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Clinico-pathological alterations in Holstein dairy cows with digital dermatitis with therapeutic intervention

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ABSTRACT

This study was performed on a commercial dairy herd of 127 *Holstein* cows with an outbreak of digital dermatitis (DD). Fifty-four DD-affected cows and 20 clinically healthy cows of the same days in milk (DIM) were selected. A detailed examination of each cow feet was recorded. Gross description and anatomic distribution of DD lesions were recorded. Lesions were biopsied and evaluated histopathologically. Lameness scoring was performed. Milk yield and BCS of the DD-affected and healthy cows were compared. Effectiveness of topical oxytetracycline treatment was assessed. The disease was contagious, a total of 54/127 cows were affected in which 70 lesions were identified. The consistent findings were lameness, reduced mobility and a localized painful dermatitis on hind claws. Most cows were clinically lame (62.9%). In the majority (92.6%) of cows only one hind leg was affected, and the left was more affected than the right one (51.9% vs 40.7%). The lesions most commonly (55.6%) were located on the back of hind legs on heel region. Majority of lesions were M2 (44.3%), large size (74.3%), painful (82.9), pink/red (62.9%), deep (42.9%), and circular (60%). Milk yield and BCS significantly decreased in DD-affected cows as compared to healthy cows. Lesions were responsive to topical oxytetracycline treatment. In conclusion, DD is a major cause of lameness in dairy farms and has a negative impact on milk yield and BCS. High incidence of lesions on the back of hind claws is characteristic for DD. Topical oxytetracycline is effective in DD lesions treatment.

1. INTRODUCTION

Digital dermatitis (DD) is a worldwide, painful, contagious inflammatory skin disease affecting feet primarily of intensively managed cattle (Wells et al., 1999) and induces varying degrees of pain, discomfort and lameness (Laven and Proven, 2000) resulting in serious economic losses and reduced cattle welfare (Bruijnis et al., 2012). The great economic losses due to DD are attributed to decreased milk yield, body weight loss, impaired reproductive performance, and increased culling rate in addition to high treatment and control costs (Cha et al., 2010). The exact etiology of the disease is still unclearly understood (Wilson-Welder et al., 2015). The disease in dairy herds appears to be multifactorial caused by complex interactions of a group of factors including bacteria, host, management and environment (Laven, 1999). Multiple bacterial species have been identified in DD lesion during disease progression; however, *Spirochetes* particularly *Treponema* have been suggested to be the probable causative pathogen (Krull et al., 2014).

The disease is highly contagious and may cause up to 90% morbidity in a cattle herd (Underwood et al., 2015). Lameness, lifting of the affected limb, walking on the tip of the toe are the most recorded clinical signs in DD affected cows (Laven, 1999). Furthermore, when feet are examined, a localized painful erosive or proliferative circumscribed dermatitis is usually identified on the back of rear foot (Pavlenko et al., 2011). Several scoring systems were designed to describe the DD lesions during the course of the disease (Krull et al., 2014). The "M score" system designed by Döpfer et al. (1997) that was adapted by Berry et al. (2012) is the commonly used system for description of DD lesions during disease progression. The gross diagnosis of the disease can be confirmed with the histopathological criteria of the DD lesions (Read and Walker, 1998). Ameliorating environmental hygiene and immediate treatment are crucial to control DD (Oliveira et al., 2017). The use of antimicrobials like tetracyclines as paste under light bandage or as topical spray is highly effective at treating cows with DD (Laven and Logue, 2006; Relun et al., 2012). This study was undertaken to characterize clinical picture and lameness scores in DD-affected cows, describe gross appearance, histopathological changes and anatomic distribution of DD lesions, investigate the impact of DD on milk production and body

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condition score (BCS), and evaluate the response of lesions to topical treatment with oxytetracycline.

2. MATERIAL AND METHODS

2.1.Study population and case definition
The study was conducted on a commercial dairy herd of 127 first lactation *Holstein* cows with an outbreak of DD (54 cows) (42.5%) belonging to Beni-Suef province, Egypt. Fifty-four DD-affected cows and 20 healthy cows of the same DIM were enrolled in this study. The herd was imported and introduced to the farm at December 2018 as late pregnant heifers. All heifers calved during February and March 2019. All cows were managed under the same conditions and nutritional regimen. Cows were fed a total mixed ration and milked three times daily. Cows were housed in free-stall barns under continually dirty wet conditions due to accumulated fecal waste. Historically, Footbaths and routine foot trimming were not performed in this farm. An outbreak of DD occurred in May, about 2-3 months post-calving.

2.2. Physical examination:
All cows were subjected to comprehensive physical examination (Radostits et al., 2000).
2.2.1. Gross description of DD lesions:
For identification and description of DD lesions, each foot was lifted, cleaned, and trimmed. A detailed examination of each leg was performed for evidence of DD lesions. Pain and size scores of identified DD lesions were evaluated as reported by Hernandez et al. (1999). In brief, existence and scoring of pain were based on the response of the cow to direct splashing of water on the lesions from a medium-pressure water hose. Pain scores ranged from 0 - 2 (0 = no pain; 1 = mild pain; 2 = severe pain). Size scores ranged between 0 and 2 (0 = no visible lesion; 1 = lesion ≤ 2.5 cm; 2 = lesion > 2.5 cm diameter). Additionally, anatomic location, number, surface characteristics, color, shape, and depth of DD lesions were recorded. Photographs of lesions were taken and saved.

2.2.2. Lameness scoring:
Locomotion was scored when cows were walking and standing on a 15 m alley with concrete flooring on their way to the milking parlor at morning milking. The existence and severity of lameness was evaluated based on the scale 1 to 5 as described by Sprecher et al. (1997).

2.2.3. Clinical scoring of DD lesions:
All identified DD lesions were assessed and clinically scored as previously described by Döpfer et al. (1997) and Berry et al. (2012).

2.3. Biopsies and histopathological evaluation:
Lesions were locally anesthetized with 2% lidocaine, and 6 mm-diameter, full-thickness punch biopsy specimens from lesions were obtained and placed in 10% formalin solution (Read et al., 1998). Each specimen was paraffin embedded, sectioned at 4µm thickness, and finally stained with H & E (Suvarna et al., 2017).

2.4. Effect of DD on milk production and BCS:
For evaluation of the effect of DD on milk production and BCS, Milk yield and BCS data of DD-affected cows were recorded at day of enrollment (day 0) and one week later (day 7). The obtained data were compared to those of clinically healthy cows (n=20) of the same days in milk. BCS was assessed based on the 1 to 5-point scale designed by Ferguson et al. (1994).

2.5. Efficacy of topical oxytetracycline hydrochloride for treatment of DD lesions:
The lesions were initially evaluated on day 0 just prior treatment, and the lesions characteristics were recorded. All DD lesions were treated regardless of their clinical stage. Treatments were carried out at milk parlor after the cows being milked. The affected foot was cleaned using strong pressure water hose then the lesions were topically treated using oxytetracycline hydrochloride solution (25mg/ml) as spry daily for 5 consecutive days then every other day for a 30-day period. The lesions were reevaluated, and the data recorded at day 7, 14, 21, and 30 posttreatment.

2.6. Statistical analysis:
The results of milk yield and BCS were analyzed using SPSS program (SPSS for windows Version 22). Independent two samples T-test was used to figure out the statistical difference between control and DD affected cows on 5 % significance level. Data expressed as (mean values ± SE).

3. RESULTS

3.1. Clinical examination findings:
The affected cows (n=54) showed variable degrees of discomfort and lameness, lifting the affected leg, standing and walking on toe tip, reduced mobility, prolonged lying down, weight loss, painful standing up and lying down, and localized painful dermatitis most commonly on the back of rear feet. Severely affected cows in which both feet were concurrently affected showed an inability in standing and walking.

3.2. Lameness scoring:
The majority of affected cows (n=34) (62.9%) were moderately to severely lame and scored 3-5. Twenty-four cows (44.4%) were lame to severely lame and scored 4-5, 10 cows (18.5%) were moderately lame and scored 3, 10 cows (18.5%) were mildly lame and scored 2 and 10 cows (18.5%) were non-lame and scored 1 (Table 1).

Table 1 Lameness score and anatomic distribution of lesions in 54 *Holstein* cows with DD in a dairy herd.

		Cow	
		Number	%
Number affected limb/cow	one hind limb/cow	50	92.6
	2 hind limbs/cow	4	7.4
Anatomic distribution of DD lesions	Heel	30	55.6
	coronet	8	14.8
	Heel and coronet	10	18.5
	Declaw	4	7.4
	Interdigital cleft	2	3.7
Lameness score	1	10	18.5
	2	10	18.5
	3	24	44.4
	4	10	18.5

3.3. Anatomic distribution of DD lesions:
All clinically affected cows showed DD lesions. The lesions were confined only to hind feet (Table 1). Only one of the hind limbs was affected in 92.6% of enrolled cows, but both hind feet were affected in 7.4%. Our observations

showed that the left hind limb was more affected than the right one (51.9% vs 40.7%). The lesions were characteristically located on the back of rear feet on or between heel bulbs, on both heel and coronet, at the coronet, around declaw and in interdigital cleft in 55.6%, 18.5%, 14.8%, 7.4% and 3.7% of affected cows respectively.

A total of 70 DD lesions were identified on 58 feet of the affected cows under study. The data revealed 79.3% of the affected feet had only one lesion and 20.7% had 2 lesions. In affected cows, 62.9% of lesions were found on plantar aspect of rear limbs on or between bulbs of heel, 25.7% at the coronet, 8.6% around declaws, and 2.86% in the interdigital cleft (table 3).

Table 2 Location, M stage, size and pain scores of DD lesions in 54 *Holstein* cows with DD in a dairy herd.

		Cow		Lesion	
		Number	%	Number	%
Limb	Right hind limb	22	40.7	30	42.9
	Left hind limb	28	51.9	32	45.7
	Both hind limbs	4	7.4	8	11.4
	Forelimbs	0	0	0	0
M score	M1	10	18.5	14	20
	M2	26	48.1	31	44.3
	M3	12	22.2	14	20
	M4	2	3.7	3	4.3
	M4.1	4	7.4	8	11.4
Size score	1	14	25.9	18	25.7
	2	40	74.1	52	74.3
Pain score	0	10	18.5	12	17.1
	1	10	18.5	14	20
	2	34	63.0	44	62.9

Table 3 Gross appearance of 70 DD lesions in 54 *Holstein* cows in a dairy herd.

Lesion characteristic		Number	%
Lesion shape	Circular	42	60
	Oval	16	22.9
	Linear	12	17.1
Lesion stage	Early	12	17.1
	Ulcerative	36	51.4
	Healing	14	20
	Proliferative	8	11.4
Lesion surface	Deep	30	42.9
	Raised	25	35.7
	Flat	15	21.4
Lesion color	Pink/red	44	62.9
	Creamy/yellow	12	17.1
	grey/brown	14	20
Heel/coronet/declaw/ Id	1- Heel:	44	62.9
	Between heel bulbs	16	22.9
	Lateral heel bulb	16	22.9
	Medial heel bulb	12	17.1
	2- coronet	18	25.7
	3- Around declaws	6	8.6
	4- Interdigital cleft	2	2.9

3.4. Gross appearance and clinical scoring of DD lesions:

Our observations showed that the clinical appearance of DD lesions varies with disease progression (Fig.1). Initially, the lesion begins as a small (< 2cm) reddish round to oval or elliptical less painful area (M1 stage) on the caudal aspect of hind legs most commonly on or between heel bulbs. The inflammation, redness, pain, and swelling increases as the disease progress. With time the affected area becomes severely inflamed, painful, reddened, and progressively swollen until the lesion burst at the weakest point leaving moist, bright red or red grey color ulcerative surface (>2cm) that prone to bleed when rubbed (M2 stage). The ulcerative lesions undergo healing (M3 stage) and are covered with firm, scab-like material and characterized by no or minute pain on palpation. Later on, unhealed lesions show progressive surface proliferation and with time appear less painful pale, gray, or brown wart-like (M4 stage). Some cases present reddish or hemorrhagic foci in the proliferative lesions (M4.1 stage).

Dynamic macroscopic changes were present among those previously mentioned stages (Fig.1). At the time of enrollment (first observation or examination), 20% of identified DD lesions were at M1 stage, 51.43% at M2 stage, 20% at M3 stage, 2.86% at M4 stage, and 5.71% at M4.1% stage.

Regarding pain response (table 2), The M2 stage was the most painful sage over the disease course. In the majority of cows (62.9%), DD lesions were moderate to severely painful. Also, the findings showed that most of the lesions (74.3%) were large in size and scored 2, while 25.7% of the lesions were small size and scored 1. Majority (60%) of lesions appeared circular, 22.9% were oval and 17.1% were linear. Most of the lesions showed pink/red (62.9%) discoloration and deep surface (42.9%).

3.5. Histopathological findings:

The biopsy specimens of DD lesions revealed loss of all epidermal epithelial cells. The dermis was heavily infiltrated with neutrophils, lymphocytes, eosinophils, and plasma cells. The subcutaneous blood vessels were highly congested, and hemorrhages and edema occurred in some areas. Other biopsy specimens showed massive proliferative reaction of dermal fibrous connective tissue accompanied by massive mononuclear cells and neutrophils infiltrations (Fig. 2).

3.6. Milk production and BCS findings:

The data revealed that DD had a significant impact on milk yield and BCS. The results showed that cows affected with DD had a significantly lower milk yield and BCS than healthy cows (table 4).

Table 4 Milk yield and BCS of DD- affected cows and healthy control cows (mean ± SE).

	Control cows	Cows with dd	P-value
Milk yield	26.35 ± 0.20	20.75 ± 0.26	<0.001**
BCS	2.9 ± 0.04	2.73 ± 0.06	0.024*

*Healthy and diseased cattle significantly different at p<0.05. **Significant at p<0.001.

3.7. Response of DD lesions to topical treatment with oxytetracycline HCl:

The DD lesions showed high response to topical treatments with oxytetracycline as spray. Good improvement was observed by day 7 and 14 post-treatment. A complete therapeutic response was observed by day 21 post-treatment. Response was characterized by complete transformation from raw, red, wet, painful surfaces to dark brown, dry, non-painful keratinous surfaces (Fig.1 M3). These changes were observed in 62.23 % of DD lesions by day 14 post-treatment. By day 21 post-treatment, diminution in lesions size and partial restoration of hair growth were observed.

Fig. 1 Skin lesions in the feet of cattle with different M grades of clinical digital dermatitis. M0: Normal healthy skin, M1: small ulcerative lesion <2cm in diameter, M2: small ulcerative lesion >2cm in diameter with a "strawberry" like appearance, M3: lesions treated with topical oxytetracycline antibiotic for seven days prior to examination (the ulcerative lesions were covered with firm, scab-like material), M4: chronic lesions with occasional epidermal proliferation, M4.1: M4 lesions with small active foci.

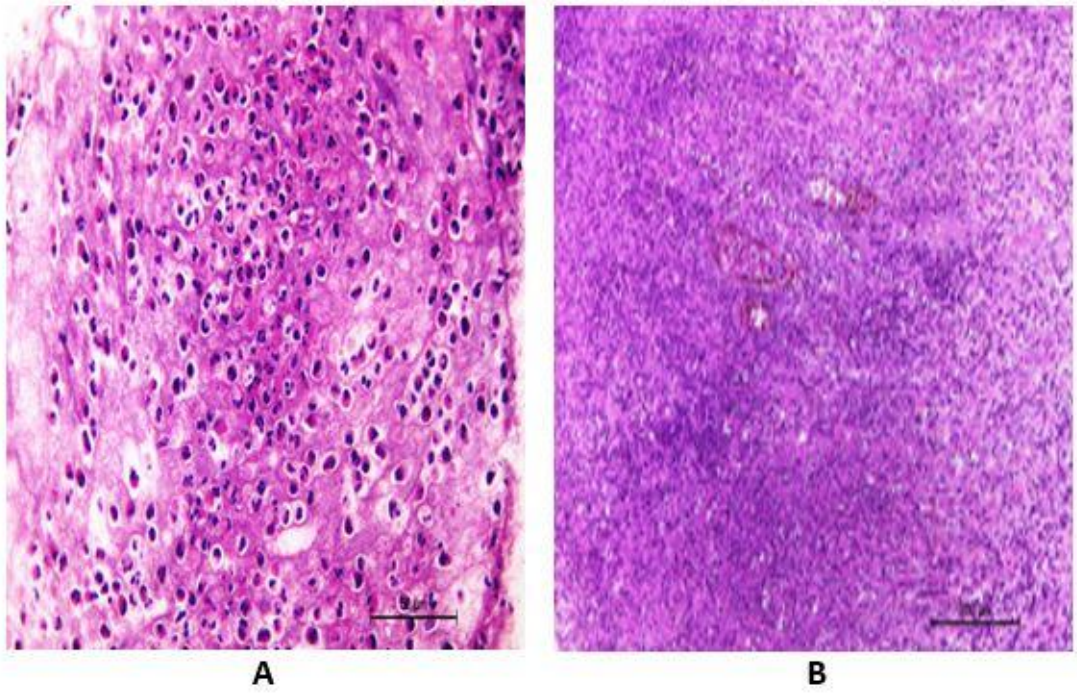

Fig. 2 (A) A biopsy specimen of hind foot skin of a dairy cow with DD showing loss of all epidermal epithelial cells and heavy infiltration of the dermis was with neutrophils, lymphocytes, eosinophils and plasma cells (H&E X400). (B): A biopsy specimen of hind foot skin of a dairy cow with DD showing massive proliferative reaction of dermal fibrous connective tissue associated with massive mononuclear cells and neutrophils infiltrations (H&E X100).

4. DISCUSSION

In this study, the clinical examination, lameness score, milk production and BCS findings in conjunction with gross appearance, histopathological features and anatomic distribution of DD lesions in Holstein dairy cows with DD were recorded. The clinical examination findings revealed occurrence of an outbreak of DD in a commercial newly imported dairy herd of 127 first lactation Holstein cows approximately 2-3 months post-calving. The disease occurs as outbreaks of lameness in newly infected dairy herds (Manske et al., 2002). Outbreaks usually occur following the introduction of newly imported or purchased animals from infected herds (Kofler, 1997). The disease is usually brought into dairy herds by infected heifers newly imported or introduced from other farms (Wells et al., 1999). Researchers established a close association between prevalence of DD and introduction of infected cattle (Wells et al., 1999).

The disease is highly contagious and may cause up to 90% morbidity in a cattle herd (Underwood et al., 2015). In the present study, the morbidity rate was moderate (42.5%). A total of 54/127 first lactation cows were diagnosed with DD lesions. The increased morbidity of DD in this herd could be attributed to the high exposure of the cows in this farm to several risk factors associated with DD among which breed, parity, stage of lactation, the introduction of infected newly introduced heifers and poor managemental measures. In respect to breed, Holstein cows are at greater susceptibility to DD than other cow breeds (El-shafaey et al., 2019). The disease is more common in the first lactation than older cows (Laven and Logue, 2007). The risk for developing DD is reducing with increasing parity. This could be attributed to increased local immunity in cows with age (Read et al., 1998).

Concerning the clinical picture, DD-affected cows in the present work showed variable degrees of discomfort and lameness, lifting of the affected leg, standing and walking on toe tip, reduced mobility, weight loss, (Read and Walker, 1998), increased number of lying times with prolonged lying bouts (Walker et al., 2008), difficulties in getting up and lying down, and localized painful dermatitis most commonly on the back of rear feet (Blowey and Sharp, 1988). Furthermore, severely affected cows in which both feet were affected showed an inability in standing up and walking. Affected cows were not systemically ill.

In the present study, the locomotion scoring revealed that the majority of DD-affected cows (62.96%) were moderate to severely lame and scored 3 to 5. These findings are in agreement with Berry et al., (2004) who stated that DD is one of the major causes of lameness in cattle worldwide.

Lameness, reduced mobility, prolonged lying, difficulties in lying down and standing up were attributable to the pain associated with DD lesions (Walker et al., 2008). Pain, reduced mobility and prolonged lying on the floor result in reduction in the total feeding time and subsequently weight loss and reduced milk production (Palmer et al., 2012). A significant reduction in milk yield and BCS were recorded in DD-affected cows than healthy cows in our study. These findings revealed that DD and the associated lameness have negative impact on milk yield (Pavlenko et al., 2011) and BCS (Singh et al., 2018).

Regarding the anatomic distribution of DD lesions, the findings showed that all lesions were confined to the rear feet most commonly on the back of rear feet on or between heel bulbs. No lesions were identified above the level of the declaws (Read et al., 1998). The characteristic predilection of DD lesions on the caudal aspect of hind limbs is a hallmark to DD (Read et al., 1998). The rear feet of the cows in this herd were continually submerged in manure slurry and remained wet than were the forelimbs especially during feeding. Other explanations have been speculated among which the shorter heels of hind feet and the more exposure and injury of the plantar aspect of hind legs to stall mats when the cow is laying down in normal position (Read et al., 1998).

In the current study, both hind feet were concurrently involved in 7.41% of cows, while in most cows (92.59%) only one rear foot was involved. The left hind foot was more affected than the right one (El-shafaey et al., 2019).

With regards to gross appearance, the early gross lesions of DD appeared as small (1cm) flat to raised erythematous masses with papilliform projections. Histologically, there was loss of stratum corneum/or granulosum, \ epidermal hyperplasia, and reactive inflammation (Walker et al., 1995; Read et al., 1998). Over time, lesions were larger, developed frond-like projections and had a tendency to ulceration or physical trauma. Lesions were usually prone to bleed when touched (Holzhauer et al., 2008).

The topical application of oxytetracycline was very effective in the treatment of DD-affected cows. Remarkable improvement was observed by day 7 and 14 post-treatment. Similar findings were obtained by Read et al. (1998), Laven and Logue (2006) and Relun et al. (2012).

5. CONCULSION

Digital dermatitis can occur in the form of outbreaks and is worsened after introduction of newly purchased heifers. The disease is a major cause of lameness in dairy herds. Lameness, discomfort, lifting of the affected leg, decreased mobility, prolonged lying, and weight loss were the consistent signs. High incidence of lesions on back of hind legs most commonly on the heel region is a hallmark of DD. DD has a negative impact on dairy cow's health, milk yield, and BCS. Poor housing circumstances are most probably high predisposing for DD. Lesions of DD are highly responsive to topical oxytetracycline treatment as spry.

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6. REFERENCES

1. Berry, S.L., Read, D.H., Famula, T.R., Mongini, A. and Döpfer, D., 2012. Long-term observations on the dynamics of bovine digital dermatitis lesions on a California dairy after topical treatment with lincomycin HCl. Vet J 193:654–658.
2. Berry, S.L., Walker, R.L., Read, D.H., Hird, D.W. and Ertze, R.A., 2004. The current state of knowledge on (papillomatous) digital dermatitis in dairy cattle: with particular reference to control. Pages 130–137 in Proc. 13th Intern. Symp. 5th Conf. Lameness Ruminants, Maribor, Slovenia.
3. Blowey, R.W. and Sharp, M.W., 1988. Digital dermatitis in dairy cattle. Vet. Rec. 122:505–508.

4. Bruijnis, M., Beerda, B., Hogeveen, H. and Stassen, E., 2012. Assessing the welfare impact of foot disorders in dairy cattle by a modeling approach. Animal 6: 962–970.
5. Cha, E., Hertl, J.A., Bar, D. and Grohn, Y.T., 2010. The cost of different types of lameness in dairy cows calculated by dynamic programming. Prev. Vet. Med. 97: 1–8.
6. Döpfer, D., Koopmans, A., Meijer, F.A., Szakáll, I., Schukken, Y.H., Klee, W., Bosma, R.B., Cornelisse, J.L., van Asten, A.J. and ter Huurne, A.A., 1997. Histological and bacteriological evaluation of digital dermatitis in cattle, with special reference to spirochaetes and Campylobacter faecalis. Vet. Rec. 140:620–623.
7. El-shafaey, E., Sayed-ahmed, M., Shoeib, S., El-khodery, S., 2019. Occurrence and risk factors of digital dermatitis in Egyptian dairies. Slov vet res., 56 (suppl 22): 465–74.
8. Ferguson, J.D., Galligan, D.T. and Thomsen, N., 1994. Principal descriptors of body condition score in Holstein cows. Journal of Dairy Science. 77: 2695-2703.
9. Hernandez, J., Shearer, J.K. and Elliott, J.B., 1999. Comparison of topical application of oxytetracycline and four nonantibiotic solutions for treatment of papillomatous digital dermatitis in dairy cows. J. Am. Vet. Med. Assoc. 214: 688–690.
10. Holzhauer, M., Bartels, C.J., Dopfer, D. and van Schaik, G., 2008. Clinical course of digital dermatitis lesions in an endemically infected herd without preventive herd strategies. Vet. J. 177: 222–230.
11. Kofler, J., 1997. Outbreak of digital dermatitis ('Mortellaro disease') in a dairy herd in Lower-Austria. Wiener Tieraerztliche Monatsschrift 84: 254–265.
12. Krull, A.C., Shearer, J.K., Gorden, P.J., Cooper, V.L., Phillips, G.J. and Plummer, P.J., 2014. Deep sequencing analysis reveals temporal microbiota changes associated with development of bovine digital dermatitis. Infect. Immun. 82: 3359–3373.
13. Laven, R. and Proven, M., 2000. Use of an antibiotic footbath in the treatment of bovine digital dermatitis. Vet. Rec. 147: 503–506.
14. Laven, R.A. and Logue, D.N., 2006. Treatment strategies for digital dermatitis for the UK. Vet. J. 171: 79–88.
15. Laven, R.A. and Logue, D.N., 2007. The effect of pre-calving environment on the development of digital dermatitis in first lactation heifers. The Veterinary Journal 174: 310–315.
16. Manske, T., Hultgren, J. and Bergsten, C., 2002. Topical treatment of digital dermatitis associated with severe heel-horn erosion in a Swedish dairy herd. Prev. Vet. Med. 53: 215–231.
17. Palmer, M.A., Law, R. and O'Connell, N.E., 2012. Relationships between lameness and feeding behavior in cubicle-housed Holstein–Friesian dairy cows. Appl. Anim. Behav. Sci. 140: 121–127.
18. Pavlenko, A., Bergsten, C., Ekesbo, I., Kaart, T., Aland, A. and Lidfors, L., 2011. Influence of digital dermatitis and sole ulcer on dairy cow behaviour and milk production. Animal, 5:8: 1259–1269.
19. Radostits, O.M., Mayhew, I.G. and Houston, D.M., 2000. Veterinary clinical examination and diagnosis. Harcourt Publisher Limited, W. R. Saunders, London.
20. Read, D.H., Nordhausen, R.W. and Walker, R.L., 1998. Pathogenesis of experimental papillomatous digital dermatitis (footwarts) in cattle: Bacterial morphotypes associated with early lesion development. In Proc. 10th Int. Symp. Lameness in Ruminants, Lucerne, Switzerland. Pp.271
21. Read, D.H. and Walker, R.L., 1998. Papillomatous digital dermatitis (footwarts) in California dairy cattle: clinical and gross pathologic findings. J Vet. Diagn. Invest. 10:67–76.
22. Relun, A., Lehebel, A., Bareille, N. and Guatteo, R., 2012. Effectiveness of different regimens of a collective topical treatment using a solution of copper and zinc chelates in the cure of digital dermatitis in dairy farms under field conditions. J Dairy Sci. 95:3722–35.
23. Singh, A., Singh, S., Gupta, K. and Bansal, B., 2018. Relationship of lameness to body condition score, udder health and milk quality in crossbred dairy cattle. Veterinarski Arhiv 88 (2): 179-190.
24. Sprecher, D.J., Hostetler, D.E. and Kaneene, J.B., 1997. A lameness scoring system that uses posture and gait to predict dairy cattle reproductive performance. Theriogenology 47(6): p. 1179-87.
25. Suvarna, K.S., Layton, C. and Bancroft, J.D., 2017. Bancroft's theory and practice of histological techniques E-Book. Elsevier Health Sciences.
26. Underwood, J., Blauwiekel, R., Delano, L., Gillesby, R., Mischler, A. and Schoell, A., 2015. Laboratory Animal Medicine (Third Edition). Chapter 15. In: Biology and Diseases of Ruminants (Sheep, Goats, and Cattle). Pp. 623-694.
27. Walker, R.L., Read, D.H., Loretz, K.J. and Nordhausen, R.W., 1995. Spirochetes isolated from dairy cattle with papillomatous digital dermatitis and interdigital dermatitis. Vet. Microbiol. 47: 343–355.
28. Walker, S., Smith, R., Jones, D., Routly, J. and Dobson, H., 2008. Chronic stress, hormone profiles and estrus intensity in dairy cattle. Horm. Behav. 53, 493–501.
29. Wells, S.J., Garber, L.P. and Wagner, B.A., 1999. Papillomatous digital dermatitis and associated risk factors in US dairy herds. Prev. Vet. Med. 38:11–24.
30. Wilson-Welder, J.H., Alt, D.P. and Nally, J.E., 2015. Digital Dermatitis in Cattle: Current Bacterial and Immunological Findings. Animals 5: 1114-1135.