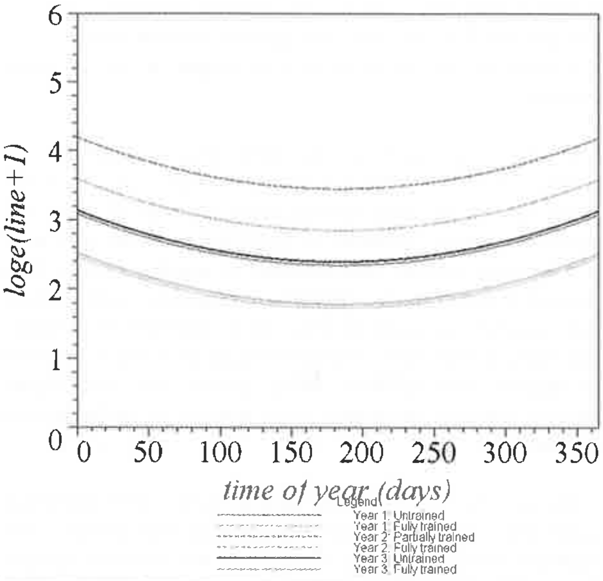


Figure 1. Fit for white line and sole lesion scores for the 6 treatment by year combinations, plotted as a function of time relative to calving for average time of year



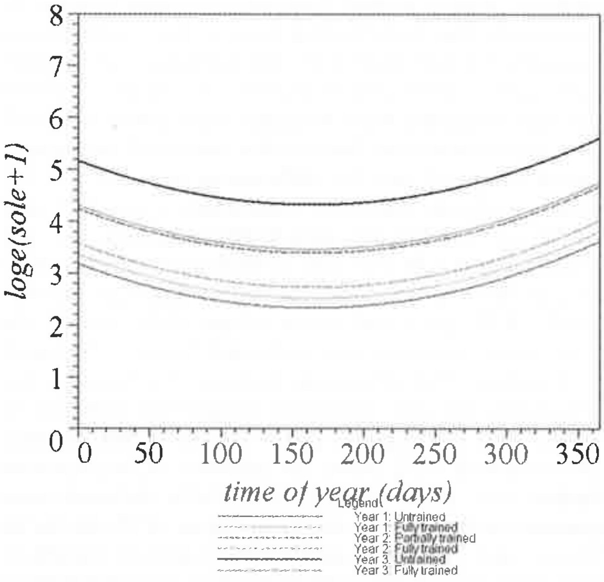


Figure 2. Fit for white line and sole lesion scores for the 6 treatment by year combinations, plotted as a function of time of year for average time relative to calving

Table 2. Predicted treatment means for white line and sole lesions (log transformed

	Untrained	Partially trained	Fully trained	Average SE Diff	P Value
White line lesions	2.92	2.67	2.29	0.329	0.041
Sole Lesions	3.50	3.75	2.95	0.317	0.007

The effect of training regime on claw horn lesion development was smaller than expected from field observations. It may be that the excellent building and cubicle design coupled with good stockmanship limited the extent of this effect.

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Acknowledgements: We gratefully acknowledge farm staff. SAC receives financial support from SEERAD

CLAW CONDITION AND MEAT QUALITY FACTORS IN FATTENING BULLS IN TWO DIFFERENT HOUSING SYSTEMS

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Introduction

Meat production is the economic goal in fattening cattle, and claw soundness is only a "by-product". Only a few studies investigated possible correlations between the condition of claws, meat quality and production factors (SUGG et al., 1996). In a study where bulls were fattened under standard, claw condition, claw diseases and daily weight gain, dressing-out percentage of carcasses and various factors of meat quality were investigated.

Materials and methods

In this study 60 Simmental bull calves were bought at Austrian Simmental Breeder Association auctions and fattened on the federal research farm Koenigshof. At the age of 75 days the calves were assigned to four groups of 15 bulls each. Two groups were kept indoors in a tethering system, while the other two groups were kept outdoors in free paddock with a shelter. In each housing system, two feeding regimes were applied, one with a pelleted complete diet, the other with maize silage and additional concentrate. All bulls were slaughtered at 450 days in the research farm's slaughterhouse to minimize any transport stress. The claw condition of all bulls was recorded on the 80th, 200th and 320th day and immediately after slaughter using a scoring system described by BOOSMAN et al. (1989) and modified by STANEK (1994). Various parameters were recorded during the fattening period including daily weight gain, feed intake and height at withers. Among the parameters that were recorded after slaughtering were dressing out percentage, percentage of significant meat cuts, shear force measurements of meat (as a parameter of tenderness according to Warner Bratzler) and several sensory parameters. The subjective sensory evaluations(taste flavour) were made by a 6 person test team, who used a testing procedure developed at the federal centre for meat research in Germany (RISTIC, 1987; GUHE, 1991). After thawing the meat in a refrigerator it was grilled up to a temperature of 70 °C. The trained team of three women and three men assessed the parameters like juiciness, tenderness and flavour with a 6 point scale.

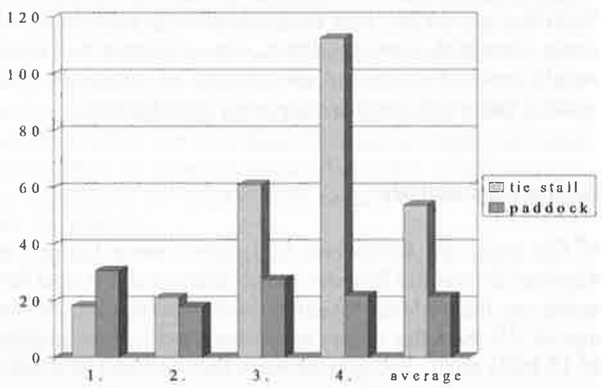
Results and discussion

Claw scores showed higher values in both subgroups kept tethered indoors, indicating more pathological conditions. Mainly pathological claw forms contributed to this fact. In the initial fattening period the claw condition was comparable in both housing systems.

The major differences between the housing systems were found towards the end of the fattening period (Table 1). The tethered group showed also a tendency to pathological conditions like sole ulcers and other types of septic conditions.

No significant influence of the feeding regime on the claw scores was detected. In general, claw scores were within the range found in well managed to average farms. The bulls of the paddock system fed with maize silage reached the highest average live weight before slaughter (669 kg) followed by the paddock system fed with pellets (622 kg) and the tethered group fed with pellets (597 kg).

Table 1: Claw score in different housing groups at mths 3, 7, 11, 15 (significant differences on mths 11, 15 and average)



The lowest weights were reached in the tethered group fed with maize (581 kg). The influence of the stable system was greater than that of the feeding regime. Average feed intake was significantly higher in the paddock groups. Average daily weight gain was highest in the paddock /maize group (1534 g / day 125 - 450). Dressing-out percentage was highest in both paddock groups (56.3%), as was the EUROP classification score. The area of rib eye muscle was larger in the paddock groups.

The following meat quality data also highlighted the superiority of the paddock group. Authors have had different points of view about meat quality of extensive housing systems. JURIE et al. (1998) found out that bulls in a tethered system were superior in meat quality to bulls housed in loose barns. In that investigation meat of bulls was tougher because of the higher content of collagen and flavour was rated lower because of lower content of fat depots. AUGUSTINI (1995) points out that higher feeding intensity increases meat quality of cattle housed on pasture.

In the current trial lower fat depots (inter- and intramuscular) were found too, but meat quality was better in the paddock system with less fat than in the tethered system. One theory to explain the lower fat depots of bulls in the paddock system is that they needed more energy for movement. The high feeding level in both systems and especially the high level of animal constitution in the paddock system contributed to a better meat quality. Some of the most important traits to assess meat quality are juiciness, tenderness and flavour. The results of the sensory evaluation point out the differences between the two housing systems. Significant differences in juiciness were found out between the two paddock groups fed with maize silage (4.24 pts.) and pelleted complete diet (4.23 pts.) on the one hand and the tethered groups fed with pellets (4.20 pts.) and maize silage (4.04 pts) on the other hand. Juiciness was noticeably better in the paddock system. The differences between the housing systems were not only significant in juiciness but also in flavour and total points. Cattle housed in the paddock system scored 4.23 points for juiciness, 4.19 points for flavour and 12.27 total points. Cattle tethered were assessed with 4.12 points for juiciness, 3.95 points for flavour and 11.95 total points. There was no significant difference in tenderness between the housing and feeding systems. Cattle from the paddock system reached 3.85 points compared to cattle from the tethered system with 3.89 points. AUGUSTINI et al. (1990) wrote about the high significance of feeding intensity and higher daily gains causing better sensory results. This statement agrees with the results in the current investigation. Warner - Bratzler Shear force values were collected to obtain an objective measure of meat tenderness. The lowest value was estimated for the paddock group fed with maize silage (2.73 kg), the highest for the group fed with pellets (3.45 kg). The tethered group fed with maize silage had a shear force value of 3.15 kg, the pellet group reached 2.98 kg. Significant differences were only detected within the paddock groups. Differences in meat colour were also observed between the housing and the feeding systems. Cattle fed with maize silage had lower values for L10*-brightness (39.20) than cattle fed with pellets (40.51). Compared with tethering, paddock cattle had higher values in a10-redness (10.66 vs. 9.70) and lower values in L10*-brightness (39.44 vs. 40.27). Cattle from the paddock system fed with silage had higher values (15.70 and 14.79) in Cab*-Metric chroma (colour saturation) than cattle from the tethered system fed with pelleted feed (12.70 and 13.61). Meat colour of paddock cattle was darker, more intensive red and showed higher Cab*-metric chroma.

Of major interest is the correlation between total claw score and meat quality traits. Claw condition in the tethered housing system is worse, with an obvious influence of the increase in body weight. The following correlations were estimated between total claw score on the one hand and pH-value, recorded 24 h p. m. (r=0.27**), drip loss (r=-0.18*), shear force (r=-0.14), L10*-brightness (r=0.09), a10-redness (r=-0.14) and flavour (-0.13***) on the other hand. The negative correlations between claw score, drip loss and shear force give the impression that meat quality increases with higher claw score. But dark cutting beef (DCB) is also characterized by more tender meat with lower drip loss but poor flavour. Although correlations are not closely related, the conclusion can be drawn that bulls with damaged claws and therefore higher claw score are placed under stress which has a negative effect on meat quality.

This investigation shows the great advantages of an open feeding and housing system with access to a paddock compared to a conventional tethered system, as still found in various countries in central Europe. Not only were fattening and slaughtering performances better but also meat quality. The correlations between claw condition and meat quality factors were not closely related.

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THE DEVELOPMENT OF A SOFTSEPARATOR™ FOR DAIRY CATTLE THAT IS EXAMINED FOR LAMENESS BY AN AUTOMATIC LAMENESS DETECTION SYSTEM

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Abstract

To obtain bodyweight measurements of cattle, herdsmen have to walk the herd through a scale, one animal at a time. This practice imposes extra work on the animal caretakers. Pneumatically activated entrance and exit gates and a passive S shape gate that is operated by the animals are viable commercially available solutions with one drawback. Gates slow down the animal traffic and introduce extra work for the animal caretakers. This paper introduces a softseparatorTM technology that eliminates the need for gates by decomposing a limb-movement-variable record of multiple cows into multiple records of a single animal. This technology works with a recently developed Lameness Detection System that was introduced in the 2002 International Symposium on Lameness in Ruminants. This patent pending system is being commercialized by Bou-Matic, LLC. The softseparatorTM technology enables the herd to freely walk through the lameness detection system in groups, yet the system keeps records of each individual animal in a separate file.

The current paper discusses the logic structure of the program, the structure of the recorded data, and the developed algorithms used. The separation of various numbers of cattle, walking freely in groups through the system, will demonstrate the system capabilities and its inevitable limitations.

Introduction

Bodyweight (BW) variations have been correlated to changing health condition of dairy cattle (Maltz et. al., 1997) and BW measurements have been one of the parameters producers record on dairy farms. BW measurements are obtained by walking the herd through a scale, one animal at a time, a practice that requires the animal caretakers to perform extra work.

Walk through scales deal with animal singulation by using pneumatically actuated entrance and exit gates or by a passive S shape gate that is operated by the animals. Slowing down the animal traffic and occasional necessary attention by the animal's caretakers are the main drawbacks of the gating solutions.

This paper introduces a softseparatorTM technology that eliminates the need of gates by decomposing a limb-movement-variable (LMV) record of multiple cows into multiple records of a single animal. This technology works with a recently developed Lameness Detection System that was introduced in the 2002 International Symposium on Lameness in Ruminants (Rajkondawar et al., 2002). This patent pending system is being commercialized by Bou-Matic, LLC. The softseparatorTM technology enables the herd to freely walk through the lameness detection system in groups, yet the system keeps records of each individual animal in a separate file.

Material and Methods

The lameness detection system generates animals' ID records (YYMMDD.id file) and ground reaction force (GRF) data (YYMMDDxxx.ana file). The id records determine the number of animals (n) recorded in each analog GRF file. If n=1 separation is not required. Nevertheless, if n>1, inductive animal separation procedure is performed. In this iterative procedure, the record of a single animal is rewritten into a separate file, and the original n animal record is rewritten to reflect (n-1) animals. This iterative procedure is performed j times, until (n-j) = 1 and no more separation is required.

The algorithm takes into account the fact that a single time-zone represents limb/floor contact of one, two, or three limbs at most. A time zone is defined as the time period during which there is a continuous contact between the floor and the animal's limbs. Two and three time-zone limbs undergo special operations that separate the GRF records of the multi limb time-zones into multiple zones of a single limb. After obtaining single limb time-zones the algorithm calculates seven statistical parameters associated with the contact locations and five parameters associated with GRF values. The former include average, minimum, and maximum values of the longitudinal hoof/floor coordinate, and the minimum and maximum values of the contact locations and their corresponding times of occurrence. Note that finite values (larger than a selected threshold) of varying hoof/floor contact locations reflect two or three limb time-zones. The GRF parameters, on the other hand, include maximum and average GRF values, number of positive and negative GRF slopes of a running window of selected size, and the number of times the GRF signature of a tested limb changes its slope direction from positive to negative values.

Through the examination of the twelve statistical limb parameters, the algorithm determines whether a tested limb is a fore (F) or a hind (H) limb. Once the limbs' pattern is identified, when a hind limb is followed by a fore one, the latter must belong to a new animal. This is true since the animals do not walk backward, when passing through the lameness detection system. By detecting F/H timings of the left and right floor plates, the GRF record of a single animal is identified and written into a separate file. By following this iterative process the GRF records of individual animals are rewritten into individual files and the animals are softly separated. This separation procedure is performed as soon as a GRF record is closed and its execution is in real time.

Results

As an example, the GRF records of cows 199 and 1027 are depicted in Figure 1. A red line identifies the end of the GRF records of cow 199 and the beginning of 1027. The two distinct GRF records are rewritten into two separate file as shown in Figures 2 (199) and 3 (1027), respectively.

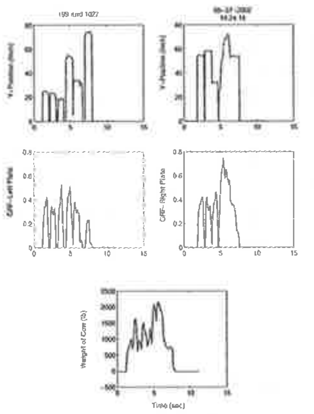

Fig. 1

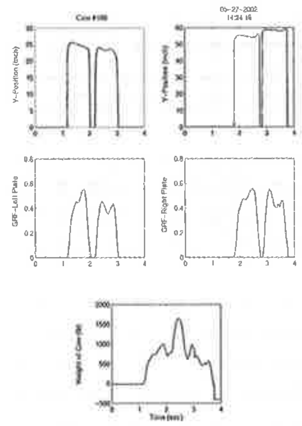

Fig. 2

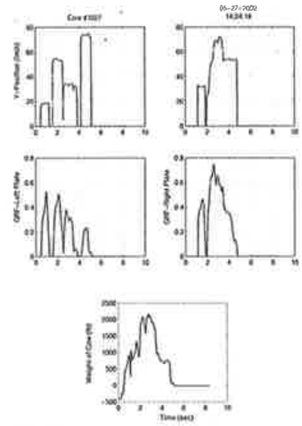

Fig. 3

Fig. 1. Cows 199 & 1027 walk through the RFD together. The softseparator marks (red line) the end of cow 199.
Fig. 2. The GRF signatures of cow 199 are written into a separate file.
Fig. 3. The GRF signatures of cow 1027 are written into a separate file.

Discussion

This paper introduces an algorithm that separates the GRF records of multiple cows into multiple files each containing the GRF records of a single animal. The GRF records are generated by having dairy cattle walk freely through a lameness detection system. This soft animal separator eliminates the need of gates that result in slowing down the animal traffic and impose extra work on the animal caretakers.

Acknowledgements

The authors acknowledge the support of Bou-Matic, LLC and the partial support of the USDA through award No. NRICGP 2000-2017. The authors also acknowledge the support of the University of Maryland Experiment Station.

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A POSTAL SURVEY OF THE INCIDENCE OF LAMENESS AND CLAW LESIONS IN DAIRY CATTLE IN THE UK: A PRELIMINARY REPORT

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Introduction

Lameness in dairy cattle reduces their welfare (Whay et al., 1997) and impacts on economic performance (Green et al., 2002). The incidence of lameness in cows in the UK has been estimated to be between 38 and 70 cases per hundred cow years (Clarkson et al., 1996; Hedges et al., 2001). Hoof lesions cause over 90% of lameness in dairy cattle (Murray et al., 1996) and UK studies have reported that sole ulcers and white line lesions are the most common hoof lesions in cows (Murray et al., 1996, Blowey et al., 2004).

This paper presents interim results from the first nine months of the UK arm of an EU project (LAMECOW OLRT-2001-00969). The ultimate aim of this study is to minimise lameness in dairy cows. The paper presented here describes the lesions that 155 farmers have recorded in their cattle during hoof trimming since February 2003.

Materials and methods

In February 2003, 497 UK dairy farmers were contacted from a database of farms used in a previous project (Peeler et al., 2002). They were invited to participate in a study investigating risk factors for lameness in dairy cows. We asked them to complete a form (Figure 1) each time they trimmed a cow's foot, for whatever reason. We also indicated that they would be asked to complete a questionnaire in early 2004. A total of 170 farmers agreed to participate in the study, 174 declined (62 of these were no longer in dairy farming) and 153 did not reply. Of the initial 170 participants, 155 have returned at least one lesion recording form. Fifty-three of these farmers have joined our more detailed longitudinal study (Barker et al., 2004). This paper presents the results from the lesion forms returned to date (October 2003).

Figure 1. The lesion form. Farmers complete one row per foot with at least one lesion when the cattle's feet are trimmed. They are asked to indicated the cause of lameness with a *

Cow number identity	Site of lesion	Foot affected	Lame	Lesions seen	Date today	Time in milk (weeks)	Who trimmed foot
	(place a cross)	(circle one only)	(circle one)	(circle all appropriate, star * cause)			(circle all appropriate)
			Sound	Sole ulcer White line			
		LF RF	Not sound	Digital dermatitis Foul			Farmer
			Definitely	Other (please state)			
			lame	______			Foot Trimmer
		LH RH	Hobbling				
							Vet

We ask farmers to assess whether the cow was lame using a four point score: sound, not sound, definitely lame and hobbling. We also request cow's identity, date of trimming, weeks in milk and who trimmed the foot. To reduce between farmer error we have provided a colour atlas with photographs of 16 types of lesion with a written description of each. To improve the validity of the reports (correct lesion identification) we ask farmers to indicate the site of the lesion with a cross on a diagram of the volar aspect of the foot, e.g. if a sole ulcer is present the cross should be in the sole not the toe region. This was sent to each participant. Forms are returned in prepaid envelopes at the end of each month. Continued participation has been encouraged by providing progress reports and prompt answers to queries, by phone if possible.

Data analysis. The occurrence of lesions is presented as the percentage of each lesion from all records with a lameness score of unsound, definitely lame or hobbling (hereafter called lame cows). The number of farms with no cases and the incidence rate of the four most common lesions; sole ulcer, white line disease, digital dermatitis and foul, are presented for those farms where herd size at the start of the study is known. The distributions of the four most common lesions are presented by month and by weeks in milk.

Results

Four farms have withdrawn from the study. The herd size of participating farms ranged from 27-350 cows (median = 84) at the start of the study. A total of 3623 records had been returned by 24th October 2003, 78% of which were for lame cows. A total of 1711 (56%) cows had a primary lesion identified excluding pre-calved cows. This group have been used for analysis below. Of these 1711 cows, 83% were recorded lame on one date only, 15% were lame twice and 1.5% were lame three or more times.

The most common lesions reported were sole ulcer (27%), white line disease (20%), digital dermatitis (16%) and foul (8%) (Figure 2). Incidence data for these lesions is presented in Table 1.

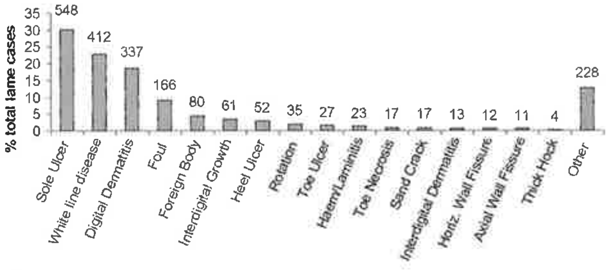

Figure 2. Percent of lesions by form from lame cows, including repeats (n=2043). Number of cases indicated above each bar.

Table 1. Incidence rate of the four most common lesions for the 95 farms with identified herd size.

Lesion	No. of farms with no cases	Minimum	Median	Maximum
	Cases/100 cows/Feb-Oct (farms with positive cases only)			
Sole ulcer	19	0.7	4.1	18.6
White line disease	24	0.4	3.7	16.7
Digital dermatitis	44	1.0	4.3	60.8
Foul	46	0.4	2.1	11.3

The most common lesions show a general decrease in incidence from the start of the study onwards (Figure 3), apart from foul, which is less consistent in its pattern of occurrence. Digital dermatitis was at its highest in the two months post-calving. Sole ulcer increased from 13- 24 weeks and white line disease was higher from 17 - 28 weeks in milk (Figure 4).

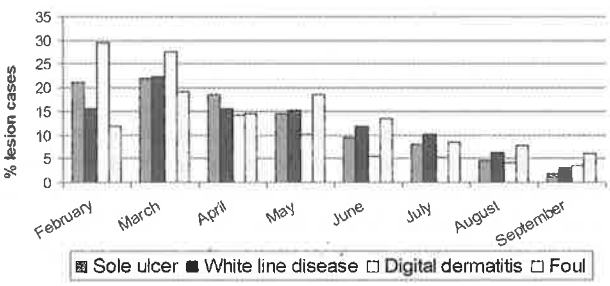

Figure 3. Percent of four most common lesions by calendar month

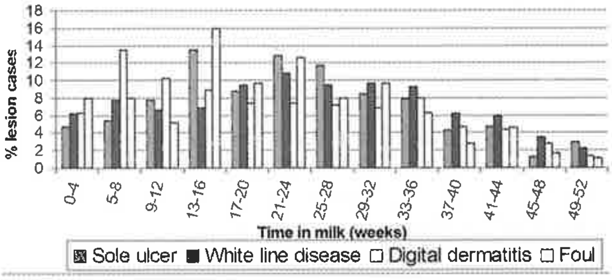

Figure 4. Percent of lesions by stage of lactation

Discussion

To improve quality of data the form was made as clear and simple as possible, with precise definitions of lesions and a colour atlas. There has been a high level of participation in the project, with only four farms knowingly withdrawing from the study. There is a reduction in the proportion of lesions by calendar month from February 2003. This may be because lameness is more common in the winter but may be due to a reduction in responses as the study continues. This is difficult to assess at this point as many farmers only return forms when cows are lame and so whether farms are still recording cannot be determined until the retrospective questionnaire is administered at the end of the recording period.

Sole ulcers and foul are at their greatest from three to four months post-calving as reported in other studies (Green et al., 2002; Blowey et al, 2004) and white line disease starts to increase about 4 weeks later. Digital dermatitis was most commonly found in the two months post-calving as reported in Blowey et al., (2004). This gives further credence to the hypothesis that the early lactation cow is either immunosuppressed or more environmentally challenged at this time than at other stages of lactation.

Further work

These results will be combined with the data from a postal questionnaire that will be sent out in early 2004 that will collect data on the environment and management of these cows. The high variation in lesions and lameness cases between farms suggests that these farms will provide a good sample for further study.

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DOES ROBOTIC MILKING AFFECT THE CLAW CONDITION AND THE OCCURRENCE OF LAMENESS IN DAIRY COWS ?

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Introduction

A future step in the automation of dairy herds will be the increased use of milking robots, a development that will be limited only by costs and herd size. Statistical problems arise in the adaptation of the AMS (automatic milking system) to small herds in Austria. Possible negative factors include the need for animals to learn new behaviour and to adapt their attitude to the system, waiting time in front of the AMS, combined with prolonged standing times and reduced human supervision. These factors can be considered as additional stress. No scientific study, has been published evaluating possible negative effects of this system on lameness and claw condition. Our special interest was focused on the direct comparison of two groups of dairy cattle kept under similar conditions except the different milking system (4). In a controlled trial on one experimental farm under otherwise equal conditions, a milk roboter (AMS) and a conventional milking system were compared as to management, behaviour, internal health and mastitis, as well as to the occurrence of lameness and the condition of claws (2). With respect to the conditions in Austria, Austrian Fleckvieh (= Simmenthal) and Austrian Braunvieh (=Brown Swiss) dairy cattle were used. The orthopaedic aspects are published here, while management, behaviour and economic questions are to be published elsewhere.

Material and methods

One large experimental stable was divided into two similar areas except for the milking system. In the cold stable made of timber, a cubicle housing system with 30 high boxes each (width: 120 cm, length 185cm, height of the floor 22,5cm, rubber mattresses, coverd with chopped straw; manure removal through slatted floor) was installed. In the AMS a selection door between the lying area and the AMS forced cattle to pass the AMS, if they wanted to get concentrate feed.

A herd of 60 cows, 30 Fleckvieh and 30 Braunvieh, was selected from the offspring of the experimental farm due to the AMS adaption of the cows. 30 Braunvieh cows were bought at auctions. During the recording period from March 2001 until October 2002 37 cows had to be replaced. The 60 cows were divided into two groups of 30 cattle (15/15 of each breed) : one group was milked in an AMS (Lely Astronaut F6, NL), the other group in a conventional herringbone parlour (HP). The average daily milk yield during the recording period was 20,8 l in the AMS group, 21,9 l in the HP group.

A locomotion and lameness score was recorded every 2 weeks over a period of 18 months, using a slightly modified MANSON and LEAVER (1989) system. Modification included higher grades 5, 5,5 and 6 for severe to no weightbearing lameness, and summarizing the subranges to a score between 0 and 4. Claw trimming on a rilt table was carried out every six months, starting in month 1. The condition of all claws of the animals was recorded using the BOSMAN et al. (1989) scoring system, modified by STANEK (1994) (3). Claw condition was recorded at the beginning of the claw trimming procedure by the first author 4 times, in months 1, 7, 13 and 18. In addition, heel horn erosions were scored separately. The statistical methods included Mann-Whitney-U-tests, Friedman-test and the Markoff-model.

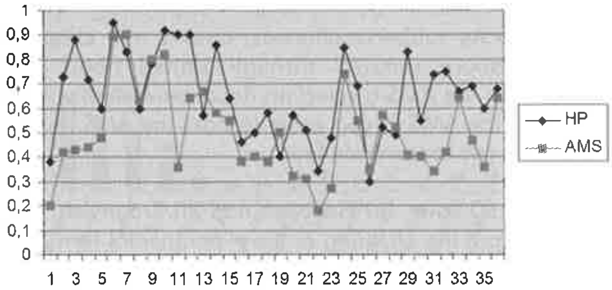
Results

The animal claw scores in comparison between the AMS group and the HP group are recorded in Table 1.

	AMS	AMS	AMS	AMS	HP	HP	HP	HP
Score #	1	2	3	4	1	2	3	4
min	0	0	0	0	0	0	0	0
max	22	33	32	66	64	33	49	92
median	5	6	6	7	4	8	4	7
Mean r	6	8	7	10	10	9	9	13
SD r	6	7	7	13	15	8	14	18

Generally, no significant difference was found between groups. Towards the end of the study a deterioration of the claw situation was seen in both groups, significant only in the HP group. Between groups, a significant difference (lower in AMS) of hemorrhages and sole ulcers was recorded only at time 1.

Tab. 1 Comparison of mean lameness score between AMS group and HP group on 37 recording terms during March 2001 and October 2002



The AMS group showed a higher incidence of heel horn erosion in the early phase of observation, whereas the HP-group had a higher frequency of erosion towards the end of the period. Lameness score was not different between groups (Table 1). At any time the percentage of lame cows was lower than 30%. Most affected cows were lame in the hind feet. Many cows had to be replaced for various reasons.

Discussion

The hypothesis was that the stress caused by adaption to the AMS system would induce a marked decrease in claw condition. This was investigated in a controlled comparison of two groups, kept under otherwise similar conditions under one roof of an experimental farm. The assumption was not confirmed. Generally, the claw score was similar to other studies. Neither the general claw score nor the analysis of the serious complications like sole ulcers revealed any loss of claw condition in the AMS. Claw judging was performed by the first author allowed a trendless comparison. The lameness scoring, that was performed independently from claw-scoring, revealed a similar situation. Heel horn erosion score increased in both groups, due to wet conditions.
Generally, neither a positive nor a negative effect of the AMS on the occurrence and severity of claw disease was recorded. In contrast to that fact, the culling rate was high. 37 cows had to be replaced, 23 of them in the AMS group, with 9 exhibiting udder problems or adaptive problems to the milking robot, 7 fertility problems and 3 due to claw problems.

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LINKING FARM PHYSICAL CONDITIONS, HERD MANAGEMENT AND COW BEHAVIOUR TO THE DISTRIBUTION OF FOOT LESIONS CAUSING LAMENESS IN PASTURE-FED DAIRY CATTLE IN NEW ZEALAND

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Introduction

New Zealand has a pasture-based dairy industry. Dairy cattle often walk further than one kilometre twice daily along gravel tracks to the milking shed from pasture. The cows congregate in concrete yards and stand for up to " hours as they wait to enter the milking parlour. Both these processes predispose the feet to damage. Herd sizes are increasing dramatically and it is not unusual for a herd of 400 or 500 cows to be handled as one group. The manner in which they are handled can further predispose the animals to foot damage. It is not surprising that greater the 75 per cent of lameness in New Zealand is traumatic in origin (Chesterton, 1988; Tranter and Morris, 1991).

The two most common lesions causing lameness are: damage to the sole (bruising, worn soles, penetrations of the sole) and separation of the white-line with subsequent abscessation under the wall ascending to the coronet. These lesions occur in front and back feet, inside or outside claw. This paper describes the occurrences of lesions causing lameness in 2468 cows.

Materials and Methods

The following information was gathered for each lame cow:

Age (first calver or adult cow)
Foot affected
Claw affected (medial/lateral)
Type of lesion

All the cows included in the study were those observed, treated and recorded by the author over a period of five years to 2003 from 60 herds.

Description of lesions:

White-line injury - separation of white-line with abscessation migrating proximally under the wall.
Sole injury - bruising, thin sole, penetration of sole
Axial wall lesion - injury or lesion involving axial groove or axial wall
Foot rot - interdigital necrobacillosis
Solar ulcer
Proximal to claw lesions - any injury or infection above claw
Miscellaneous - miscellaneous or undiagnosed lameness

Results

Table 1: Occurrence of types of lesions in 2468 lame cows:

White-line injury	(37.5%)
Sole injury	(27.9%)
Axial wall lesion	(13.5%)
Foot rot	(8.7%)
Solar ulcer	(1.5%)
Proximal to claw lesions	(5.6%)
Miscellaneous	(4.3%)

Detailed Analysis of Main Lesions from Table 1 :

Sole Injuries - First Calvers and Adult Cows have the same distribution.

Figure 1: Schematic Diagram of feet, showing claw involved.
Numbers indicate injury occurrence in percentages

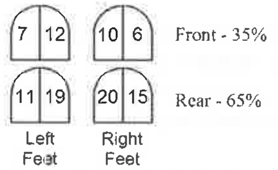
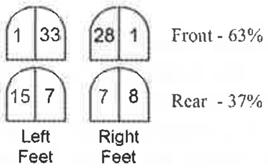
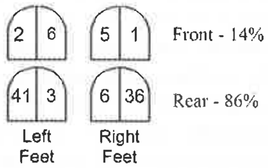


Figure 2: Schematic Diagram of feet, showing claw involved.
Numbers indicate injury occurrence in percentages



White-line injury - First Calvers

Figure 3: Schematic Diagram of feet, showing claw involved.
Numbers indicate injury occurrence in percentages



White-line injury - Adult Cows

Discussion

In a previous study traumatic lameness in general in New Zealand was found to be associated with track conditions and the patience of the herds-person (Chesterton et al, 1989)

The results in this present study show that by far the predominant type of lesions (of traumatic lameness) seen were white-line injury and sole injuries. (Table 1)

The distribution of sole injuries was found to be different to the distribution of white-line injuries. This suggests a difference in causes

Sole injuries occurred fairly randomly. In both heifers and adult cows the distribution was similar. Whatever the cause of sole injury in dairy cattle it was affecting young and adult cattle in a similar way.

Being prone to sole injuries seems to be linked to farm physical conditions.

The possible explanation of the distribution of sole injury (Figure 1) is as follows:
Both cows and heifers walking on long and damaging tracks wear the soles of their feet making them more prone to random bruising and penetration by sharp stones. The hind feet suffer more wear because they are the feet that provide the main propulsion, explaining the greater chance of a hind foot being affected.

The medial front claw is slightly more likely to be affected than the lateral claw. This may be because greater weight is carried by this medial claw in normal walking. However on the back foot the author cannot understand the greater prevalence of damage to the medial claw. One would have expected the lateral claw to have greater chance of damage.

With white-line injuries the distribution of the lesion was not random and varied greatly between first calvers and adult cows. (Figures 2 and 3)

In first calvers with white-line injury the front feet were more likely to be affected and overwhelmingly the medial claw. In adult cows the rear feet were more likely to be affected and overwhelmingly the lateral claw.

White-line injury appears to result from cow behaviour in response to poor herding management.

The possible explanation of the distribution of white-line injury is as follows:
When an adult cow is herded under pressure on a concrete surface, she leans against the cows next to her to push away from cows of higher dominance or from a potentially noxious situation (eg electric side fence, motorised backing gate, drains along the edges of the track). The adult cow pushing to the right leans to the right and uses her right hind foot for propulsion. Because she is leaning against cows on her right, the wall of the lateral claw, takes most of the force. This causes separation of the wall from the sole allowing gravel to enter the white-line. In a large proportion of cases the white-line of both lateral hind claws is separated because the cow sometimes pushes to the left and sometimes to the right. So the lesion is bilateral, not random.

When a first calver is herded under pressure on a concrete surface, she reacts differently to an adult cow. Instead of pushing sideways as do adults when trapped, she reverses out of the situation. The front feet provide the propulsive force. If the heifer is trying to escape backwards and away from say an electric fence on her left, she pushes with her left front foot. Because she is leaning at an angle away from the fence, greater weight bearing is taken by the medial front claw and the lateral wall in particular. This action again causes the separation of the wall and sole allowing gravel to enter at the white-line. Again the lesion is bilateral because the heifer is sometimes pushing from the left and sometimes from the right.

In veterinary practices in New Zealand we often find herds with a predominance of one type of lameness. Study of the distribution of injuries causing lameness in herds of dairy cattle points to links between the different types of lesions and farm physical conditions, management practices and cow behaviour. This will be of assistance in the diagnosis of causes.

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ASSOCIATIONS OF SOLE ULCER AT CLAW TRIMMING AND FERTILITY, UDDER HEALTH AND CULLING IN SWEDISH DAIRY CATTLE

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Introduction

Sole ulcers are caused by damage to horn-forming tissues in the sole corium and are an important cause of lameness (Murray et al., 1996). Most cattle with sole ulcer are, however, to some extent affected bilaterally, and the lesions may not be detected until claw trimming because lameness is not obvious. Sole ulcers are found at claw trimming in as much as ~10% of Swedish dairy cows, mostly not recorded as lame (Manske et al., 2002). Studies of lame cows with sole ulcers have detected an association with decreased fertility (Collick et al., 1989). The present study aimed at clarifying the relationships between the presence of sole ulcer at routine claw trimming, and measures of reproductive performance, udder health, and culling during the same lactation in Swedish dairy cattle.

Materials and methods

One-hundred and two study herds were selected at random from the replies to a questionnaire delivered to all 4204 dairy farmers of five counties in southwestern Sweden (reply frequency 47%) (Hultgren et al., 1998). For details of herd and animal inclusion criteria and selection procedures, see Manske et al. (2002). The hoof health of cows in study herds was examined (by a claw-trimming technician (71%) or by one of the authors (TM, 21%; CB, 8%) at routine claw trimming 1-4 times during the housing seasons 1996-97 and/or 1997-98. The presence of a sole ulcer was defined as exposed corium through a defect in the solar surface of the claw capsule. A lactation was considered affected (ULCER=1) if sole ulcer was found in at least one foot. Cows with sole ulcer were consistently treated with corrective trimming and application

of a plastic block (Cowslips®; Giltspur Scientific, Ballyclare, N. Ireland) to the unaffected claw.
In this study, cows trimmed 60 to 180 d after calving between October and May were included. Lactations without such lesions were considered unaffected (ULCER=0). Only the first complete lactation of each cow was included. The number of lactation records obtained was 2368. The analysis was performed at cow level, accounting for herd-level clustering by introducing Herd as a random effect in mixed-regression models.
The associations of first-service conception rate (binary), number of services per pregnancy (count), number of days from calving to the successive calving (continuous, log-transformed), case of anoestrus (binary), clinical case of veterinary-treated mastitis (binary), high milk somatic-cell counts (udder health class score ≥ 6 [Brolund, 1986] at least once; binary), and culling (binary) in the studied lactations to sole ulcer during the same lactations were examined.
Each lactation started with a calving and ended with either a subsequent calving in the home herd (63%), culling (34%), leaving the home herd alive (2%), or remaining in the home herd >465 d after calving without calving again (1%). Potential confounders considered were: year when trimming took place (1996-97, 1997-98), breed (SLB, SRB, other/mixed breeds), milk yield index (continuous), parity (1, 2, ≥ 3), calving-month category (Febr.-July; Aug.-Oct.; Nov.-Jan.), days from calving to first service (continuous), anoestrus, high SCC, and occurrence of other clinical (veterinary-treated) diseases at least once during the lactation (0, 1; one variable for each disease trait).
The associations between ULCER and each outcome were analysed separately using the MIXED procedure (for continuous outcomes) or GLIMMIX macro (binary and count outcomes) in SAS 8.01 (SAS Institute Inc., 2000). In all, 7 mixed models were fitted, in each case specifying herd as a random effect and all other covariates as fixed effects.

Results and discussion

Sole ulcer was found in 15% of the cows (whereas 6% of the cows were lame). Sole ulcer was associated with a lower first-service conception rate during the first study year (OR=0.59; PWald=0.001), a prolonged calving interval (2% longer; PLR=0.013), and a higher odds of receiving treatment for anoestrus (OR=1.61; PLR=0.024). A borderline-significant association was found between sole ulcer and the number of services per pregnancy (10% higher; PLR=0.053). There were no significant relationships between sole ulcer and udder health or culling.
Relationships between hoof disease and infertility have been investigated and found in many studies (e.g. Barkema et al., 1994; Sprecher et al., 1997; Hernandez et al., 2001; Melendez et al., 2003). These studies were based on clinical cases of acute lameness. In our study, the lactations were categorized as either diseased or non-diseased based on records from routine claw trimmings during a critical part of the lactation, and regardless of any locomotor disturbances. To our knowledge, this has not been done before.
It is generally agreed that poorly trimmed hooves and lameness impair the lying-down and rising behaviour and thereby act as risk factors for teat injury (Rajala-Schultz and Gröhn, 1999), which in turn are very powerful predictors of clinical mastitis (Oltenacu et al., 1990; Elbers et al., 1998). Associations between poor foot health or poor hoof care and impaired udder health were found by □ster□s and Lund (1988), Ekman (1998), and Vaarst et al. (1998). In the present study, we did not find evidence of associations between sole ulcer and the occurrence of clinical mastitis or high milk-cell counts. This lack of relationship might be due to our cases being less severe. All participating herds were on routine claw-trimming programs and lesions were identified and treated promptly. Moreover, the longitudinal design of the main study, in which data were collected, might have increased the awareness of hoof health matters in participating herds (Ducrot et al., 1998), and thus resulted in more rapid treatment of affected cows. The treatment of sole ulcers may also have been more careful than in general claw-trimming practice, e.g. by a more frequent use of plastic blocks. Thus, the study design may have led to an underestimation of the strength of the studied associations by decreasing the severity of lesions and promoting effective treatment.
This study provides evidence that sole ulcer is related to impaired reproductive performance in the same lactation.

Acknowledgements

The authors gratefully acknowledge the help of all 22 participating claw trimmers, especially Anette Svensson, who made most of the lesion scoring. We also thank the 102 dairy farmers for giving us access to their facilities and herds, Dr. Hans Gustafsson for advice on fertility measures, and Dr. Henrik Stryhn for statistical advice regarding mixed modelling of categorical data. The study was funded by the Swedish Farmers' Foundation for Agricultural Research and AGROVÄST.

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THE INFLUENCE OF REARING ENVIRONMENT ON THE BEHAVIOUR OF HEIFERS IN CUBICLES

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Introduction

The environment encountered by dairy heifers during their rearing period can have a significant impact on the development of hoof-horn (Livesey et al 2000). However, the influence of rearing on hoof-horn may have a behavioural as well as an environmental component. This study was designed to allow the relative importance of these two factors to be established, and to provide significant information on how rearing affects the way heifers adapt to cubicles post-calving.

Materials and methods

Animals

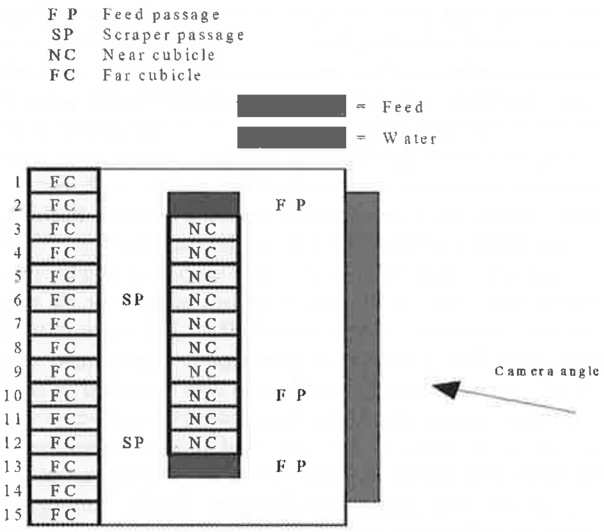
60 Holstein heifers were randomly assigned to cubicles or straw yards from first service until calving. During this period all heifers in the straw yard group had two weeks on cubicles, so that they could be trained to use individual feeders.

Behavioural monitoring

Heart rate monitors (Polar Vantage, Polar Electro) were fitted to 5 heifers from each rearing group in the pre-calving period, and to 10 heifers from each rearing group at housing in the milking group, and 8 weeks after calving. The heart rate monitors recorded heart rate (as beats per minute) every 15 seconds for a period of 48 hours.

Pedometers (Digi-walker, Yamax Ltd.) were fitted to 5 heifers from each rearing group in the pre-calving period, and to 10 heifers from each rearing group at housing in the milking group, and 8 weeks after calving. The pedometers were used to record the number of paces over a 48-hour period. Therefore one datapoint was recorded per cow on each occasion

One 25-cow cubicle yard was assigned for video recording. The yard plan is shown in below.



Video recording took place on three occasions one month apart. Recording periods lasted for 48 hours. The videotapes were viewed by trained observers and scan samples were made every 10 minutes with the videotape fast-forwarded between samplings. Observed behaviours were categorised as 'standing' (feeding/drinking vs. ruminating/idle) or 'lying in cubicle (correctly vs. incorrectly)

Results

Pedometers

Table 1 shows the effect of treatment and time post partum on activity, as measured using pedometers. There was no significant effect of treatment on pedometer reading (P=0.8). Heifers in both treatment groups showed an increase in pedometer reading between pre and post partum. This effect of time was almost significant at the 5% level (P=0.06), but there was no significant interaction between treatment and time (P=0.5).

Table 1 Effect of treatment and time on mean pedometer readings (metres) (SEM)

	Time of measurement		
Treatment	Pre-calving	Week 1	Week 8
Cubicle	2305 (± 465)	3124 (±501)	2796 (±260)
Straw	1664 (±333)	3496 (±955)	3526 (±486)

Video data

For each period Table 2 shows the total number of cow observations for each activity. This table shows the number of heifer observations where the identity of the group was observed and the number where no group identity could be determined. Analysis of the uncorrected data showed that there was a significantly higher proportion of heifer observations of standing in heifers reared on cubicles (P<0.001) than heifers reared in straw yards (59% of all observations as opposed to 56%). When the data were transformed using the non-identified cattle, this difference was still significant (P<0.001, 44% as opposed to 39%). The percentage of standing observations that were idling/ruminating was significantly higher in the cubicle-reared cattle than the straw-reared cattle (P<0.001, 46% as opposed to 35% uncorrected and 49% as opposed to 39% corrected). All identified heifers observed lying incorrectly in cubicles were in the cubicle-reared group

Table 2 The total number of heifer observations for each

	Standing						Lying					
	feeding / drinking			ruminating / idle			Correctly			Incorrectly		
	C	S	?	C	S	?	C	S	?	C	S	?
Time 1	448	460	451	335	344	601	292	594	2063	119	0	190
Time 2	510	407	495	415	158	524	476	300	2244	143	0	0
Time 3	501	402	381	481	181	357	669	597	2307	171	0	2
All	1459	1269	1327	1231	683	1482	1437	1491	6614	433	0	192

c = cubicle-reared heifers s= straw-reared heifers ?= not identified

Heart rate

There was no significant difference between mean heart rate on the first day of monitoring and the second day at any timepoint (P>0.4). Thus, only the overall mean for each treatment group was analysed. Table 3 shows the effect of treatment and time post partum on heart rate, as measured using heart rate monitors. The effect of treatment on heart rate was almost significant at the 5% level (P=0.06), with straw-reared heifers having lower mean heart rates at all three timepoints, although there were no significant differences at any timepoint (P>0.01) and by week 8 the difference between the means was only one beat per minute). Heifers in both treatment groups showed a significant change in heart rate with time (P<0.001). However there was no significant interaction between treatment and time (P=0.5).

Table 3: Effect of treatment and time on mean heart rate (bpm) (SEM)

	Time of measurement		
Treatment	Pre-calving	Week 1	Week 8
Cubicle	85.7 (± 2.39)	95.8 (± 3.52)	90.4 (± 2.66)
Straw	81.7 (± 1.41)	90.2 (±1.81)	89.4 (±1.21)

Discussion

This study showed no significant effect of rearing heifers in cubicles or straw yards on activity (as measured using a pedometer) or on heart rate. However, it did find a significant effect of housing type, during pregnancy, on behaviour after calving. Heifers reared in cubicles were significantly more likely to be seen standing than heifers reared in straw yards, and, when standing, they were more likely to be not eating or drinking. Furthermore, the only identified heifers observed lying in the cubicle incorrectly were heifers that had been reared in cubicles. All of these behavioural changes have been suggested as being associated with poorer hoof horn health, particularly an increased risk of hoof horn haemorrhages. However, in this study the reverse is the case as heifers reared on cubicles had significantly less hoof horn haemorrhages post partum. This suggests that the effects of rearing on hoof confirmation and hoof horn quality were able to overcome the effects on behaviour. Additionally, further research is required to identify why cubicle rearing affects behaviour in this way. The differences seen would seem to suggest that the straw-reared heifers were better adapted to cubicles than heifers that had been in cubicles from puberty until one month before calving. It is possible that this is because the heifers were reared in cubicles designed for cows and they learnt bad habits during the rearing period. This is almost certainly the cause of the significant difference in the number of heifers lying incorrectly in the cubicles. When younger, the heifers were able to lie head outwards in the cubicles, and once learnt this behaviour persisted in around 30% of heifers after calving. However, why rearing in cow-sized cubicles increased standing is unclear.

LONGITUDINAL STUDY INTO EFFECTS OF CLAW DISORDERS ON LOCOMOTION AND BEHAVIOUR IN DAIRY CATTLE

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Introduction

Lameness in dairy cows is a serious welfare problem. Almost 90% of all lameness cases originate from claw diseases. The concrete floor is considered an important factor in the development of claw disorders and lameness. In a previous study, we demonstrated that soft surface was most beneficial to claw health and prevalence of claw lesions differed significantly between traditional concrete flooring systems (Somers et al., 2003). The number of cows affected by claw lesions was on average 70 to 80 percent.

We performed a longitudinal study to investigate the effect of claw lesions on locomotion and behaviour over time. The effect of type of flooring was also taken into account. This paper highlights the effect of digital dermatitis and heel erosion on lameness as well as lesion development over time in both diseases.

Material and methods

The longitudinal study was performed on 12 dairy farms. The farms had either a cubicle house with slatted floor (n=3), solid concrete floor (n=3), grooved floor (i.e. emission-reducing floor system existing of flat concrete floor elements with grooves right-angled to the span of the floor elements and equipped with a manure scraper) (n=3), or a straw yard (n=3). Per farm, twenty cows were selected randomly but stratified for parity (1, 2, 3, 4). Hind hooves were examined at monthly intervals between March 2002 and May 2003 for severity and extent of heel erosion (HE; scale 0-4) and active lesions of digital dermatitis (DD; scale 0-2). Lesion score per cow for HE and DD was calculated as the sum of HE or DD score in both hind feet. The presence of interdigital hyperplasia and ulceration in the sole and white line was recorded as well. After claw examination, the locomotion of each cow was scored on a scale of 1-5 with half points according to Manson and Leaver (1988). Cows were trimmed at the start of housing (September/October 2002). This paper presents preliminary results of six monthly measurements, starting two weeks after claw trimming.

Results

Lesions of HE develop gradually over time. Mean lesion score for HE increased from 1.0 by two weeks after claw trimming to 2.8, 3.8, 4.2, 4.6, and 4.9 at 6, 10, 14, 18, and 22 weeks, respectively.

Changes in mean DD lesion and locomotion score after claw trimming are shown in Figure 1. Both mean DD lesion and locomotion score were lower in straw yards at any time. At week 6, 10, 14, and 18, locomotion score was significantly lower ($P < 0.05$) in straw yards than on slatted floors (reference). Locomotion score was always highest on grooved floors, with $P < 0.05$ at week 10, 14, and 18.

Then locomotion score was classified into three categories: normal (score 2), tender (score = 2.5), and disturbed/lame (score 3). Overall, about half of the cow population showed a normal locomotion, a quarter had an uneven gait with tenderness of feet, and 23% was lame. The cows in a straw yard had by far the highest proportion of normal locomotion, as well as the lowest number of lame cows (Table 1).

Table 1. Distribution of cows with a normal, tender, and disturbed locomotion on different floor types.

Locomotion	Slatted floor	Solid concrete	Grooved floor	Straw yard
Normal	54.8	45.7	34.7	82.5
Tender/at-risk	24.jan	27.jul	25.jul	16.jul
Disturbed/lame	21.jan	26.jun	39.7	0.8

The percentage of lame cows two weeks after claw trimming was high in cubicle houses: 18, 20, and 32% on slatted, solid concrete, and grooved floors, respectively. As housing season progresses, lameness increases slightly on slatted floors (up to 24%), while on solid concrete and grooved floors lameness cases increase considerably during the following four months (up to 35 and 51%, respectively).

When comparing DD-negative cows with those who are slightly (lesion score 1) and seriously (lesion score 2) infected, the proportion of normal locomotion declined from 64 to 47 and 33%, respectively, while lameness increased from 15 to 24 and 37%, respectively.

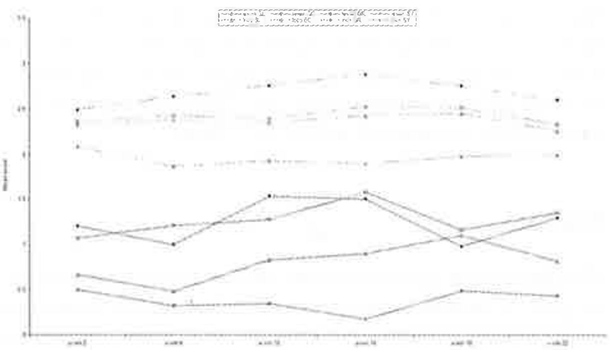

Figure 1. Changes in mean lesion score for digital dermatitis and locomotion score with weeks after claw trimming in dairy cows on slatted floors (SL), solid concrete floors (SC), grooved floors (GR), and in straw yards (SY).

Discussion

Over a third of the cows that had a serious DD infection were lame. This confirms that DD is an important disease associated with pain and discomfort, and often resulting in lameness. The presence of DD has increased considerably over the last ten years in The Netherlands, resulting in a current prevalence level of about 30 percent in cows kept in cubicle houses (Somers et al., 2003). In the present population, prevalence of DD among cows in cubicles houses was even higher: 44%.

Lesions of HE become more severe with time after housing. As a consequence, the stability and cushioning function of the heel becomes less properly and may result in pain en discomfort at walking. This may explain the increase in disturbed locomotion during the housing season.

Normal locomotion was most frequently observed in straw yards. This may be due to a better claw health effected by a drier hoof environment in the deep litter area. The lower mechanical stress to the claw sole because of a soft surface might also help. In addition to good lying comfort, the soft area relieves cows' claws and subsequently, cows may have less difficulty facing the uncomfortable concrete floor.

Claw trimming is generally suggested an effective tool in the treatment of claw diseases. It restores the original claw shape, and subsequently provides a more natural pressure distribution, which is essential to perform normal locomotion (Toussaint Raven, 1989). Nevertheless, prevalence of lameness two weeks after claw trimming was surprisingly high in cows kept in cubicle houses with concrete flooring. This result indicates that there is a growing need to explore the applicability of local soft surface areas in traditional concrete housing systems as concrete flooring often result in impaired locomotion and thus being detrimental to animal welfare.

Acknowledgements

The Dutch Technology Foundation (STW) and the Ministry of Agriculture funded this work. We thank the dairy farmers for their co-operation.

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PRIORITISING RISK FACTORS FOR DAIRY HEIFER LAMENESS THROUGH ITERATIVE CONSULTATION

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Abstract

A questionnaire survey was carried out to prioritise the risk factors associated with lameness affecting dairy heifers. Respondents were asked to score the importance of risk factors in the development of lameness associated with 4 classes of lesion and with reference to two distinct time points; birth to three months pre-calving and three months pre-calving to three months post-calving. The responses received gave some ranking of on farm risk factors for lameness in dairy heifers and provided the basis of a mechanistic approach to identifying points of lameness control on farm.

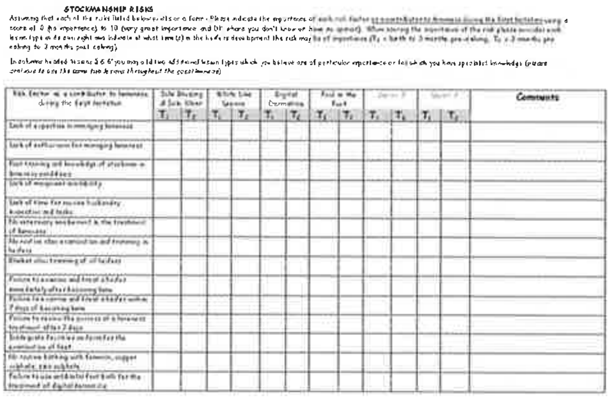
Introduction

Dairy heifers have been identified as a high-risk group for deterioration in claw health (Webster, 2002) so represent an important stage in the process of lameness prevention for dairy cattle during their first and subsequent lactations.

The survey described is the initial stage in the development of a process for identifying risk factors for dairy heifer lameness on individual farms and determining those factors which represent critical points in the control and prevention of lameness. The survey was carried out using the Delphi consultation technique (Linstone & Turoff 1975). This involved the use of postal questionnaires to gather the opinion of people who had demonstrated a professional interest in cattle lameness through publication or presentation of work at one of the three international symposia on lameness in ruminants held since 1998. A postal survey was used because it allowed consultation with a broad base of international opinion and did not require those consulted to meet. In addition the Delphi consultation is an iterative process in which the summarised results of the first round of consultation are fed back to the panel of respondents so they can further rank and comment on the views of all other panel members. In this way, some consensus of opinion is reached.

Methods

The initial questionnaire was compiled as a list of possible risk factors present either during the heifer rearing period from birth to three months pre-calving and/or at the time of first parturition from three months pre-calving to three months post-calving. The risk factors were subheaded as rearing strategy and general management risks, nutrition risks, feeding system risks, housing management risks, outdoor risk factors, animal-based risks and stockmanship risks. Respondents were asked to consider each risk presented with reference to the time period when it may be present and consider the importance of the risk factor for lameness during the first lactation. The four common lesion types associated with heifer lameness presented to the respondents for consideration were sole bruising and sole ulcer, white line lesions, digital dermatitis and foul-in-the-foot. The importance of each risk factor was scored on a scale of 0 (no importance) to 10 (very great importance) as a contributor to lameness during the first lactation with specific reference to four lesion types commonly associated with lameness in dairy heifers. An example of the questionnaire is shown in figure 1.


STOCKMANSHIP RISKS

Comments

Figure 1 The questionnaire filled in by all respondents. Each page contained risk factors associated with a specific risk category; general management risks, nutrition risks, feeding system risks, housing management risks, outdoor risk factors, animal-based risks or stockmanship risks.

The zero to ten scores ascribed to each risk factor in the first survey were then summed and ranked from highest to lowest. The top twenty risk factors for each lesion type during each time period were listed in order of rank and returned to the respondents. The respondents were asked to indicate whether they agreed with the ranking of risk factors and if not indicate how they would change the ranking.

The outcome of this iterative process of prioritising risk factors was then applied, using the rationale of HACCP (hazard analysis, critical control point) (Bell et al. 2003), to develop pro forma for the assessment of risk factors which could be applied at the farm level.

Results

Fifty-four initial questionnaires were sent electronically or by post to potential respondents. Completed responses were received from 22 people, a further 8 people returned either partially completed questionnaires or replied that they did not feel able to complete the questionnaire.

In total the questionnaire identified 110 potential risk factors for heifer lameness which were scored at differing levels of importance as risks for the four identified lesion types. The maximum possible total score for a risk factor would be 220 if all respondents had given a score of 10. In all cases the three months pre-calving to three months post calving period were given higher total scores for the risk factors than the same risk factors being present during the period from birth to three months pre-calving. The three factors most frequently identified in the top twenty risk factors as being of greatest importance were (in order) stockmanship, housing management and nutrition. The second round of the questionnaire gave respondents the opportunity to comment on the ranking of the top twenty risk factors for each lesion type at both time points. Many respondents took the opportunity to make some adjustment to the order.

Discussion

This process of consultation has provided a comprehensive but unwieldy expression of opinion as to the identity and relative importance of individual and multiple risk factors for lameness in heifers. Inspection and analysis of all this information revealed that many different risk factors could be placed in the same category; e.g. maintenance of good foot hygiene. Other named risk factors, e.g. poor stockmanship, were obviously important but could not be related to critical control points. By a process of refinement we were able to categorise all this information into nine distinct mechanisms involved in the aetiology of lameness and thereby define nine critical control points for the management of risk.

This framework of nine critical control points (e.g. biosecurity, foot hygiene, routine foot care) is used to identify the most important points of risk on individual farms and recommend options for their control. These will form the basis of a Lameness Control Plan, devised by farmers in association with their own veterinarians. The Bristol group has identified risk factors for 60 dairy farms as the basis for an intervention study whereby control options will be exercised on 30 of these farms. We shall assess the outcome as measured by compliance with the control programme, foot lesions and lameness after twelve months.

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ENVIRONMENT: LAMENESS AND MASTITIS

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Abstract

Lameness and mastitis are the most costly diseases in milk production. Farmers are often suffer economic losses in milk production due to an inadequate management of the herd. The environment in stables is very important for development of various lesions and infections of the extremities and the mammary gland in cattle. One crucial factor is improper design of the cubicles. Due to short beds, claw and udder lesions are very common, despite adequate preventive measures. Injured areas are often colonized by different microorganisms, including Staphylococcus aureus (S. aureus), which is very frequently isolated from the animals skin and from various surfaces in the stable. S. aureus is a common cause of infection in the bovine mammary gland and also an important pathogen in humans. The percentage of S. aureus udder infections in Slovenia was during the past years about 50%.

In our recent study a total of 1282 milk samples taken from clinically healthy udder quarters were examined. S .aureus was isolated from 150 (11.7%) samples and other major pathogens from 158 (12.4%) samples. The results of sensitivity tests showed that all S. aureus strains were sensitive to kanamycin and oxacillin and 88% of strains were sensitive to Amoksicillin + clavulanic acid. 31% of the strains were resistant to ampicillin and 36.8% to penicillin.

We conclude, that most farmers underestimate the impact of environmental factors on the health of cattle.

Introduction

In an international and wider perspective, mastitis and lameness can be assumed to be the most important production diseases. This refers to the incidence and also to the considerable animal welfare issue. Depending on detection rate, the frequencies differ from herd to herd and from country to country (Clarkson 1996).

The health of hooves, legs and the mammary gland is of crucial importance for dairy performance, longevity and for production economy. It influences the animals' well being and is therefore of concern not only for the animal and dairy industry, but ethical aspects also make it of concern for society and consumers. When lameness and clinical cases of mastitis affect among more than 10% of the cows in a herd it could be considered to be a herd health problem. A cow suffering from lameness and/or mastitis loses her rank in the herd and changes her eating behaviour, which could affect the performance. A negative energy balance, is often the result and implies reduced milk production and body condition.

Compared to cows' natural environment when grazing, today's confined dairy systems hardly satisfay requirements for comfortable lying, standing and walking. Hygiene is often poor. A higher risk for lameness and mastitis is found in large and high producing herds especially when housed. There is no indication that production will decrease in the future and tomorrow's management systems must thus be planned for even higher demands than they are today (Bergsten 1996).

Material and method

Data for our research were collected on 18 dairy farms across Slovenia. 350 cows were examined and 1282 milk samples were collected. Samples were bacteriologically examined on blood agar plates and the agar diffusion method was performed for susceptibility testing. On dairy farms a questionnaire for farmers was used to determine the influence of various factors (environment, treatment, preventive measures, animal) on the prevalence of S. aureus infections. In this article only environmental impact will be discussed. In the questionnaire the environment was analysed on the basis of cubicle design, pasture conditions, milking techniques, milking machine functions, bedding materials and climate conditions.

Results

A total of 1282 milk samples taken from clinical healthy udder quarters were examined. S. aureus was isolated from 150 (11,7%) samples and other major udder pathogens from 158 (12,4%) samples.

Sensitivity tests showed that all S. aureus strains were sensitive to kanamycin and oxacillin and 88% strains were sensitive to Amoksicillin + clavulanic acid, while 31% of strains were resistant to ampicillin and 36.8% to penicillin.

Statistical analysis of the results indicated that the influence of the environment was more important than other factors (preventive measures, diagnosis, treatment, animal).

Table 1. Statistical analyses of the questionnaire with ANOVA

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
PREVENT	Between Groups	8996,604	14	642,615	1,397	,494
	Within Groups	920,205	2	460,102		
	Total	9916,809	16			
ZD.DG	Between Groups	4936,962	14	352,640	4,166	,210
	Within Groups	169,280	2	84,640		
	Total	5106,242	16			
OKOLJE	Between Groups	11831,249	14	845,089	33,324	,030
	Within Groups	50,720	2	25,360		
	Total	11881,969	16			
KRAVA	Between Groups	1555,246	14	111,089	,346	,911
	Within Groups	641,225	2	320,613		
	Total	2196,471	16			

ANOVA-analiza variance
Prevent- preventive
Zd.dg-therapy and diagnosis
Okolje- environment
Krava-animal
*-largest influence

Discussion

Environment conditions are a link between lameness and mastitis prevalence in a herd. Our current study revealed a significant influence of the environment on animal welfare in general and particularly on the health status of the mammary gland. Harmful influences from the environment are very important in the development of various lesions and infections of the extremities and mammary glands in dairy cows. In our circumstances the shape of the cubicles was the most critical factor. In old-fashioned projected cowsheds the cubicles are mostly too short for the high producing dairy cows due to improvement in genetics of the animals. Different studies (Vokey et al, 2001; Manson 1989) demonstrate the important role that alley surfaces and stall beds play in providing optimal comfort to dairy cows. Rubber-surfaced alleys combined with concrete stalls, despite the favourable effect on claw net growth, is not recommended because several cows preferred to lie in the alleys, causing hygiene and mastitis problems. In our survey concrete stall surfaces were predominant. Due to short cubicles and inappropriate stall surfaces claw and udder lesions were very common. Among these examined dairy herds, S. aureus was the most frequently isolated pathogen from mammary glands. Matos et al. (1991) reported that sources other than the infected udder of lactating cows are probably involved in the epidemiology of S. aureus intramammary infections in the dairy herd. Although these alternative sources may well play a smaller role than the udder of infected cows in spreading the disease in a dairy herd, measures should be used to minimize the spread from such sources in order to control or eradicate S. aureus. In this study 40% of the 70 lesions sampled on the skin of adult cows were positive for S. aureus, showing that this organism readily colonizes wounds. In a similar survey, Roberson et al, (1994) reported that S. aureus appears to be ubiquitous in the environment and was isolated at least once from all general sites on dairy farms. It has also been shown that lame cows lie down more than healthy cows (Manson 1989; Singh et al. 1993) and thus have a higher risk for leg injuries, teat tramps and mastitis (Vaarst et al. 1998).

Lameness and mastitis remain a major problem in dairy cattle worldwide. In future the improvement of environmental conditions will be an important task in order to reduce their prevalence.

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INFLUENCE OF DIFFERENT FLOOR CONDITIONS ON CLAW DEVELOPMENT, METABOLISM AND MILK YIELD IN DAIRY COWS HOUSED IN STALLS WITH FREE COW TRAFFIC

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Introduction

Floor condition in cattle housing systems significantly affects performance. Concrete floors lead to an increasing number of claw disorders and leg injuries. Providing flooring which allows the claw to sink into its soft surface

covering the concrete floor as is the case on pasture might partly help to solve the above problems.
Therefore, research conducted at the Ludwig-Maximilians-University experimental farm in Oberschleissheim, Germany, evaluated the influence of soft floor conditions on claw development and milk yield in dairy cows.

Materials and methods

F1 cross-bred animals of German Holstein and German Fleckvieh (Simmental) were compared for the duration of one lactation period. The control group's floor was a conventional concrete slatted floor. The slatted floor of the experimental group was covered with precision cut rubber mats (system "KURA S", Co. Kraiburg).
Claw trimming is performed at predetermined intervals (day -21, 150, 305 of the lactation). At the same time linear measurements were taken, disorders were documented, and horn hardness was determined at defined points on each claw (Fig. 4).
A caliper (Co. Mitutoyo) was used to measure claw dimensions, claw angles were determined using a goniometer (Co. Frei) and the hardness of the horn was tested with a hand held hardness testing set for test standard Shore D ('Hardmatic series 811', Co. Mitutoyo) (Fig. 1).
The test standard Shore D includes materials such as harder plastics and harder elastomers.
The hardness scale ranges from 1 to 100. The scale of the hardness testing set and the hardness of the horn are proportional to each other.

Fig.1 Measurements at the claw with caliper, goniometer and hardness testing set

Results:

Initial results showed a trend concerning linear measurements of the horn capsule on soft floors. especially a higher value of the wall length (Fig.2) and an increasing angle of the bulb (Fig. 3).

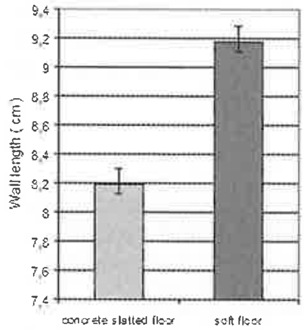

Fig.2 Influence of different floor conditions on wall length at day 150 of the current

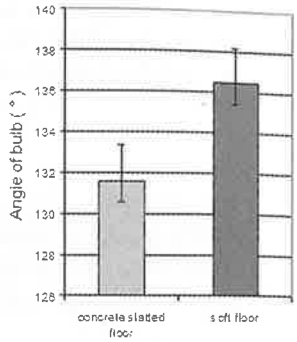

Fig.3 Influence of different floor conditions on angle of the bulb at day 150 of the current

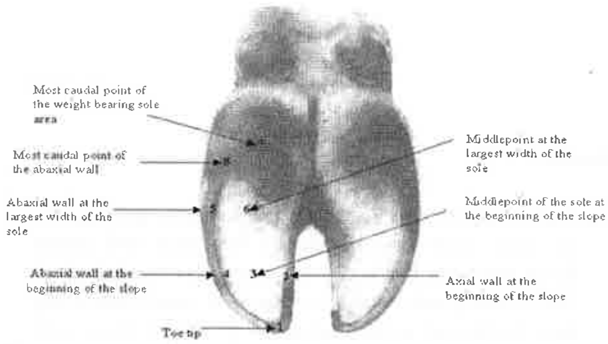

Fig.4 Location of the hardness testing points (modified after T.Raven 1977)

In addition the horn hardness measurements showed, that horn becomes softer on elastic floor (Fig. 5, 6). For explanation of the hardness testing points see Fig. 4.

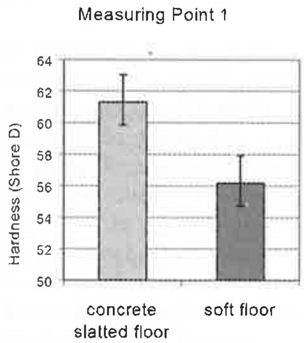

Fig.5 Influence of different floor conditions on the hardness of the horn at the tip of the toe
(Point 1) at day 150 of lactation

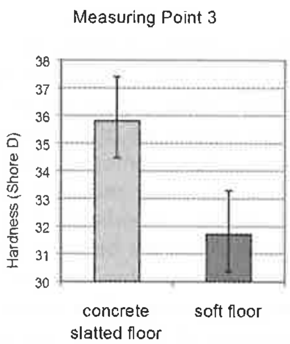

Fig.6 Influence of different floor conditions on the hardness of the horn at the midpoint of the cranial sole
(Point 3) at day 150 of lactation

Discussion

The preliminary results confirm that soft floor conditions have a significant influence on the development of the horn capsule. The abrasion of the horn is reduced due to sinking of the claw into the soft surface, followed by increased length of the horn capsule.
The angle of the bulb increases as a direct result of the