

## **Research Article**

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# Histopathological changes in uncomplicated sole ulcers in dairy cattle

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**Abstract:** This study aimed to investigate the histopathological changes that occurred in the wound of the sole ulcer located on the hooves of dairy cattle. A total of 12 Holstein–Frisian dairy cows suffering from sole ulcer were studied. Sole ulcers in the lateral hoof of the left or right foot of hind legs containing the corium and a part of the epidermis were collected and examined histopathologically. There were subacute inflammatory changes in the superficial dermal vessels. A slight perivascular lymphocyte infiltration and congestion and hyalinization of arterial walls were observed in the corium. Thickening was observed in the endothelium of dermal vessels, and revascularization areas were also noticed. Although epithelial proliferation (acanthosis), degeneration, and necrosis were found, the more obvious changes observed were eosinophilia and mineralization of the connective tissue. In addition, congestion and edema were also seen in the connective tissue.

Key words: Sole ulcer, cattle, histopathology

## Introduction

A sole ulcer is an area of damaged sole horn that has lost most of the hoof horn tissue except the corium, which is the alive horn-producing tissue (1-4).

Sole ulcers are the most common foot disease and most typically they occur in the lateral hoof of the hind foot. Sole ulcers commonly affect one or both lateral hind claws, predominantly in heavy, high-yielding dairy cattle kept under confined conditions (5).

The typical location of the lesion is around the axial border of the heel–sole junction of the lateral claws of the hind feet. Sole ulcers on the front feet

are rare; however, when they occur, they are usually located on the medial hooves. Bilateral lesions of the hind feet are also commonly seen. In some cases, loss of sole is not evident, but there is hemorrhage beneath the sole or the sole is yellow and soft; the ulcer becomes apparent when the undermined sole is trimmed away. Damage to the dermis is associated with a circumscribed zone of localized hemorrhage and necrosis (6). The ulcer sometimes appears as a hemorrhage with a softened and yellowish horn and progressive necrotic tissue together with infection. Granulation tissue may protrude from the ulcer area. Severe lameness is not usually seen until granulation tissue develops from the exposed corium and

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protrudes from the defect present in the sole (3,4,6).

Although the literature contains broad and very detailed knowledge on angiographic evaluation of vascular changes, no detailed information about connective tissue and vascular pathological changes in the sole lesions located in the hooves of cattle is available. The aim of the present study was to investigate the histopathological changes in the wound of the uncomplicated sole ulcer on affected dairy cattle claws.

### Materials and methods

In this study, the hind claws of 12 Holstein–Frisian dairy cows suffering from sole ulcer were studied. Sole ulcers on the lateral claw of the left or right foot of hind legs consisting of the corium layer and a part of the epidermis were collected (Figure 1a) and examined histopathologically. The collected samples were fixed in 10% buffered formalin and embedded in paraffin. Sections 3–5  $\mu m$  thick were cut from the paraffin-embedded tissue and stained with hematoxylin and eosin (H&E). Beside this, some selected samples were additionally stained using van Gieson's method for collagen fibers (7).

#### Results

Changes related with subacute inflammation of the superficial dermal vessels were observed. The pathologic changes are presented in the Table.

A slight perivascular lymphocyte infiltration and congestion and hyalinization of arterial walls were seen in the corium (Figure 1b). There was an increase in the thickness of the epithelial layer of dermal vessels, and revascularization areas were also noticed.

Although an epithelial proliferation was observed, rete ridge formation was characterized with the extensions of the epithelium into connective tissue and the degeneration and necrosis of the basal and squamous epithelium. The other changes observed were accumulation of eosinophils and mineralization of connective tissue (Figure 1c). Congestion and edema were observed in the connective tissue of the corium layer. Hyaline degeneration and mineralization, especially around the ulcer sites, were noticed in collagen bundles (Figure 1d). Dyskeratotic changes were seen in the stratum basale and spinosum. Ruptures were seen in the epidermisdermis border in some areas close to calcified areas.

Table. The histopathological changes in uncomplicated sole ulcer in dairy cattle. Lesions are scored as +: weak, ++: moderate, and +++: marked, according to the severity of the changes.

Case no.	Inflammation	Epithelial proliferation	Mineralization of connective tissue
1	++	++	++
2	++	+++	+
3	+	++	+
4	+++	++	+++
5	+	++	+
6	++	+++	++
7	++	++	+
8	+++	+++	+++
9	++	++	+
10	+	++	+
11	++	+++	+
12	+++	++	+++

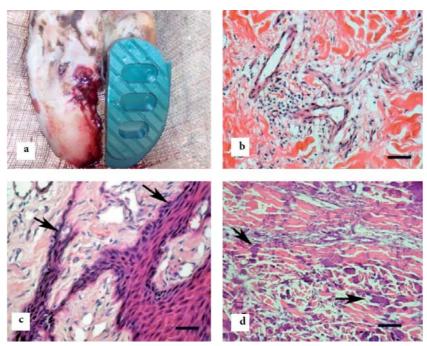


Figure 1. Images from affected cattle: a) sole ulcer; b) a slight perivascular infiltration by mononuclear inflammatory cell, bar = 40  $\mu m$ , H&E; c) rete ridge formations characterized with extension of the epithelium into the connective tissue where innermost part of the epidermal lamella is only 1 to 2 cell layers wide (arrows), bar = 35  $\mu m$ , H&E; (d) degeneration, necrosis, and mineralization (arrows) in collagen bundles near the ulcer site, bar = 80  $\mu m$ , H&E.

#### Discussion

The cause of foot diseases should be considered as a combination of predisposing factors leading to vascular reactivity and inhibition of normal keratin synthesis (1–4,8–10). In this study, uncomplicated sole ulcers were observed as a necrotic area at the sole–heel junction in hind lateral hooves. There was granulation tissue development and protrusion from the ulcer area. All animals showed signs of severe lameness and were kept under unhygienic conditions. Samples collected from the studied animals contained the corium and some part of the epidermis in the lateral claw of the left or right foot of hind legs.

The pathogenesis of hoof lesions in cattle has long been assumed to be associated with laminitis and, in analogy to the horse, to be due to a disturbance in the microcirculation of the corium of the claw with subsequent degenerative (and inflammatory) changes at the dermal–epidermal junction (8,11,12). Therefore, we compared our findings with histopathological changes of bovine laminitis (8,9,11,12) and uncomplicated Rusterholz ulcer (13) in the literature. In acute laminitis, hyperemia, edema, and hemorrhages were seen in the corium. Severe hemorrhages occurred in the solar corium at the toe and opposite to the plantar/palmar edge of the distal phalanx. Lymphocytes predominated (8,12), but focal infiltrations of neutrophils were also observed. Thrombi were found in both small and large arteries and veins (11). Thickening of the tunica adventitia, medial fibrosis and loss of medial smooth muscular cell continuity, intimal fibrosis, and ruptures in the internal elastic membrane are pathological changes related to chronic laminitis in dairy cattle (11). In the present study, the most important changes related to vessels in the sole ulcerate claw were lymphocyte infiltration and congestion and hyalinization of arterial walls. There was a thickening in the endothelium of dermal vessels, and, furthermore, revascularization areas were noticed. These findings

were different from findings of chronic laminitis of both horse and cattle reported by Ossent and Lischer (9) and Boosman et al. (11). In our findings, there was no obstruction of the vascular lumen by thrombosis, thickening or fibrosis of vessel walls, or ruptures in the internal elastic membrane. Recanalization of thrombi was not seen, but many newly formed capillaries and neocapillary formations were present. Boosman et al. (11) reported the recanalization of thrombi and neocapillarization in chronic laminitis. The present study shows that there is no proof of obstruction of the vascular lumen by thrombosis as demonstrated by Ossent and Lischer (9), Boosman et al. (11), and Azarabad et al. (13). The histological findings were in agreement with those of Azarabad et al. (13) and Singh et al. (14), who demonstrated infiltration and congestion.

On the other hand, vasoconstriction and ischemia of the peripheral dermal microcirculation of the bovine digit have been incriminated in the pathophysiology of bovine foot diseases. However, the characteristics and degree of vascular involvement are not fully understood (2). In the current study, histopathological evaluation showed that the most obvious connective tissue changes were congestion, edema, accumulation of eosinophils, and mineralization of connective tissue in dermis. Congestion and edema may be due to impairment of the blood supply of the corium. Vessel wall paralysis and vasodilatation cause the blood to stagnate and they subsequently begin to leak. Transudation leads to development of edema. The sequence of events is erythema, edema, hemorrhages, thrombi, and finally necrosis.

The understanding of histopathological aspects of sole ulcer in dairy cattle is limited (13); therefore, we presented here the histopathological changes occurring in bovine laminitis and uncomplicated Rusterholz ulcer. The histopathological evaluation of this study shows that both vascular and connective tissue changes in soles with ulcer, uncomplicated Rusterholz ulcer, and laminitis were basically the same.

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