Effect of intensive application of topical treatments in the milking parlor on the prevalence of Digital Dermatitis

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Abstract

Digital Dermatitis (DD) is an infectious claw disease of cattle, reported as having a high prevalence in several countries. Currently, some different types of topical treatments are available on the market, but not all as effective. The objective of this study was to evaluate the prevalence of DD before and after intensive use of the topical treatments with four different products for DD in the milking parlor on 10 farms in Alberta, Canada. Subsequently, the effect of the treatments was determined on the distribution of each M-stage on every of the 10 farms. To achieve this all the hind feet were scored weekly in the milking parlor. In the first week if a DD lesion was present it was randomly assigned to one of the four different topical treatments: HealMax™, Hoof-Sol, negative control (saline with green food coloring), and positive control (oxytetracycline spray). All lesions were examined for 8 weeks, and if not cured re-treated with the same treatment as in the first week. There was no difference in overall DD prevalence after eight weeks of topical treatments over the 10 farms ($P=0.13$). However, the prevalence of chronic lesions (M4) increased on every farm, whereas the prevalence of active lesions (M2 and M4.1) decreased ($P<0.05$). Eight weeks of topical treatments under field conditions did decrease the prevalence of active lesions in a herd, but did not decrease the overall DD prevalence on a farm.

Introduction

Digital Dermatitis (DD) is an infectious claw disease of cattle. It is characterized by a superficial dermatitis, which causes painful ulcerations with a fetid odor. Since it was recognized by Cheli and Mortellaro (1974), several different nomenclatures for the disease have emerged such as Mortellaro disease, footwarts, hairy heel warts, strawberry or raspberry heelwarts, hairy hoof warts and papillomatous digital dermatitis (Read & Walker 1998). Nowadays, predominantly DD is used in scientific reports. The lesions are typically located in the interdigital ridge of the rear feet (Döpfer, Holzhauer and Boven, 2012). DD lesions can also be found around the dewclaws, the heel and along the coronary band. These lesions can cause lameness which will lead to production losses and reduced animal welfare (Döpfer et al. 2012; Laven & Logue 2006). For this reason, and its reported high prevalence (Holzhauer *et al.*, 2006), it is considered a significant problem in many intensively managed herds in the European countries: Italy, the Czech Republic, Slovakia, Denmark, Slovenia, Germany, Ireland, France, England, and the Netherlands. DD has also been confirmed in various regions of the USA, Mexico and Canada (Britt *et al.*, 1996).

Despite several scientific studies, the etiology is still not fully understood. The current hypothesis is that DD is multifactorial (Read, Walker and Castro, 1992) and spirochetes, more specifically the phylogenetic group of *Treponema* spp., play an important role in the pathogenesis of DD (Read, Walker and Castro, 1992; Gomez *et al.*, 2012; Krull *et al.*, 2014). However, other bacterial species such as *Fusobacterium spp.*, *Campylobacter spp.* and *Prevotella spp*. were also isolated from acute DD lesions (Krull *et al.*, 2014). Nevertheless, there is an interaction of causative factors, including spirochetes (Read, Walker and Castro, 1992), the host (Scholey *et al.*, 2010) and the environment (Wells, Garber and Wagner, 1999). To be specific, the hosts with the highest risk for DD are: high producing cows, cows in low parity, early stages of lactation and/or cows affected by interdigital dermatitis (ID) and interdigital hyperplasia (Holzhauer *et al.*, 2006). Environmental risk factors include housing cattle in freestalls with slatted floors, unhygienic and wet conditions, and the purchase of (sub)clinically affected cattle. In the cooler months of the year, when cattle are kept indoors, the prevalence of DD is higher (Rodriguez-Lainz *et al.*, 1999; Somers *et al.*, 2005). All these different factors make the control and elimination of DD very difficult and unpredictable (Laven & Logue 2006). Furthermore, the dynamics of DD lesions are complex, and understanding the lesion progression is an important factor in implementing control methods to achieve a manageable state of disease.

One of the systems used to categorize DD lesions is the 5 M-stages scoring system (Döpfer et al., 1997), running from M0 to M4.1. M0 is attributed to feet without visible circumscribed skin lesions. M1 refers to feet with red to grey circumscribed lesions, with <2 cm in diameter. M2 is the acute and active lesion, seen as an ulcerative (bright red) or granulomatous (red-gray) stage with a diameter >2 cm and often painful on palpation. M3 is attributed to feet in healing stage after topical therapy, which shows up with a scab over the acute lesion. M4 is the chronic stage, which appears with hyperkeratosis or proliferation of the surface. Stage M4.1 is characterized as a chronic lesion with a new M1 lesion within its perimeter (Berry *et al.*, 2012). Applying this scoring method will give a better knowledge of the severity and active or non-active stages of a DD lesion (Holzhauer et al. 2008). Another system used for herd monitoring and transmission models of DD lesions in clinical trials is the division of cows in Type 1, 2 and 3. This division is based on the repeated presence of M2 lesions in the same cow. A Type 1 cow does not develop an acute lesion (M2), but can have a M1 or a M4; a Type 2 cow develops an acute lesion but follows a "normal" infection (M0→M1 →M2→M3→M4→M0); and a Type 3 cow has repeated episodes of acute M2 lesions within a period of two to three weeks (Döpfer, van Boven and de Jong, 2004).

The overall aim of control methods for DD is the reduction of exposure of cattle to the infectious agent(s). There are different types of methods available that focus on herd treatment, such as footbathing (mainly for prevention), individual treatment, via topical application, or individual systemic treatment (Laven & Logue 2006). Footbathing is considered the most accepted prevention strategy (van Amstel and Shearer 2006). Footbath treatments can be done using formalin, copper sulphate, zinc sulphate, antibiotics and disinfectants such as glutaraldehyde and hydrogen peroxide (Laven & Hunt 2002; Laven & Logue 2006). Combinations of these compounds are possible; however, neither antibiotics nor formalin and copper/zinc sulphate are recommended because of their hazardous characteristics. The use of footbaths with antibiotics is not off-label and environmental implications of copper and zinc sulphate have resulted in restricting the concentrations of copper sulphate (0.5%) and zinc sulphate (1%) (Holzhauer et al. 2008). The use of antibiotics (lincomycin and particularly oxytetracycline (OTC) spray or powder) for topical treatment is still common (Laven & Logue 2006). The spray application will provide a fresh treatment for each treated cow without the risk of contamination, as is the case for footbaths. However, the environmental limitations and public health concerns have necessitated the development of topical treatments without antibiotics or copper and zinc sulphate. One possible treatment is a topical disinfectant with glutaraldehyde. This product is used as a chemosterilizer and disinfection product, also used for leather tanning and tissue fixation for electron microscopy. Glutaraldehyde has a broad spectrum of activity and a fast killing rate for bacteria, including bacterial spores, various types of viruses and both mycelia and spore forms of fungi (Ellis and West, 1976). One of the products on the market with glutaraldehyde as the active ingredient is HealMax™[1] Herd Spray or HealMax™ Wart Spray. According to the product indications both are sprayed directly on individual DD lesions, for 1-

[1] AgroChem Inc., http://www.agrocheminc.com/index.php/products/category/healmax

2 times (either on the same or consecutive days). Three to five sprays provide sufficient product to cover the entire lesion. Another alternative is Hoof-Sol[2], a topical spray with copper and zinc chelates, as active substances, and aloe vera plant extracts. The company Intracare uses chelates to reduce the environmental implications as it is more stable and soluble. The protocol recommends applying a solution of 50% on the lesion by mixing Hoof-Sol with cold water. Although there are several treatment options, there is little evidence that these topical treatments are efficacious, and their effect on the DD prevalence in a herd when routinely applied is not known. Few studies on the efficacy of antimicrobial and some non-antibiotic topical treatments in the parlor (not the recent treatments available on the market) have been completed (Shearer and Hernandez, 2000; Berry *et al.*, 2010). Also, no scientific evidence is available about the comparison of the current topical treatments in parlor versus topical treatment with the same products in chute. On the other hand, studies about the prevalence (and epidemiology) of DD in herds have been conducted in different countries (Cramer et al. 2008; Holzhauer et al. 2008; Gomez et al. 2012; Berry et al. 2012; Döpfer et al. 1997; Holzhauer et al. 2008). Nevertheless, a few of these studies had controlled conditions or conditions which are not comparable with the present/real farm procedures, leaving some doubts if the results would be applicable in field conditions. For these several reason the current study was developed.

The objective of this study was to evaluate the effect (prevalence of DD before and after) of the intensive use of topical treatment with the different products (OTC 5g/5ml, HealMax, Hoof-Sol and as negative control saline solution) for DD on the prevalence of DD. Subsequently, the effect of the treatments was determined on the distribution of each M-stage on every of the 10 farms.

Materials and Methods

Farm enrolment criteria

Ten free-stall dairy farms in Alberta, Canada, with high DD prevalence (>15% active DD in the lactating cows) were selected. These farms were required to have accurate identification in the milking parlor and a milking parlor suitable for product application. Farms were visited with a 7-day interval for 8 weeks in January to April 2016. Farm visits started after the next routine claw trimming session (if the farm had the visit in approximately 3 weeks). This measure was taken to be sure that the treatments from the claw trimmer would not interfere with the applied treatments in this trial.

Figure 1 – Mirror on a spatula used to score the lesions situated on the hind feet

Scoring method

The 10 farms were divided over two teams, each team consisted of an observer and a topical treatment administrator. To score the lesions in the parlor, the observer had a head lamp and a

[2] Diamond Hoof Care Ltd. and Intracare BV., https://diamondhoofcare.com

mirror on a spatula (Figure 1). This tool provided a good observation angle, so that lesions situated between the heel and along the interdigital space could be seen more easily. The scorings were done by three observers over the whole trial. They received an inter-observer training to be sure that they used the same scoring criteria. All feet were cleaned before scoring, using the parlor hose. For every scored cow the identification number with the involved (left and/or right) foot and the M-stage were recorded. Each cow foot with a score ranging from M1-M4.1 was marked (with green MS Spray marker) to be treated. Each week the collected data were entered into Microsoft Excel. The researcher involved in scoring the feet was blinded throughout the study to the treatment each animal received.

Topical treatments

In this study, four different topical treatments were included: HealMax™, Hoof-Sol, negative control (saline with green coloring), and OTC spray (5g of OTC powder mixed with 5ml saline; 250mg/g of OTC; concentration=25%) as a positive control. Each treatment was prepared in the appropriate concentration and put into spray bottles of 500 ml (Figure 2). In the first week the treatment administrator picked the treatment for each cow using a randomization sheet (simple randomization). The affected feet were marked with a green mark by the scorer to identify those feet for treatment. The cow was the unit of randomization; therefore, cows with both hind feet affected had both feet treated with the same topical treatment. Each affected foot was sprayed seven times, in order to get approximately 5 ml of the treatment on the lesion.

Figure 2 – Equipment used by the topical treatment administrator: spray bottles (500 ml) and clipboard with the treatment sheet.

After every farm visit, all materials used to score and to treat were disinfected with bleach to avoid transmission between farms.

Follow up

All cows were examined weekly for 8 weeks. To keep track on the treatments from the week before, sheets (Figure 2) with the cow identification number and the treatment group were made per week. If a cow still had a lesion, the same treatment was applied again. The same happened when a cow healed but consequently developed a new lesion. A completely new lesion was randomly assigned to one of the treatment groups in the same way as in the first week of the trial.

Statistic analysis

The statistical unit used was the rear foot. Day 1 was defined as being the moment when the cows where scored and treated for the first time. The following week (on the same day of the week) was considered as being week 2. A foot was considered DD-positive (and therefore included in to the prevalence calculation) if a M1, M2, M3, M4, or M4.1 was detected on that foot. All M1-M4.1 lesions from day 1 were used. For each lesion the farm identification number, the week number, the cow ID, respective foot with the lesion and score were compiled into Microsoft Excel (Microsoft Corp., Redmond, WA, USA), and all statistical analyses were performed using Stata 13.1 (StataCorp, 2013, College Station, TX, USA). The data were checked for typing errors.

Statistical analysis was performed over the 10 farms. The DD prevalence was calculated in week 1 and 8. Secondly, the DD prevalence from the farms was tested with the McNemar test, to determine if the change in prevalence of DD after eight weeks of topical treatments was significant. To test a difference in the distribution of the M-stages after 8 weeks of topical treatment a Chi-Square test was used on contingency tables. A p-value <0.05 was considered statistically significant.

Results

In the first week, 4138 rear feet were scored and 1688 (41%) of those were treated. In week 8, 4191 hind feet were scored and 1785 (43%) treated. The average prevalence of DD was 44 and 46% at week 1 and 8, respectively. Figure 1 illustrates that the prevalence appeared to increase on 7 (farm 1, 3 and 7 decreased less than 1%) of the 10 farms; however, no significant changes in prevalence were observed; the lowest P-value was seen at farm 8 (OR=0.375; 95% CI: 0.064 - 1.562; P=0.13).

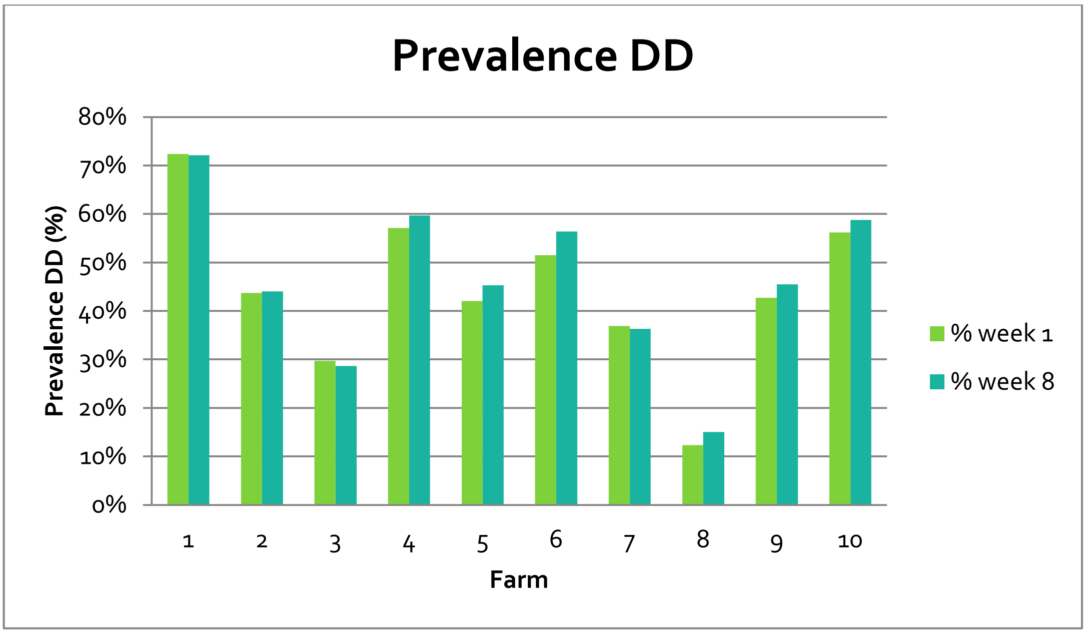

Figure 3 –DD prevalence on each farm in week 1 and week 8

Week 4 was added to visualize the development of the DD prevalence (Figure 2). No trend was visible. On seven farms the prevalence increased in week 4 and went down in week 8. The other three experienced a slow increase of prevalence.

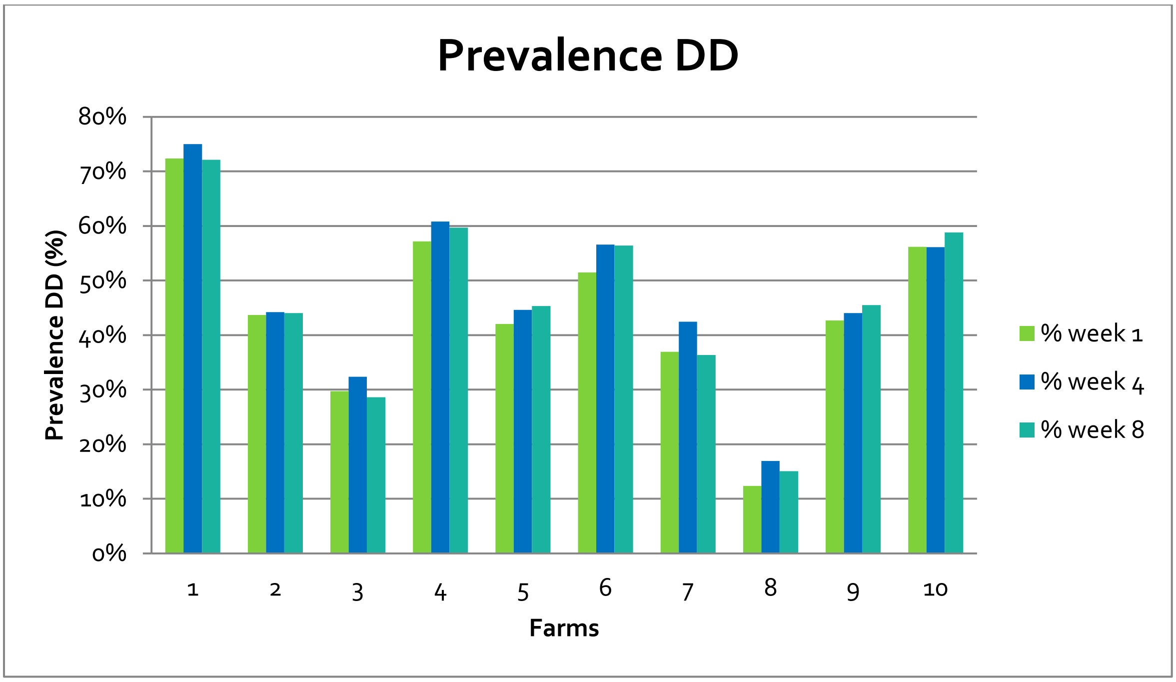

Figure 4 – DD prevalence on each farm in week 1, 4 and 8

Prevalence of M-stages per farm

Table 1 – Prevalence of the different M-stages in week 1 and week 8 for every farm

Farm	M0		M1		M2		M3		M4		M4.1	
	wk 1	wk 8	wk1	wk8	wk 1	wk 8	wk 1	wk 8	wk 1	wk 8	wk 1	wk8
1	22	27	5	9	18	6	4	2	41	50	4	3
2	57	55	9	14	4	1	6	3	21	24	2	1
3	69	70	0	0	2	0	5	4	19	23	3	2
4	42	40	0	2	8	8	9	2	36	39	4	8
5	55	55	7	7	9	3	1	1	21	31	4	3
6	46	42	5	5	4	3	10	6	30	40	2	1
7	61	63	0	0	2	0	4	4	26	31	4	1
8	88	84	2	5	0	0	1	2	9	9	0	0
9	54	55	0	0	5	2	4	1	25	38	6	4
10	43	41	1	0	7	2	7	3	32	49	1	4
Overall	53.7	53.2	2.9	4.2	5.9	2.5	5.1	2.8	26.0	33.4	3.0	2.7

Overall, the distribution of M-stages was different between the start and the end of the trial ($P<0.05$). The proportion of chronic (M4) lesions increased on every farm except Farm 8 where it remained the same, whereas the proportion of active lesions (M2 and M4.1) and the healing stage (M3) decreased (Table 1 and per farm in Figure 3-12). On farm 1, 2 and 4 the proportion of M1 (active lesions) increased instead of decreasing. For farm 4, no decrease was seen for M1, M2 and M4.1 lesions, and M0 and M3 decreased, indicating that there were still a lot of active lesions. On farm 7, the prevalence of M2 lesions decreased to 0%.

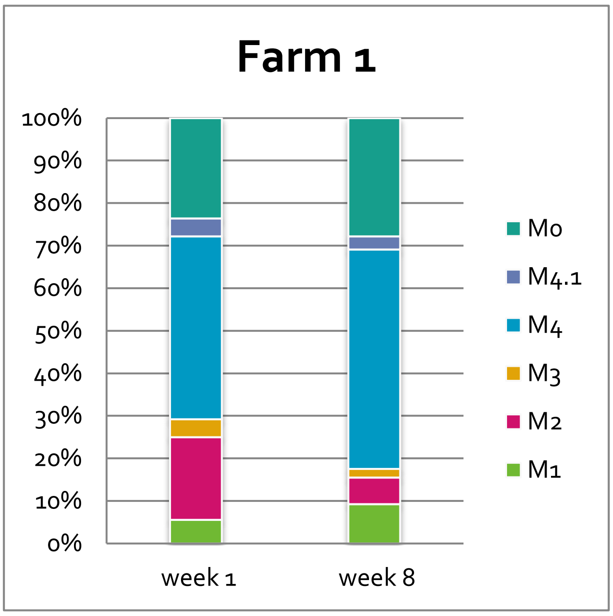

Figure 5 - Distribution of different M-stages on week 1 and 8 for farm 1

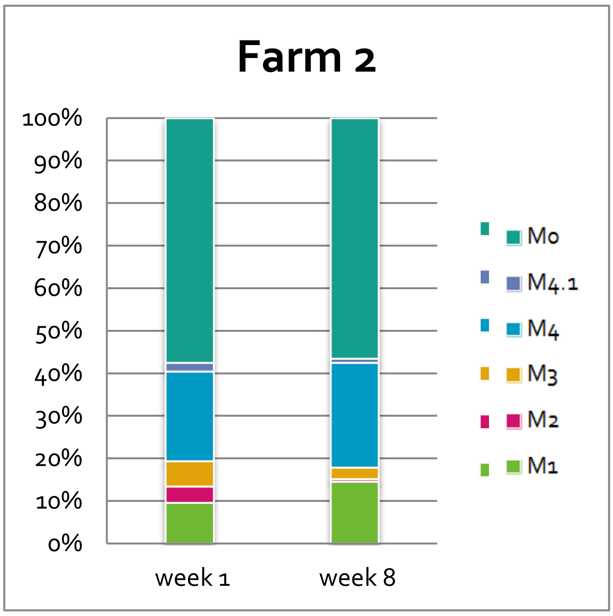

Figure 6 – Distribution of different M-stages on week 1 and 8 for farm 2

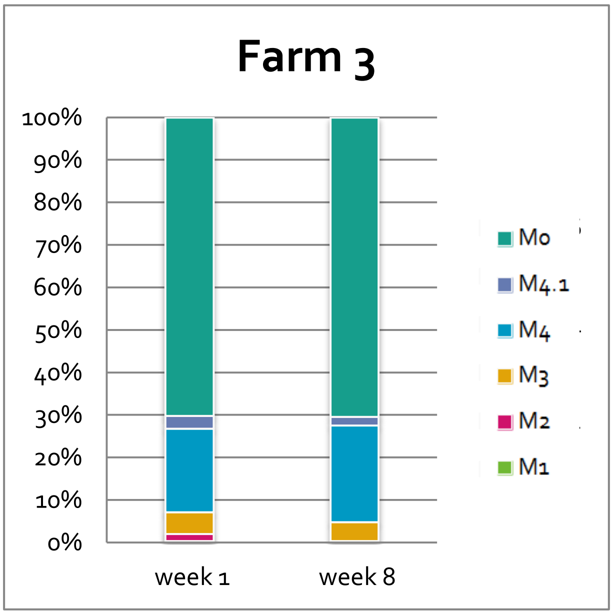

Figure 7 – Distribution of different M-stages on week 1 and 8 for farm 3

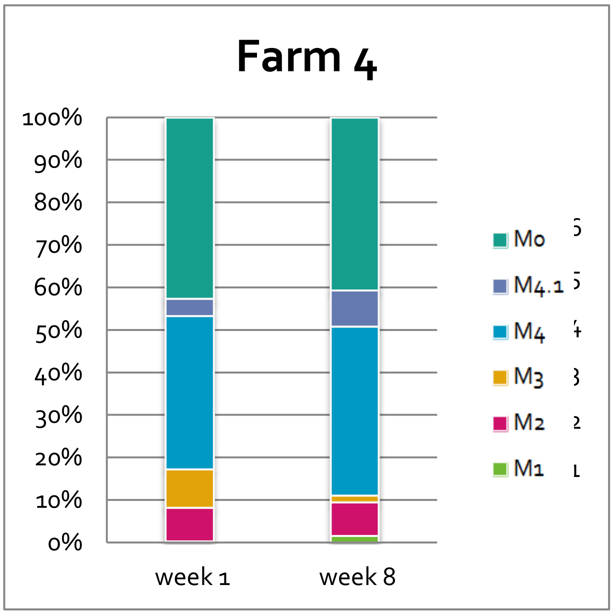

Figure 8 – Distribution of different M-stages on week 1 and 8 for farm 4

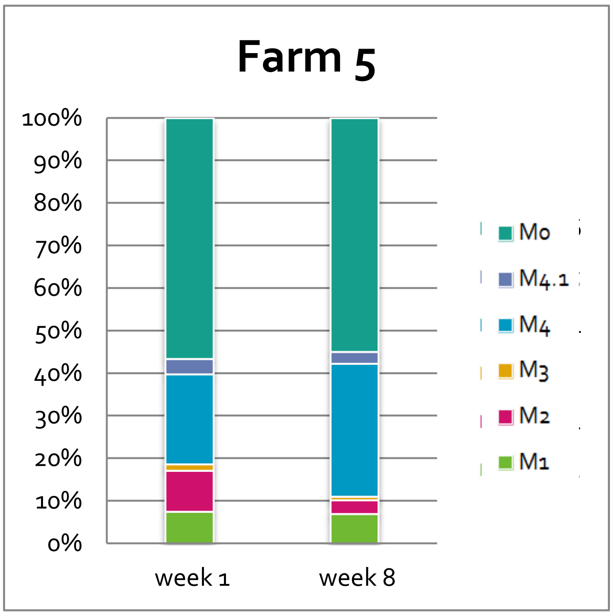

Figure 9 – Distribution of different M-stages on week 1 and 8 for farm 5

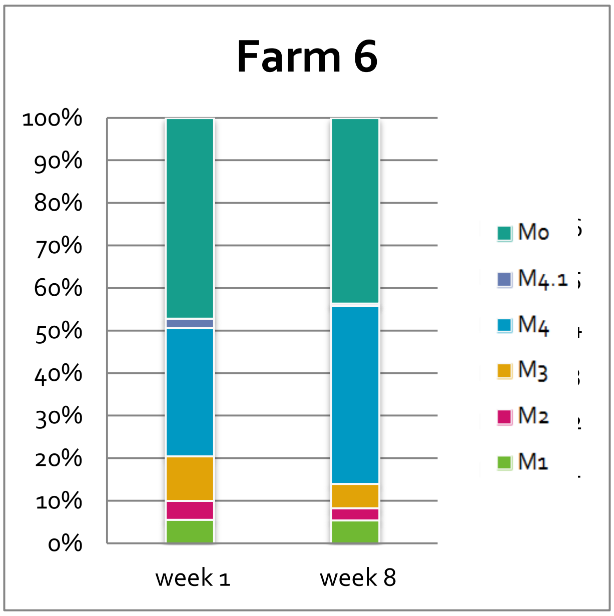

Figure 10 – Distribution of different M-stages on week 1 and 8 for farm 6

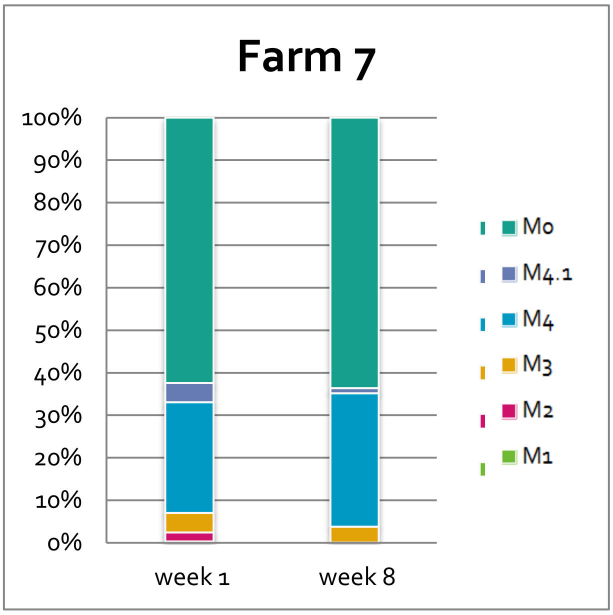

Figure 11 – Distribution of different M-stages on week 1 and 8 for farm 7

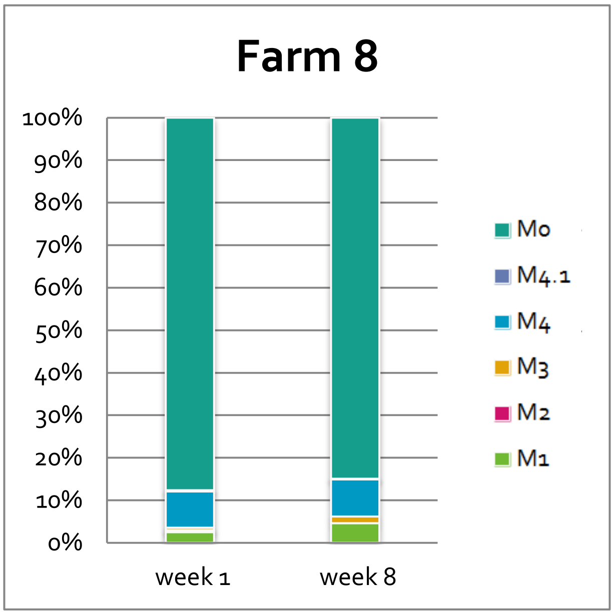

Figure 12 – Distribution of different M-stages on week 1 and 8 for farm 8

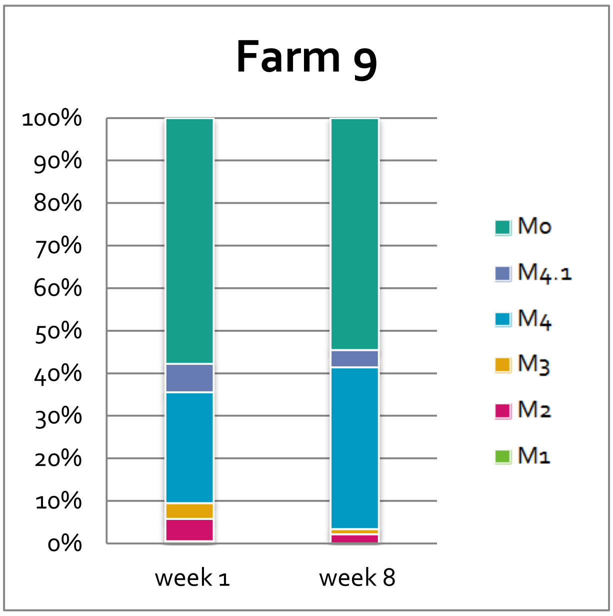

Figure 13 – Distribution of different M-stages on week 1 and 8 for farm 9

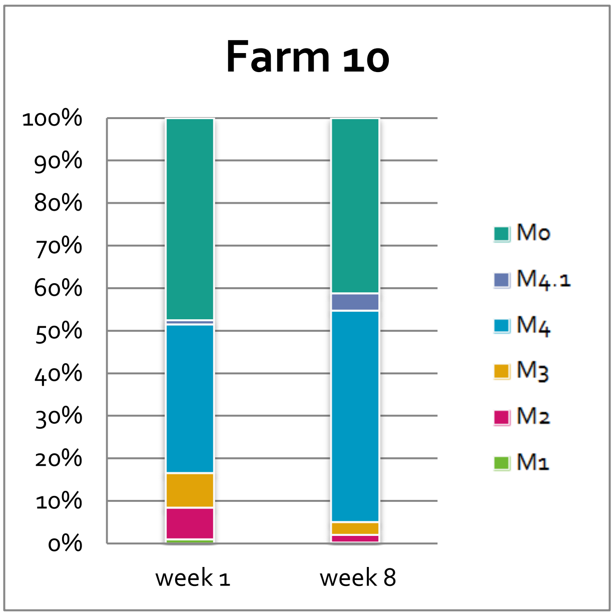

Figure 14 – Distribution of different M-stages on week 1 and 8 for farm 10

Discussion

The objective of this study was to determine if the prevalence of DD would change over 8 weeks on different dairy farms in Alberta (Canada) after weekly topical treatment of all DD lesions of the hind feet in the milking parlor. Recently, a study was conducted about the efficacy of scoring DD in the milking parlor, concluding that it is a reliable method (Relun *et al.*, 2012). As mentioned before, no scientific evidence is available about the effect of the current (on the market) in parlor topical treatments on the prevalence of DD in a herd. However, this trial demonstrates that after topical treatment on 10 dairy herds no change was seen in the overall DD prevalence (P=0.13). Comparing with other studies conducted to determine the evolution of the prevalence of DD in a herd; this trial did not have controlled conditions or conditions which were not comparable with the present farm procedures. Therefore, all cows continued in their normal routine. Also, for the overall prevalence calculation (Figure 1) the cows that were dried off, moved to the special needs pen, treated by the hoof trimmer, or culled during the trial were not excluded from the initial dataset. The same was applied to the heifers and recently calved cows, which came enter the lactating herd. Therefore, the results of our study provide a realistic indication of the prevalence in the whole adult herd. However, to test the statistical significance (with the paired McNemar test), it was needed to exclude all cows that were not in the trial the entire study period, from week 1 to week 8. Even then, no change in DD prevalence was seen on any of the farms. This was also confirmed by the study of Cramer et al. (2009) where a negative association between treating lesions topically on a routine basis and the prevalence of DD was found. This could be explained by the fact that the use of various spraying solutions have been recorded as efficacious, but the efficacy of spraying is dependent on product (Hernandez, Shearer and Elliott, 1999), frequency, duration, and concentration (Cramer *et al.*, 2009). Another explanation could be the reason that when new cows came into the herd, new DD lesions (active or not) were constantly added during the weeks. However, if we continued to treat every cow with a lesion for the longer period eventually the prevalence may decrease. To achieve this, the cows should have been treated for another eight weeks. This would give a higher assurance that cows that had been dried off during the treatment period and subsequently came back in the lactating herd were still treated. Those cows would be treated for eight weeks in total (considering the average dry off period of 60 days); another possibility could be treating topically the dry cows and the pregnant heifers also. However, putting those cows in a chute can be a factor of limitation at some farms (examples range from: no chute available, very labor intensive or a too high stress level for pre-calving cows). The necessity of eight weeks of treatment is based on three studies, Read and Walker (1998) and Berry et al. (2010 and 2012), which focused on recurrence of lesions after topical treatment with antibiotics and reported respectively recurrence after 7 weeks, after 4 weeks (32% recurrence) and after 37 days (8% of recurrence).

In this study, to calculate the prevalence of DD on farms it was assumed that only lesions scored as M0 were considered being healed and would be used to decrease the prevalence DD in the herd. However, in other studies where DD prevalence was also calculated, "healed" or was defined as grossly healed skin what could also be considered a M3 (Somers et al. 2003; Berry et al. 2010). This means that an improvement such as a transition from a M2 to a M3 was not considered. For this reason, a deeper look was given in to the evolution of the different M-stages after eight weeks of topical treatments. In general, the distribution of the different M-stages, followed an equal development as documented by Holzhauer et al. (2008a). The last mentioned reported transitions which were consistent and in sequence, for example, M1 to M2, M2 to M3 and M4 to M0. In the same study M3 to M0, M0 to M3 and M0 to M2 were found but considered unexpected, but also that more than 40% of the cows with a M0, M2 or M4 remained in the same stage (Holzhauer et al. 2008). This was also confirmed by Berry et al. (2012), as M1 and M2 were 27 times more likely to be M2 after a month than they were to be healed (M0). Another study found the transition of M2 to M4 and the transition of M3 to M4 (Capion *et al.*, 2012). Altogether gives an indication that the treated lesions (active stages) are more likely to develop into a chronic stage, although it depends of the history of the cows (type 1, 2 and 3 (Döpfer, van Boven and de Jong, 2004)). These chronic lesions are also prone to remain, as the treponemes will descend deep into the epidermis and dermis, and coil up into piles (Döpfer *et al.*, 1997). Therefore, chronic stages (M4) are considered being a reservoir of DD, turning this disease into long-term infection and disease (Döpfer *et al.*, 1997). In such manner a topical treatment will decrease the prevalence of active lesions but it will not cure the disease. However, this is preferable as cows will have less painful (less M2) lesions (Read and Walker, 1998) and it is also an important way to avoid that the chronic lesions turn into a new active lesions. In this trial, the proportion of chronic lesions (M4) increased and the proportion of active lesions (M2 and M4.1) decreased (Table 1 and per farm in Figure 3-12), confirming the results of the earlier mentioned studies. On farm 7 and 3 a good development was seen after the topical treatments as the M2 lesions decreased to 0 percent and on farm 3 it decreased to ≈0.33 percent. On farm 1, 2 and 4 the proportion of M1 (active lesions) increased instead of decreasing, this could be due to intra-observer differences (Relun *et al.*, 2011), but this was not statistically verified. For farm 4, no decrease was seen for M1, M2 and M4.1 lesions, and M0 and M3 decreased, indicating that there were still a lot of active lesions. This may lead to a thought that the topical treatments did not decrease the active lesions on this farm. This was the only farm out of the 10 with these results, indicating that an important introducing factor of DD could be present. A possible explanation for that could be the introduction of heifers with active DD lesions to the adult herd, a fact to which this farm was familiar with.

There are also some factors of interest that may have contributed to the outcome to this trial. Considering the topical treatments, four different topical treatments were used, and one of them was a negative control. For this reason essentially 75% of the animals with DD were treated with a 'positive' treatment. The remaining 25% could have given enough infection pressure so that the DD prevalence could not decrease. If one or two of the other treatments were also not very efficacious (no evidence for it), this would limit the overall treatment effect even more. Another point in the category of the topical treatments is the fact that it was established that spraying seven times would supply an amount of 5ml on the lesion. However, sometimes half of the solution dropped on the floor, as the product or was too thick to adhere to the skin (which was wet when treating) or the lesion was not easy to reach with the spray bottle. Shearer and Hermandez (2000) described this failure (not getting sufficient active substance on the lesion) as one of the major causes of reduced efficacy of topical treatment under field conditions. Additionally, some of the treatments which dropped on floor of the milking parlor were washed away which is a concern, particularly for the positive control treatment (OTC), as OTC is particularly strongly absorbed in all soils (Rabølle and Spliid, 2000). Also, Shearer & Hermandez (2000) commented that they received many comments from hoof trimmers, dairy farmers and practicing veterinarians suggesting that effectiveness of topical treatment of antibiotic solutions (including tetracycline or OTC) seemed to decrease after prolonged use.

For this trial only farms with >15% of DD were selected. This resulted in an automatic selection for farms with a high infection pressure. Therefore if weekly treatment was conducted on a farm with less DD, a higher impact on the prevalence would have occurred. Since the infection can be controlled more easily with low infection pressure. Furthermore, field conditions did not always correspond with the designed procedure plan. This was the case on farm 7 where the parlor hose was not working; as a result the DD scoring had to be done without cleaning the feet. This was in the middle of the trial so nothing could be done to repeat this visit and the visits before, as it would have affected the new initial prevalence at that farm. Another factor which could have contributed to the outcome was that on some farms the cows, after being treated in the parlor, walked through a water bath or manure footbath/alley. This will have had some effect on the period of time of lesion exposure to the treatment, which is also a key factor for success of the treatment (Shearer and Hernandez, 2000).

The method of on-farm prevalence determination in the milking parlor is good but not 100% sensitive, and will give an underestimation of the prevalence, as persistent subclinical infections cannot be detected with visual examination. In such a case, a lesion can be scored as Mo (healed) based on visual examination and after a while return to an active stage due to the persistent infection (Berry *et al.*, 2010). This fact can have contributed to a failure of DD control programs and until now no definitive solution is found for it. However, the study of Döpfer et al. (2012) focused on the dynamics of DD in populations of dairy cattle has developed a model to predict transition rates in a herd. This tool in future studies will help to improve to determine the prevalence rates on a farm. Nevertheless, more research needs to be done to improve the determination of the prevalence in a herd and to increase the knowledge about the efficacy of topical treatment with commercial treatments in the milking parlor.

Conclusions

Prevalence of DD did not change on 10 Alberta dairy farms after eight weeks of topical treatments (OTC 5g/5ml, HealMax^{TM}, Hoof-Sol and negative control). The distribution of M-stages did, however, change: the proportion of chronic lesions (M4) appeared to increase and the proportion of active lesions (M2 and M4.1). Eight weeks of topical treatments under field conditions did decrease the prevalence of active lesions in a herd, but did not decrease the overall DD prevalence on a farm.

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