Chapter

Macroscopic, Radiographic and Histopathologic Changes of Claws with Laminitis and Laminitis-Related Disorders in Zero-Grazed Dairy Cows

James Nguhiu-Mwangi and Peter M.F. Mbithi

Abstract

Laminitis and laminitis-related claw disorders are prevalent in zero-grazed dairy cows. Confinement and limited movement influences claw size and shape. Abnormal claw size and shape causes imbalanced body weight distribution on the claws. Claw horn growth and wear is impaired, further aggravating laminitis disorders. The objective of this study was to determine: macroscopic disorders on the claws, as well as radiographic features and histopathologic changes on the claws with laminitis/laminitis-related disorders. A total of 159 dairy cow forelimb and hind limb feet (318 claws) were collected from an abattoir and a slaughter slab around Nairobi, Kenya. The claws were examined for macroscopic abnormalities, dorso-palmar or dorso-plantar radiography done, sagittal claw sections done, corium gross changes observed and corium tissues harvested for histopathology. Macroscopic disorders observed were: sole bruising, claw deformities, heel erosion, subclinical laminitis sole haemorrhages, double soles, chronic laminitis and white line separation. Radiographic changes observed mainly on distal phalanges were dilated vascular channels, irregular margins, exostoses/periostitis, distal phalangeal narrowing and lysis. Histopathologic changes in the corium included arterio-venous shunts, vascular wall rupture and thickening, vascular proliferation and thrombosis, corium and connective tissue oedema, degeneration, haemorrhages and spongiosis. Hence macroscopic, radiographic and histopathologic changes in laminitis claws affect locomotion.

Keywords: laminitis, sole bruising, corium, claw horn, vascular channels, pedal bone

1. Introduction

This chapter is about a descriptive presentation of the anatomic and physiological changes of the claws, which are the most vital part of the locomotor system of a dairy cow. The health of the claws allows free movement of the cow, which enhances adequate feeding time that leads to optimal reproduction and production, thus fulfilling the farmers' main purpose for keeping dairy cows. Over 80% of the dairy cattle herd in Kenya is under smallholder production [1], in most of which the dairy cows are kept in zero-grazing units with the animals confined for a long time [2]. Many of these smallholder zero-grazing units have improperly designed cattle-rearing structures, defective and unsuitable treading floors, inadequate and inconsistent nutritional diets, as well as generally poor husbandry and management practices [2]. All these factors interact synergistically and predispose dairy cows to laminitis and related non-infectious claw disorders [2–3]. It has been observed that housing of dairy cows in confinement leads to an increased prevalence of claw disorders. This is particularly true when the cows are confined on hard unyielding floors such as concrete, which exert immense pressure against the claws after loading them heavily under the opposing animal weight to ground forces [4–5]. Some of the non-infectious claw disorders such as white line separation, sole ulcer, double (underrun) soles, sole bruising/sole erosion and heel erosion can be the result or the cause of laminitis [6].

Laminitis tends to affect all the claws simultaneously in a single cow and when it advances to the chronic phase, there are obvious macroscopic claw deformities that make it difficult to reshape them to their normal anatomical appearance. These macroscopic deformities are often accompanied by irreversible damage to the internal structure of the claws [6–9]. Initially, laminitis occurs in a subtle clinically unrecognised state referred to as subclinical laminitis, which can only be discerned through claw trimming as sole and white line haemorrhages [6, 10, 11]. The haemorrhages are associated with pododermal microvasculature changes that lead to extravasation of serum and blood elements with subsequent staining of the internal layers of the horn next to the corium of the claw and later following horn-growth towards the surface of the sole, becomes visible externally as sole haemorrhages [6, 11, 12]. If claw trimming or keen observation is not done routinely in individual dairy cows, subclinical laminitis advances to chronic phase of laminitis and predisposes to laminitis-related non-infectious claw disorders with likelihood of occurrence of irreversible internal damage in the claws [6, 13].

The structural changes in the claws that result from laminitis and laminitis-related disorders are initially evident histologically, later they can be seen macroscopically and radiographically. Publications on comprehensive description of macroscopic, radiographic and histopathologic changes occurring in the claws during laminitis and laminitis-related disorders in dairy cows are scarce. The available publications describe scantly and separately the changes discernible through these three observational methods. The aim of this chapter is to comprehensively describe in a single publication the macroscopic, radiographic and histopathologic changes in the claws of dairy cows following subclinical and chronic laminitis, as well as laminitis-predisposed claw disorders. The study observed claws collected from abattoirs, which originated from slaughtered culled dairy cows. The publication also presents a comprehensive literature review on the structural changes affecting the claws from the various non-infectious disorders, especially laminitis.

2. Literature review

Laminitis is an aseptic diffuse inflammation of the pododerm (corium), which is the dermis of the claw. There is a progressive damage of the microvasculature of the corium, which compromises oxygen and nutrient supply to the corium cells, thus resulting in the production of low quality weak claw-horn [6, 14]. The low quality horn predisposes the claws of dairy cows to occurrence of various claw disorders associated with disruption of the claw-horn. These include sole ulcer, white line separation, double (underrun) soles, sole bruising (erosion) and heel erosion [6–8]. Claw-horn disruption is mainly thought to emanate from laminitis; hence, conclusion of association between laminitis and the claw disorders mentioned above [15]. Another factor that has the likelihood of causing claw-horn disruption in dairy cows is exertion of excessive forces on the germinal epithelium responsible for production of new claw horn. These excessive forces penetrate into the claw after depletion of adipose tissue in the digital cushion and the sole soft tissue thickness. The overall digital cushion and the sole soft tissue thickness become thinner to the extent that they cannot dissipate the concussion forces [16]. These authors have stated that the likelihood of having claw lesions has been associated with thin digital cushion. Impaired claw function has also been traced to weakened claw structures including subcutaneous tissue (digital cushion), dermis (corium) and suspensory apparatus. These lead to the damage of the vascular system of the dermis, disruption of dermal-epidermal junction as well as the horn producing cells [17].

The prevailing factors during the peri-parturient period predispose dairy cows to likelihood of more claw-horn wear and negative horn-growth. This effect leads to thinned soles that subsequently allow pressure penetration into the corium exacerbating its fragility and injury, consequently producing poor quality horn. Other structural changes in the claws occurring during the peri-parturient period in dairy cows include the slight shifting of the distal phalanx within the claw horn capsule. The distal phalanx rotates and sinks lower into the claw capsule. This movement of the distal phalanx is incriminated on the rising levels of the enzyme “hoofase” that occurs in the peri-parturient period, which causes more flexibility of the connective tissue suspending this bone within the claw horn capsule [18]. Studies have shown that sole haemorrhages were absent in heifers that had not calved, while these haemorrhages occurred after the heifers calved [19]. This confirms the role played by peri-parturient factors in predisposing to the occurrence claw-horn lesions. During the peri-parturient period, digital cushion and sole soft tissue become thinner, hence the germinal epithelium of the corium becomes predisposed to pressure. This results in production of poor quality horn as well as reduced horn production, especially the horn of the sole [16]. The alterations of thickness of sole soft tissue during the peri-parturient period are thought to be influenced by reproductive hormones relaxin and oestrogens. These hormones cause activation of the enzymes metalloproteinases, which degrade collagen [20]. The scores of lesions in the sole are higher during the postpartum period. These lesions decrease in the late lactation. Wider outward angle of the hind limbs caused by the size and weight of the udder in early lactation, predisposes the claws especially the lateral claws to occurrence of lesions [21]. Apart from hormonal factors during early postpartum period and in early lactation, metabolic changes associated with parturition and lactation similarly contributes to increased locomotion as well as lesion scores in dairy cows [20]. Lesions causing structural changes in the claws are more common and with higher locomotion scores in dairy cows that have had greater number of parities/lactations, especially five or more lactations [21].

Dairy cow housing-related factors are major contributors to the occurrence of claw lesions and structural changes. This is particularly common in the more confined dairy cows such as the smallholder zero-grazing units [2]. The type of floor on which the cows live and walk is the main housing factor predisposing them to develop claw lesions. This is determined by either rough or slippery floor texture [22], as well as defective concrete floors with small or large pot-holes [2]. The distribution of the weight of the cow on the claws is influenced by the hardness of the floor. Hard floors cause most of the animal weight to be loaded on the abaxial wall of the claws, while soft yielding floors lead to significant distribution of weight to the sole. The overall effect of the weight force against the mechanical pressure from the floor depends on the architectural arrangement of the claw wall determined by the interrelationship arrangement of tubular, intertubular and laminar horn cells. The numerical density of the horn tubules determines the differences in the stiffness and elasticity of the various parts of the claw horn capsule. The degree of the effect of the mechanical forces from the animal weight and floor pressure is associated with the degree of horn capsule elasticity and stiffness [22]. The longer the dairy cow stands on hard unyielding floor such as concrete, the more the likelihood of developing claw lesions [23]. Besides the pressure transmitted to the inside of the claws from the hard floors, the floor abrasiveness has a significant contribution to the occurrence of claw lesions, mainly due to excessive wearing of the claw-horn [24]. The claws are particularly prone to this abrasiveness of the floor when the horn produced is weak following bouts of laminitis [8]. Conversely, the claw-horn does not wear off when the cow lives and walks on soft yielding floors such as earthen floors or straw yards. This leads to inevitable overgrowth of the claws with subsequent overloading of the region towards the heel bulb, which is soft, hence the ease of transmission of pressure to the inner claw tissues, thus damaging them. The result of this is the production of poor quality horn [23]. All these factors have an overall effect of causing changes in the claw shape as well as structural changes internally.

3. Materials and methods

3.1 The study area

The study was carried out in the peri-urban areas of Nairobi City in the Republic of Kenya. The area was chose owing to its high number of smallholder zero-grazing dairy units. The high number of smallholder zero-grazing dairy units is instigated by the availability of market to which farmers sell milk and the value-added dairy products. The market is from the high number of city dwellers and workers. Nairobi and its peri-urban region have an area of 696 km^2 with estimated over 2.5 million people. It is located between latitude 01° 18′S and longitude 36° 45′E with an altitude of 1798 m above sea level. The estimated annual rainfall is maximum of 765 mm in the season from March to June, and a minimum of 36 mm in the season from October to December. The other areas from where samples for the study were collected included an abattoir in Kiserian centre of Kajiado County, Kenya and a slaughter slab in Wangige area of Kiambu County, Kenya.

3.2 The study design

This was a prospective abattoir study in which claws were collected from slaughtered dairy cows. Claws from an abattoir and a slaughter slab were subject to the number of dairy cows slaughtered per given time.

3.3 The sample size and sample selection

The two places selected from which dairy cow claws were collected included Kiserian abattoir and wangige slaughter slab. They were purposively selected based on the findings of an earlier pilot survey indicating that culled dairy cows were regularly slaughtered in these two places. Slaughter of dairy cows is not commonly done, hence the difficulties in getting their claws. The dairy cows send to slaughter were culled as a result of reasons related with poor production, poor reproductive performance, non-treatable problems and old age. The combined number of dairy cow feet collected from both slaughter places was 159, which made a total of 318 claws. A total of 96 feet were from Wangige slaughter slab, which had more dairy cows slaughtered and 63 were from Kiserian abattoir. Among the 159 feet, 109 were from the hind limbs and 50 from the forelimbs. More hind limb claws were collected based on the literature knowledge that they suffer more problems than forelimb claws. Since only a small number of dairy cows are slaughtered, it necessitated collection of the available claws without any random selection. History indicated that all the cows from which the claws were collected had been kept under zero-grazing system.

3.4 Examination of the claws for data collection

Each of claws collected from the abattoir and the slaughter slab was thoroughly washed. Observation of gross appearing lesions was done. Each lesion/abnormality was recorded in data collection sheets. Each claw was radiographed in dorso-palmar or dorso-plantar views. The radiographs were viewed and examined thoroughly. All radiographic changes seen were recorded. Owing to the time lapse between slaughter and examination, the claws were partially desiccated by loss of their water content, making trimming difficult. Therefore, after radiography, the area covered by the horn capsule was dipped in water for 2–3 h to make them softer for facilitation of trimming. Trimming was done by removing 2–3 mm of the horn of the sole, while observation of any lesion on the sole was made. All lesions were recorded. About 10 claws with subclinical laminitis and another 10 claws with chronic laminitis were purposively selected. Sagittal sections of all these 20 selected claws were done starting from the fetlock joint level through proximal, middle and distal phalanges. All gross lesions observed in the exposed corium and the phalanges after sagittal section were recorded.

The horn of the sole was removed from each claw using a knife to expose the corium of the sole fully. Sections of about 5 mm thickness of the corium specimens were harvested in transverse and longitudinal planes from each of the claw samples. The corium specimens were further sectioned into 1–2 millimetre pieces, which were dehydrated using ascending concentrations of absolute isopropyl alcohol, that started from 80 to 100%. Clearing of the tissues was done in xylene, then embedded in paraffin wax, blocked on wooden chunks, followed by fixing on the microtome. The tissues that were in paraffin wax blocks were cut into 5 μm thick pieces. From each block, four pieces of 5 μm thick tissues were made. This was followed by dewaxing the tissue sections in xylene and hydrated using graded alcohol whose concentration was from 100 to 50%. It was cleared in xylene, alcohol and washed in water. The tissues were stained with haematoxylin and eosin (H&E). Following dehydration, they were mounted on microscope slides and cover slips with Destrene 80, dibutyl phthalate and Xylene (DPX) mountant according to Ref. [25]. The slides were examined under light microscope using ×10, ×40 and ×100 objective lenses. The results were recorded and photomicrographs taken as necessary. The control in this study was done by comparing the findings in the normal claws with those that had lesions or were abnormal. The corium samples from claws that did not show signs of subclinical or chronic laminitis were processed using similar procedure as the test specimens and used as controls.

3.5 Data management and analysis

Claw lesions were given numerical codes for ease of entry into the computer. The prevalence of claw lesions was computed using Microsoft Office Excel data analysis tool as the number of dairy cow abattoir feet/claws showing a specific type of lesion divided by the total number of feet/claws examined multiplied by 100 to make it into percentage. Percentages of occurrence of radiographic changes were calculated by dividing the number of claws showing a specific radiographic feature by the total number of claws examined through radiography multiplied by 100. The percentages of histopathologic changes were calculated out of the number of claws with subclinical laminitis and chronic laminitis selected for histological evaluation.

3.6 Ethical approval

Permission to collect claws from the abattoir and the slaughter slab was granted by the Director of Veterinary Services, Kenya through the Veterinary Officers in the two places, respectively. The ethical approval of the proposal was given by the Biosafety, Animal Use and Ethics Committee of the Faculty of Veterinary Medicine, University of Nairobi, Kenya.

4. Results of the study

4.1 Macroscopic changes

Various macroscopic lesions/disorders were observed in the claws collected from the abattoir and from the slaughter slab. These included claw deformities (consisting of overgrown claws, flattened claws, concave dorsal wall and corkscrew claws) at a prevalence of 78.0% (n = 248) of the claws. Sole bruising (erosion) was seen in 44.0% (n = 140) of the claws and heel erosion at 41.5% (n = 132) of the claws. Others included subclinical laminitis (only evidenced by localised sole haemorrhages after a thin layer of the sole was trimmed-off), which was found in 34.6% (n = 110) of the claws, double (underrun) soles in 23.2% (n = 74) and chronic laminitis (evidenced by some types of claw deformities and more diffuse severe sole haemorrhages) in 21.4 (n = 68) of the claws. The rest of the lesions with low prevalence can be seen in **Table 1**.

Severe congestion of the sole corium was observed in the claws that had subclinical laminitis as well as those that had chronic laminitis. The congestion was more severe in the claws that had chronic laminitis than in those that had subclinical laminitis. On sagittal section, the pedal bone appeared dark red in claws with laminitis when compared to the normal claws.

Sole bruising (erosion) caused severe thinning of the horn of the sole, which made the corium to be positioned extremely close to the surface of the sole. This was clearly evident after the sagittal section in comparison to the normal claw (**Figure 1**). The thinning of the horn was more evident in sole bruising than in heel bruising (erosion). The claws that had sole bruising were also found to manifest sole haemorrhages. When the necrotic appearing horn of the bruised sole was trimmed, the protective function of the remaining horn was compromised due to the degree of thinness. Double soles and the degree of sole erosion became clearer after claw trimming was done (**Figure 2**).

Claw deformities ranged from mild to severe and usual deformities that clearly impaired the normal gait causing the cows to show obvious signs of lameness. Depending on the extent and degree of deformity, weight distribution on the claws was grossly unequal and not according to the natural normalcy.

Claw lesions/disorders	Number of cow feet (n = 159)	Number of claws (n = 318)	Prevalence (%)
Sole bruising	124	248	78.0
Claw deformities	70	140	44.0
Heel erosion	66	132	41.5
Subclinical laminitis	55	110	34.6
Double (underrun) sole	37	74	23.3
Chronic laminitis	34	68	21.4
White line separation	26	52	16.4
Foreign bodies	5	10	3.1
Sole ulcer	1	2	0.6
Claw infection	1	2	0.6

Table 1.
Percentage occurrence of macroscopic lesions/disorders out of 159 dairy cow feets (318 claws) collected from Kiserian abattoir and Wangige Slaughter slab in Kajiado County and Kiambu County, Kenya, respectively.

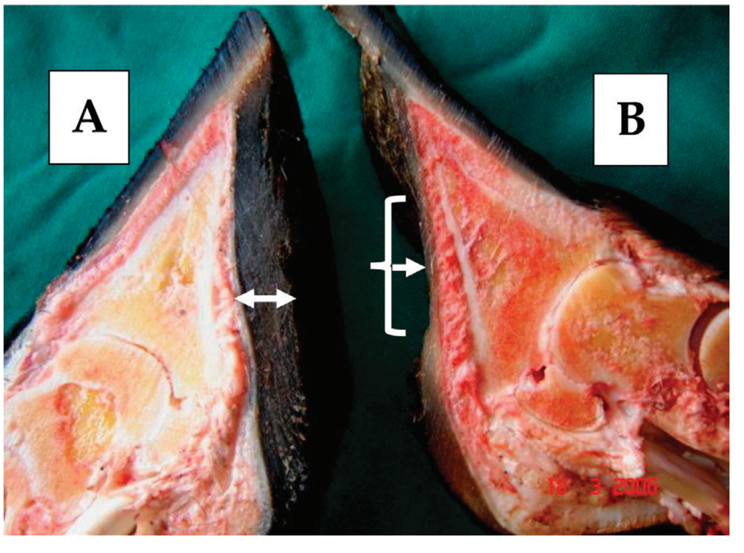

Figure 1.
Sagittal section of a normal claw (A) showing adequate thickness of the horn of the sole (double-headed arrow); and a claw with severe sole bruising (erosion) (B) showing extremely thinned horn of the sole with the corium very close to the treading surface (brace and arrow).

4.2 Radiographic features

The claws that were collected from the abattoir and the slaughter slab had varying prevalence of radiographic changes that occurred in the distal phalanges, particularly in those that had chronic laminitis. Other radiographic changes seen affected the blood vessels. Out of the 159 abattoir feet samples examined, 25.8% (n = 41) had more than one radiographic lesion. The most prevalent and consistently seen radiographic features were: prominently dilated vascular channels in 60.8% (n = 97), prominent but not dilated vascular channels in 24.1% (n = 38), irregular pedal bone margins in 13.9% (n = 22), pedal bone exostoses in 9.4% (n = 15) and narrowed/tapering pedal bone in 5.7% (n = 9) of the 159 dairy cow feet samples. Other radiographic features seen occurred in less than 5% (**Table 2**). Radiographic changes in

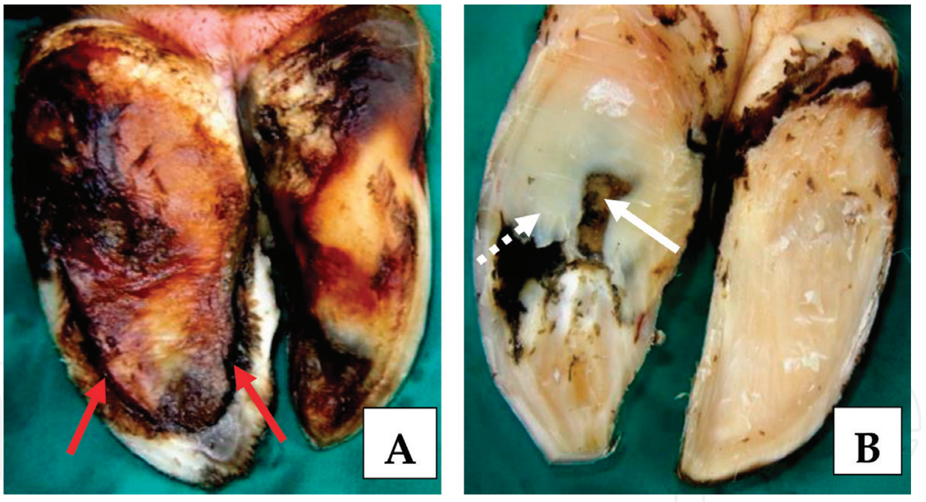

Figure 2.
Claw with double (underrun) soles whose margins are clearly distinct as shown (bold arrows) in A. The thin inner sole in B is exposed (bold arrow) after trimming-off part of the outer superficial sole (dotted arrow). This occurred concurrently with chronic laminitis in some dairy cow claws.

laminitis and other claw disorders were only observed in the distal phalanx. The middle and proximal phalanges did not show any changes. Vascular channels were markedly prominent in the distal ends of pedal bones of the claws that had laminitis.

These were more prominently dilated in claws with chronic laminitis, visible in claws with subclinical laminitis and hardly visible in claws without laminitis (**Figure 3**). The margins of the pedal bone from claws with chronic laminitis were small irregular serrations towards the distal end of the bone. The pedal bones from claws with subclinical laminitis and other laminitis-related disorders did not show any irregular margins (**Figure 4**).

Slight osseous overgrowths were seen on the periosteum at the distal parts of distal phalanges in some of the claws with chronic laminitis. Similar findings were seen in some of the claws with severe deformities. This radiographic feature seemed like exostosis (**Figure 5**). One of the claws with chronic laminitis had pedal bone with exostosis on the plantar surface (**Figure 5**). None of the claws with subclinical laminitis and other laminitis-related lesions had bone periosteal overgrowths.

Radiographic lesion	Number of feet samples (n = 159)	Number of claws (n = 318)	Percentage occurrence (%)
Dilated vascular channels	96	192	60.4
Prominent but not dilated vascular channels	38	76	23.9
Irregular pedal bone margins	22	44	13.8
Exostoses of pedal bone	15	30	9.4
Narrowed (tapering) pedal bone	9	18	5.7
Absent pedal bone apex	4	8	2.5
Fractured pedal bone	3	6	1.9
Periostitis of pedal bone	2	4	1.3
Osteolysis of pedal bone	1	2	0.6

The changes were observed in claws with laminitis and/or laminitis-related lesions/disorders.

Table 2.
Radiographic changes observed out of the 159 dairy cow feet examined from Kiserian abattoir, Kajiado County and Wangige slaughter slab, Kiambu County, Kenya.

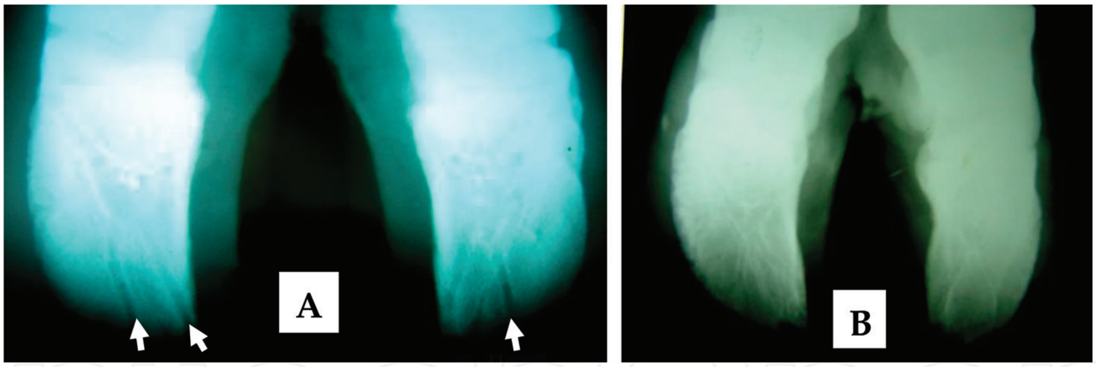

Figure 3.
Prominently dilated vascular channels (arrows) seen in claws with chronic laminitis (A), compared with unnoticeable vascular channels in a normal non-laminitis claw (B).

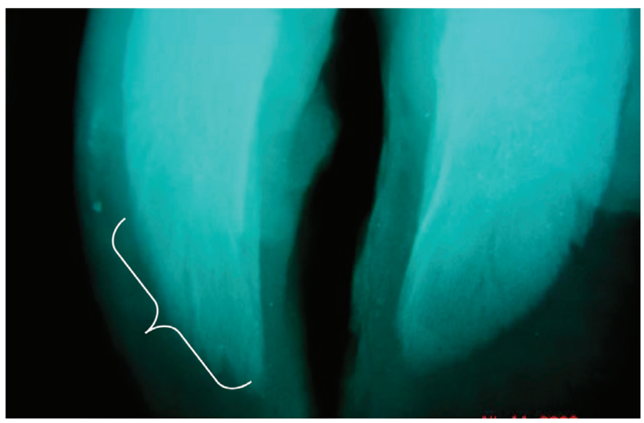

Figure 4.
Irregular serrations of the distal margin of the pedal bone (area with brace enclosure) in some of the claws that had chronic laminitis. Compare with the ipsilateral claw, which shows no serrations on the distal margin.

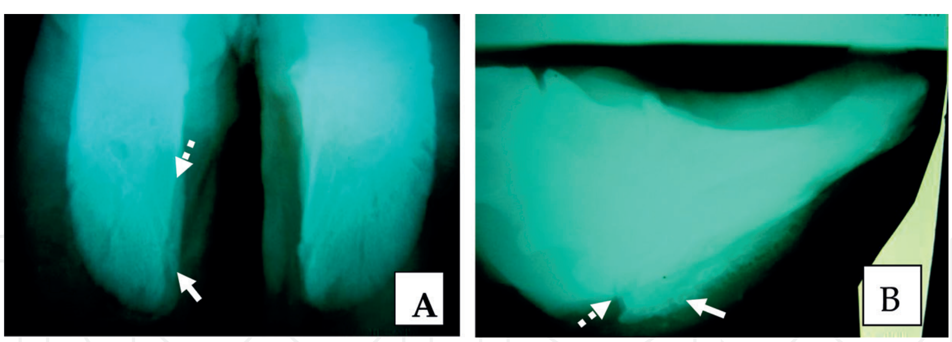

Figure 5.
Periosteal-like exostosis (bold arrow) with definite margin (dotted arrow) on the medial side of pedal bone in A. Exostosis (bold arrow) on the plantar surface of pedal bone with definite margin (dotted arrow) in B. These exostoses were found in some of the pedal bones from claws of dairy cows with laminitis.

Extreme tapering of the distal parts of the pedal bone was observed in some of the claws that had chronic laminitis with severe deformities such as twisted toe. The pedal bone was markedly narrowed (**Figure 6**). The other laminitis-related disorders including subclinical laminitis did not show this radiographic feature.

Another radiographic feature seen in claws with chronic laminitis and extreme twisting of the toe is absence of the sharp apex of the distal phalanx. The apex of the bone appeared more rounded than the normal ipsilateral claw (**Figure 7**). Claws with chronic laminitis but without deformity of the toe did not have this pedal apical feature. One of the radiographic features observed in claws with excessive sole bruising and heel erosion coupled with claw deformities was fracture-like fissures of the distal

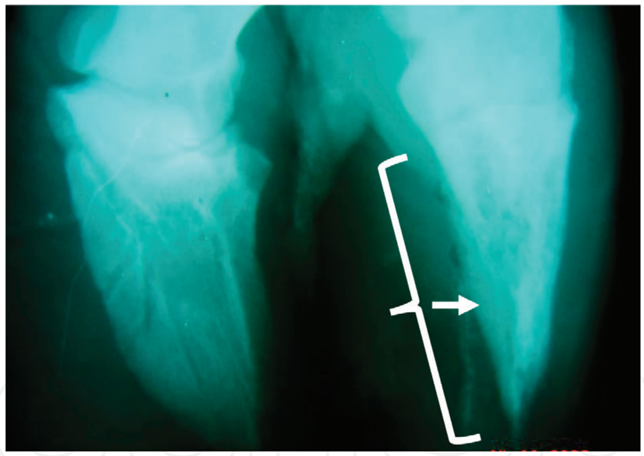

Figure 6.
Narrowed pedal bone (arrow) whose extent of tapering is shown (brace). The pedal bone of the ipsilateral claw is normal width. This radiographic change was observed in dairy cow claws with laminitis.

phalanx. The fissure was small and the part that appeared like bone fragment was still aligned to the entire bone (**Figure 8**). One distal phalanx was found to have osteolysis, which was clearly manifested by radiolucency of the bone appearing like the bone has a large medullary cavity (**Figure 9**), which is naturally absent in the pedal bone.

4.3 Histopathological features

Arteriovenous shunts (AVs), disruption of the dermal-epidermal junction, rupture of the vascular wall, vascular thrombosis and vascular proliferation were the main histological changes frequently observed in the claws with laminitis and the laminitis-related lesions. Other histological changes observed but rare included oedema in the dermis of the sole, connective tissue oedema, thickened arterial wall, degeneration and necrosis of the connective tissue that supports the distal phalanx, damage of the epidermal cells, spongiosis of the stratum basale and stratum spinosum, corium haemorrhages, connective tissue degeneration and enlargement of veins (**Table 3**).

Arteriovenous shunts (**Figure 10**) were observed in 40 and 50% of the corium specimens from claws with subclinical laminitis and chronic laminitis, respectively. Vascular wall damage (**Figure 11**) and dermal-epidermal junction disruption (**Figure 12**) were each observed in 30 and 50% of the corium specimens from claws with subclinical and chronic laminitis, respectively. Spongiosis was seen in 30% of

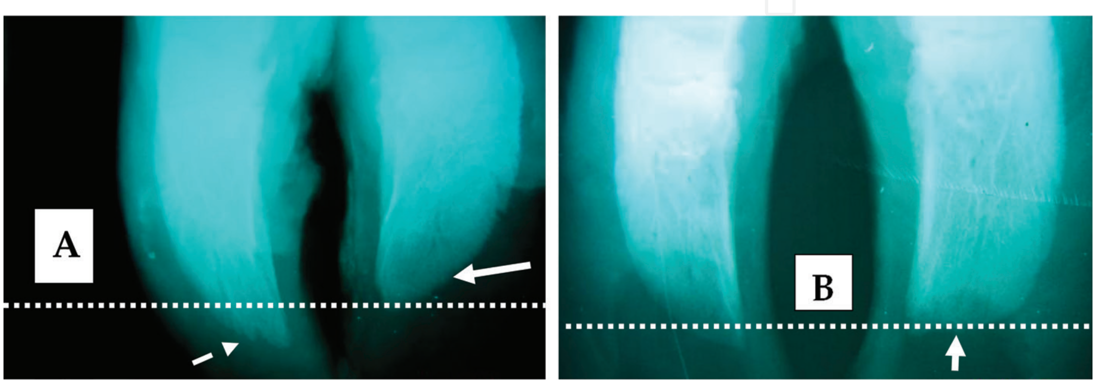

Figure 7.
A rounded (missing) apex of the pedal bone (bold arrow) making the bone look shorter than the ipsilateral pedal bone as shown (dotted line) in A and B. The projected apical part of the ipsilateral pedal bone is slightly irregular with serrations (dashed arrow). These radiographic changes were observed in dairy cow claws that had laminitis.

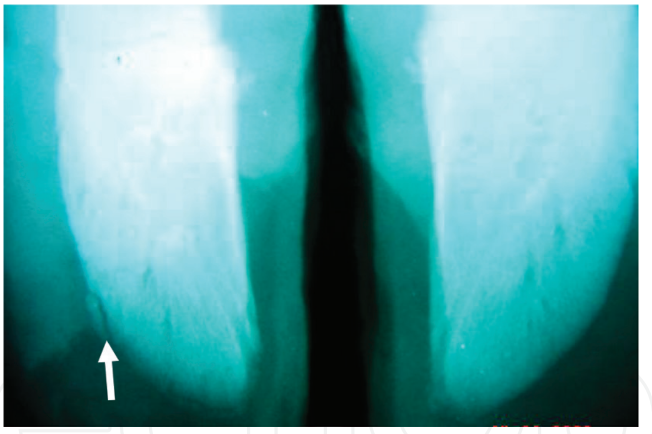

Figure 8.
A small fracture-like fissure (arrow) observed in three pedal bones from dairy cow claws that had laminitis.

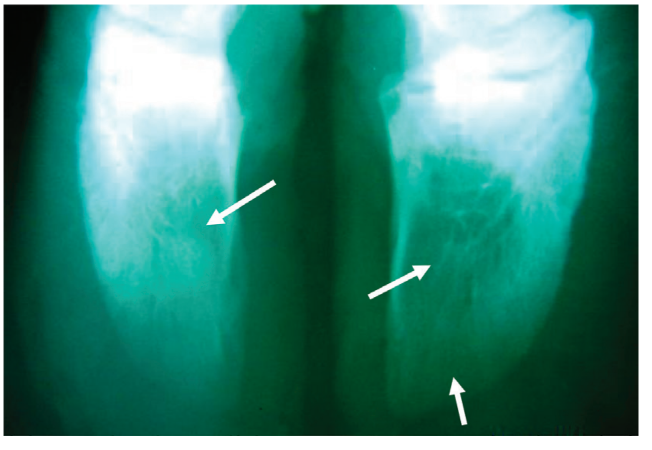

Figure 9.
Pedal bones showing radiolucent areas representing bone lysis (arrows). The areas look similar to medullary cavity, which is normally absent in normal pedal bones. This was observed in one dairy cow claw that had chronic laminitis.

Histopathological lesions	Prevalence (%) in subclinical laminitis (n = 10)	Prevalence (%) in chronic laminitis (n = 10)
Arteriovenous shunts	40	50
Vascular wall damage	30	50
Dermal-epidermal junction disruption	30	50
Vascular thrombosis	40	100
Vascular proliferation	30	30
Oedema of corium and connective tissue	20	20
Vascular wall thickness	30	10
Connective tissue degeneration	0	30
Corium haemorrhages	10	20
Spongiosis	0	30

The claws were collected from Kiserian abattoir of Kajiado County and Wangige slaughter slab of Kiambu County, Kenya.

Table 3.
Histopathological changes observed in the corium tissue specimens harvested from dairy cow claws with subclinical laminitis and chronic laminitis.

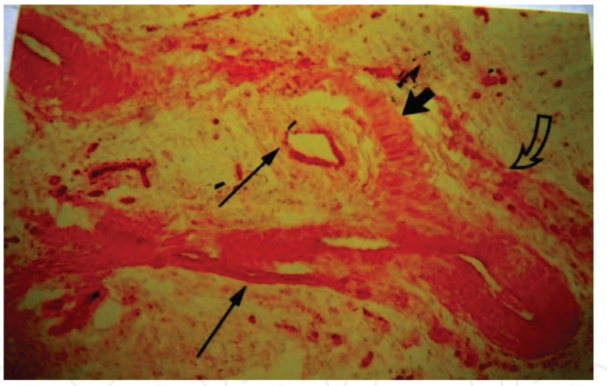

Figure 10.
Arteriovenous shunts shown by branching out of the wall of the artery (bold arrow) and inclusion of veins within the arterial wall boundaries (thin arrows) and the position of capillary-beds (curved arrows). These changes were seen in the corium of dairy cow claws with laminitis. (H&E stain, ×400).

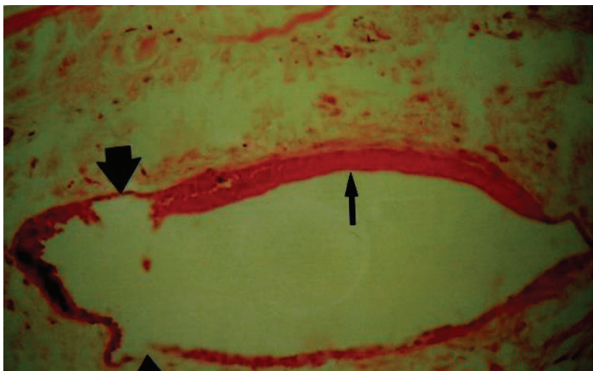

Figure 11.
A vein showing damaged wall (thick arrow) and a thrombus attached to the wall (thin arrow). This was a common feature in the corium of dairy cow claws with laminitis. (H&E stain, ×400).

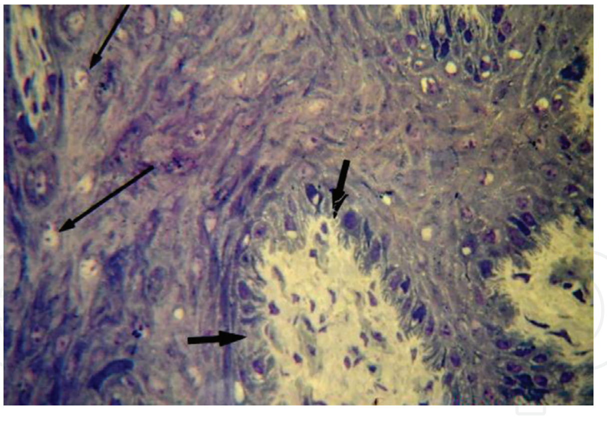

Figure 12.
Dermal-epidermal junction disruption shown by areas where the cells are not in contact and continuous with each other (short arrows). Intracellular oedema (spongiosis) (long thin arrows). These were common findings in cases of claws with laminitis. (Toluidine blue stain, ×400).

the specimens from claws with chronic laminitis (**Figure 12**), but it was not seen in claws with subclinical laminitis. Vascular wall damage was seen more regularly in the arterioles and venules than in the larger blood vessels. Endothelial disruption was seen more frequently than damage of the rest of the vascular wall. Rupture of the entire thickness of blood vessel wall followed by haemorrhage into the surrounding tissues was seen in 10 and 20% of the corium specimens from claws with subclinical and chronic laminitis, respectively. All the histological changes mentioned above were more common, extensive and severe in the corium specimens from claws with chronic laminitis than in those with subclinical laminitis.

Vascular thrombosis (**Figure 13**) was found in 70% of all corium specimens. All the corium specimens from claws with chronic laminitis were found to have vascular thrombosis, but only 40% of the corium specimens from claws with subclinical laminitis. Thickened vascular wall particularly of the arteries was found in 40% of the specimens, 30% from claws with subclinical laminitis (**Figure 13**). The vascular layer that appeared more prominently thickened was tunica media and in some of the specimens, it was evident that this thickening was due to oedema. Degeneration of the connective tissue that supports the distal phalanx was observed in 40% of the specimens, all of which were from claws with chronic laminitis. Oedema in the dermis of the sole and in the connective tissue (**Figure 14**) was found in 40% of the specimens, half from claws with subclinical laminitis and half from claws with chronic laminitis. Evidence of haemorrhages in the corium was found in 30 and 10% of the specimens from claws with chronic laminitis and subclinical laminitis, respectively. Mild degeneration of the sole connective tissue was seen in 30% of the specimens from claws with chronic laminitis (**Figure 15**). In 20% of all the specimens of claws with subclinical laminitis and chronic laminitis, the veins had enlargements that appeared like dilatations.

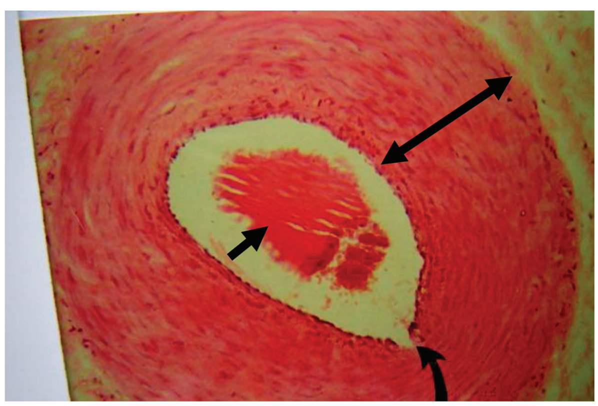

Figure 13.
Thickened arterial wall (double-headed arrow), disrupted arterial endothelium (curved arrow) and a thrombus in the lumen of the artery (single-headed arrow) (H&E stain, ×400). This was a feature observed in the corium vasculature of dairy cow claws with laminitis. (H&E stain, ×400).

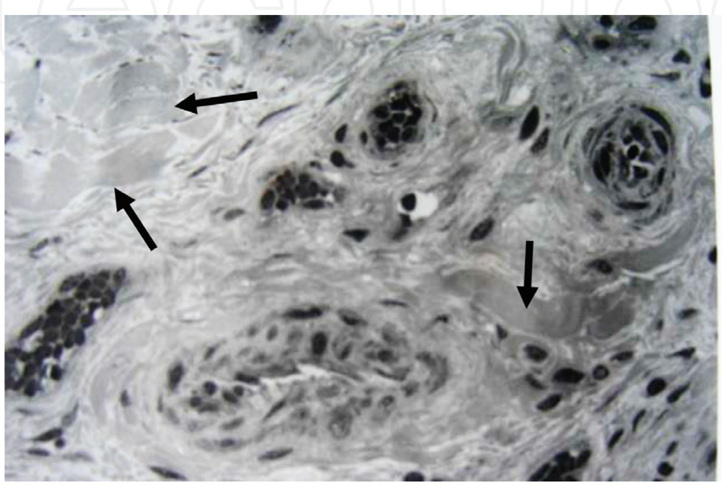

Figure 14.
Connective tissue oedema (arrows) seen in the corium of dairy cow claws with laminitis. (H&E stain, ×400).

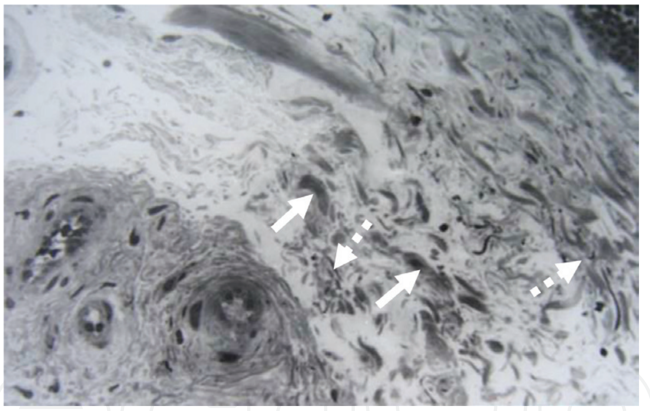

Figure 15.
Connective tissue degeneration manifested by deeply stained degenerated fibroblasts (bold arrows) and fibres (dotted arrows) in the histological sections of the corium of dairy cow claws with chronic laminitis (Toluidine blue stain, ×200).

5. Discussion

Laminitis influences the occurrence of other claw lesions and disorders such as sole ulcer, white line separation, double (underrun) soles, sole bruising (erosion) and heel erosion [6–8, 15, 26]. The occurrence of laminitis in dairy cows is predisposed by an interactive array of multiple risk factors [2, 6]. The high prevalence of laminitis and laminitis-related lesions/disorders in the dairy cow claws from Kiserian abattoir and Wangige slaughter slab can probably be attributed to the types of modern farming systems adopted in the dairy business enterprises in which the dairy cows are confined for long hours on hard unyielding concrete floors with much concentrate feeding. Such housing and feeding factors have been incriminated as risk factors for the development of laminitis [6, 7, 23, 27, 28]. The smallholder zero-grazing dairy units expose the cows to additional suboptimal management conditions such as prolonged presence of slurry, lack of claw trimming, and defective concrete floors apart from concentrate feeding [2, 8].

The high prevalence of prominently dilated vascular channels in the distal phalanges of the claws with laminitis particularly chronic laminitis corroborates previous documented observations that dilated vascular channels are consistent observations in subclinical and chronic laminitis. These previous reports indicated that in laminitis, both dilated and non-dilated vascular channels could be found [29]. The more prominent and excessively dilated vascular channels seen in chronic laminitis compared to subclinical laminitis could probably be associated with the greater damage internally in the claws during chronic laminitis [8].

The observed exostoses in the distal phalanges of claws with chronic laminitis are similar to an old report, which found exostosis in the pyramidal process of the pedal bone [30]. Occurrence of these exostoses could be associated with periostitis of the distal phalanges owing to prolonged inflammatory stimuli in chronic laminitis [8]. Although the factors associated with exostosis were not determined in this study, non-infective claw-horn disruption lesions (CHDL) have previously been incriminated as being associated with extra bone development particularly on the caudal aspect of the distal phalanx. The cows that had suffered chronic lameness from CHDL were found to have greater extra bone growth on the caudal aspect of distal phalanx, which was verified both by morphological gross observations and and histologically. This extra bone growth was more in quantity on the lateral claws of the hind limbs and medial claws of the forelimbs [31]. Extra bone development on the distal phalanges was reported to be positively correlated with the age of the cow [32]. Since, the claws in the current study were from culled aged dairy cows and had signs of non-infective claw-horn disorders, the findings corroborate these previous reports. It has also been suggested that infectious claw lesions are not associated with extra bone growth in the distal phalanges [31].

Narrowing (tapering) of the pedal bone towards the apex is a feature not previously described. Closely resembling this, is the atrophy of pedal bone reported in cattle with chronic laminitis [33]. This narrowing of the pedal bone could probably be attributed to osseous tissue necrosis (osteosis), which is linked to the deprivation of the nutrients and oxygen especially when local blood supply is compromised similar to what occurs in the claws with chronic laminitis [34]. Thromboembolic blockage of blood vessels with chronic local anaemia may also lead to osteosis unless supplemented by the presence of effective collateral circulation. However, bones have poor and inefficient collateral circulation owing to small anastomosing vasculature incapable of adequate compensatory dilatation. This makes them prone to osseous ischaemia that leads to necrosis with subsequent resorption of the necrotic bone by the osteoclasts [34]. If this process takes place during chronic laminitis with extreme twisting of the toe, then there would be likelihood of pedal bone narrowing. This could also explain the observation in some of the pedal bones that had absence of the apex from claws with chronic laminitis. Moreover, atrophy of the pedal bone reported previously in chronic laminitis could also be incriminated in the pedal apical absence [33].

Pedal bone osteolysis could be associated with reduced overall density of the bone in chronic laminitis as reported previously with crooked toe [30]. Productive periostitis of the pedal bone could be a response to irritation of the periosteum in claws with excessive inflammation of the corium in chronic laminitis. This could also lead to the irregular margins observed towards the distal margins of the pedal bone. The pedal bones in the lateral claws of the hind limbs had more severe and extensive radiographic changes than those of the medial claws. This corroborates earlier reports of higher prevalence and severity of lesions occurring on the lateral claws of the hind limbs of cattle than the medial claws [35, 36].

The observation of arteriovenous shunts (AVs) in this study was similar to previous reports in which the same was found [6, 37]. These shunts are normally not seen in healthy claws [38]. The arteriovenous shunts mainly involved the arterioles and venules. Arteriovenous shunts could be attributed to the fact that narrowed vessels have a likelihood of being affected by changes in blood pressure involved in the microcirculation of the corium [39], in which the arteriovenous shunts form as structural change adjustment [40].

The vascular damage in this study, which affected mainly dermal arteries, has been reported previously [38]. The pressure build-up from intermittent vasodilation and vasoconstriction of the corium microvasculature during laminitis may be the cause of this vascular damage [6]. The build-up of local blood pressure due to the initial increase in blood flow and pooling of blood within the corium capillary-bed, may lead to blood vessel rupture followed by haemorrhages and seepage of serum [6]. This eventually leads to the observation of haemorrhages seen macroscopically on the sole in cases of laminitis.

Thickening of vascular wall particularly of the arteries was similar to proliferated tunica intima, hypertrophied tunica media and fibrotic tunica adventitia reported earlier in laminitis [41]. The thickened arterial wall would obviously interfere with the function of the blood vessel. Vascular thrombosis is an almost invariable occurrence in the corium of claws with laminitis whether subclinical or chronic [6, 42]. The formation of thrombi might result from vascular damage particularly the endothelium, or from influence of the vasoactive substances within vascular circulation [6, 43]. Numerous capillary network is normal in the corium of the claws [40]. However, the vascular proliferation observed in this study, which led to an increased capillary network, could be a response to inflammatory reaction occurring in laminitis [14]. Enlarged veins seen only in chronic laminitis, which remained widely open appeared like venous dilatation rather than thickening of the wall. This feature has not been described previously in laminitis.

Oedema that was seen in the corium and in the connective tissue of the sole has been reported previously [6]. It occurs as a result of rising capillary pressure together with post-capillary resistance, which enhances transvascular fluid seepage followed by increased pressure within the tissues. Digital venous constrictions are thought to be the initial step in these processes [39]. Subsequently, oedema expands the corium, which causes pain that leads to lameness seen in laminitis [6, 44].

Although chronic degenerative change in the bovine corium of the claw has been reported in chronic laminitis previously [45], the disruptive connective tissue damage particularly involving fibroblasts has not been reported previously. This disruptive damage is probably a degenerative change associated with enzymatic action of matrix metalloproteinases (MMPs) and gelatinolytic protease known as hoofase [46]. It may also be a biochemical alteration in the connective tissue [20]. The degenerative disruption of the connective tissue was observed only in specimens from claws with chronic laminitis.

Disruption of the dermal-epidermal junction is an invariable finding in subclinical and chronic laminitis owing to compromised microvasculature of the corium, which causes nutrients and oxygen to be diminished to the extent of not reaching the epidermal cells. This causes breakdown of the stratum germinativum in the epidermis, the corium becomes degenerated and eventually a breakdown in dermal-epidermal junction that results in separation between the stratum germinativum and the corium [44]. Spongiosis and hyperplasia of the epidermis have not been reported previously. Both of these histological changes occurred in subclinical as well as in chronic laminitis. Probably, they are due to inflammation triggered by pressure irritation on the soft tissues located between the pedal bone and the horn of the sole after the claw comes into contact with the hard treading surface when the cow is in standing posture or in locomotion.

Previous studies have shown that laminitis and laminitis-related claw disorders have various cow-level and farm management-level risk factors [2, 6–8]. Although the cows whose claws were used in this study had been pregnant several times, the number of pregnancies and the stage of lactation were not ascertained from the zero-grazing units in which they were kept. It would therefore be recommended that further study be conducted relating macroscopic, radiographic and histopathologic findings on the claws with number of parities, state of pregnancy and stage of lactation of the cows. This will reveal the nature of correlation between these factors, thus indicating the factors exacerbating morphological changes on the dairy cow claws. This should also include correlation with the seasons of the year.

6. Conclusions

The study concluded that claw disorders are prevalent in dairy cows kept under zero-grazing system. The most commonly occurring macroscopic disorders are sole bruising (erosion), claw deformities, heel erosion, double soles, both subclinical and chronic laminitis and white line separation. The radiographic changes found in the claws were mainly associated with chronic laminitis. These occurred on the pedal bone and included dilated vascular channels, irregular bone margins, narrowing of the bone towards the apex and osteolysis. The histopathologic changes were common in the corium of both subclinical and chronic laminitis claws, which included mainly changes in the vasculature such as venular wall damage, arterial wall thickening, vascular thrombosis, connective tissue changes including oedema and degeneration. Most of these changes are evidently irreversible, hence making dairy cows suffering chronic laminitis coupled with extreme claw deformities to have poor prognoses.

Acknowledgements

We are grateful to the University of Nairobi Deans Committee for provision of funds to carry out this research. We thank the Chairman of the Department of Clinical Studies for allowing us the use of Departmental laboratory and imaging facilitation to carry out this research.

Conflict of interest

There is no conflict of interest for this project and publication whatsoever.

Author details

James Nguhiu-Mwangi[1]* and Peter M.F. Mbithi[1,2]

1 Department of Clinical Studies, Faculty of Veterinary Medicine, University of Nairobi, Kenya

2 Office of Vice-Chancellor, University of Nairobi, Kenya

*Address all correspondence to: jamesnguhiumwangi@gmail.com

References

[1] Wanyoike MM, Wahome RG. Small-scale farming systems. In: Workshop Proceedings on Cattle Production in Kenya-Strategies for Research Planning and Implementation. December 2003 KARI HQ. Published 2004; pp. 87-133

[2] Nguhiu-Mwangi J, Mbithi PMF, Wabacha JK, Mbuthia PG. Factors associated with the occurrence of claw disorders in dairy cows under smallholder production systems in urban and peri-urban areas of Nairobi. Veterinarski Arhiv. 2008;**78**(4):343-355

[3] Nguhiu-Mwangi J, Mbithi PMF, Wabacha JK, Mbuthia PG. Prevalence of laminitis and the patterns of claw lesions in dairy cows in Nairobi and the peri-urban districts. Bulletin of Animal Health and Production in Africa. 2009;**57**:199-208

[4] Van der Tol PPJ, Metz JMH, Noordhuizen-Stassen EN, Back W, Braam CR, Weijs WA. The vertical ground reaction force and the distribution on claws of dairy cows while walking on a flat substrate. Journal of Dairy Science. 2003;**86**:2875-2883

[5] Van der Tol PPJ, Metz JMH, Noordhuizen-Stassen EN, Back W, Braam CR, Weijs WA. The force and pressure distribution on the claws of cattle and the biomechanical effect of preventive trimming. In: Proceedings of the 13th International Symposium and 5th Conference on Lameness in Ruminants; 11-15 February; Maribor, Slovenija. 2004. p. 102

[6] Nocek JE. Bovine acidosis: Implications on laminitis. Journal of Dairy Science. 1997;**80**:1005-1028

[7] Belge A, Bakir B. Subclinical laminitis in dairy cattle: 205 selected cases. Turkish Journal of Veterinary and Animal Sciences. 2005;**29**:9-15

[8] Nguhiu-Mwangi J. Characteristics of laminitis and associated claw lesions in dairy cows in Nairobi and its environs [PhD Thesis]. Nairobi, Kenya: University of Nairobi; 2007

[9] Nguhiu-Mwangi J, Mbithi PMF, Wabacha JK, Mbuthia PG. Risk (predisposing) factors from non-infectious claw disorders in dairy cows under varying zero-grazing systems. In: Perez-Marin CC, editor. A Bird's-Eye View of Veterinary Medicine. Rijeka, Croatia: InTech; 2012. pp. 393-422. DOI: 10.5772/29795.ch21. ISBN: 978-953-51-0031-7

[10] Bergsten C. Haemorrhages of the sole horn of dairy cows as a retrospective indicator of laminitis: An epidemiological study. Acta Veterinaria Scandinavica. 1994;**35**:55-66

[11] Nguhiu-Mwangi J, Mbithi PMF, Wabacha JK, Mbuthia PG. Prevalence of sole haemorrhages and its correction with subclinical and chronic laminitis in dairy cows. Bulletin of Animal Health and Production in Africa. 2007;**55**:232-242

[12] Hirschberg RM, Plendl J. Pododermal angiogenesis and angioadaptation in the bovine claw. Microscopy Research and Technique. 2005;**66**:145-155

[13] Manske T, Hultgren J, Bergsten C. The effect of claw trimming on the hoof health of Swedish dairy cattle. Preventive Veterinary Medicine. 2002;**54**:113-129

[14] Bell EL. Description of claw horn lesions and associated risk factors in dairy cattle in the Lower Fraser Valley [MSc Thesis]. British Columbia, Canada: The University of British Columbia; 2004

[15] Nguhiu-Mwangi J, Mbithi PMF, Wabacha JK, Mbuthia PG. Disorders

of the claw and their association with laminitis in smallholder zero-grazed dairy cows. International Journal of Veterinary Science and Medicine. 2017;**6**(2):64-69

[16] Newsome RF, Green MJ, Bell NJ, Bollard NJ, Mason CS, Whay HR, et al. A prospective cohort study of digital cushion and corium thickness. Part 1: Associations with body condition, lesion incidence and proximity to calving. Journal of Dairy Science. 2017;**100**:4745-4758

[17] Mülling CKW, Greenough PR. Applied physiopathology of the foot. In: Proceedings of the 14th World Buiatrics Congress; 15-19 October 2006; Nice, France. 2006. pp. 103-117

[18] Blowey R. Factors associated with lameness in dairy cattle. In Practice. 2005;**27**:154-162

[19] Knott L, Tarlton JF, Craft H, Webster AJF. Effects of housing, parturition and diet change on the biochemistry and biomechanics of the support structures of the hoof of dairy heifers. Veterinary Journal. 2007;**174**:277-287. DOI: 10.1016/j.tvjl.2006.09.007

[20] Tarlton JF, Holah DE, Evans KM, Jones S, Pearson GR, Webster AJ. Biomechanical and histopathological changes in the support structures of bovine hooves around the time of first calving. Veterinary Journal. 2002;**163**:196-204

[21] Chapinal N, de Passillé AM, Rushen J. Weight distribution and gait in dairy cattle are affected by milking and late pregnancy. Journal of Dairy Science. 2009;**92**:581-588

[22] Franck A, Cocquyt G, Simoens P, De Belie N. Biomechanical properties of bovine claw horn. Biosystems Engineering. 2006;**93**(4):459-467

[23] Somers JGCJ, Frankena K, Noordhuizen-Stassen EN, Metz JHM. Prevalence of claw disorders in Dutch dairy cows exposed to several floor systems. Journal of Dairy Science. 2003;**86**:2082-2093

[24] Vokey FJ, Guard CL, Erb HN, Galton DM. Effects of alley and stall surfaces on indices of claw and leg health in dairy cattle housed in a free-stall barn. Journal of Dairy Science. 2001;**84**:2686-2699

[25] Luna LG. Mannual of the Histological Staining Methods of the Armed Forces Institute of Pathology. 3rd ed. New York: McGraw Hill; 1968

[26] Nguhiu-Mwangi J, Mbithi PMF, Mbuthia PG. Laminitis and laminitis-associated claw conditions in smallholder zeo-grazed cows. In: Claw Disorders in Dairy Cows Under Smallholder Zero-grazing Units. Germany. ISBN: 978-3-639-66879-7, ch3: Scholars' Press, OmniScriptum GmbH & Co. KG; 2015. pp. 86-130

[27] Somers JGCJ, Schouten WGP, Frankena K, Noordhuizen-Stassen EN, Metz JHM. Development of claw traits and claw lesions in dairy cows kept on different floor systems. Journal of Dairy Science. 2005;**88**:110-120

[28] Vermunt JJ. Herd lameness: A review, major causal factors and guidelines for prevention and control. In: Proceedings of the 13th International Symposium and 5th Conference on Lameness in Ruminants; 11-15 February 2004; Maribor, Slovenija. pp. 1-15

[29] Boosman R, Nemeth F, Gruys E, Klarenbeek A. Arteriographical and pathological changes in chronic laminitis in dairy cattle. Veterinary Quarterly. 1989;**11**:144-155

[30] Maclean CW. A postmortem X-ray study of laminitis in barley beef animals. Veterinary Record. 1970;**86**:457-462

[31] Newsome R, Green MJ, Bell NJ, Chagunda MGG, Mason CS, Rutland CS, et al. Linking bone development on the caudal aspect of the distal phalanx with lameness during life. Journal of Dairy Science. 2016;**99**:4512-4525

[32] Tsuka T, Ooshita K, Sugiyama A, Osaki T, Okamoto Y, Minami S, et al. Quantitative evaluation of bone development of the distal phalanx of the cow hind limb using computed tomography. Journal of Dairy Science. 2012;**95**:127-138

[33] Greenough PR, Vermunt JJ, Mckinnon JJ, Fathy FA, Berg PA, Cohen RDH. Laminitis-like changes in the claws of feedlot cattle. Canadian Veterinary Journal. 1990;**31**:202-208

[34] Jubb KVF, Kennedy PC, Palmer N. Pathology of Domestic Animals. 3rd ed. Vol. 1. New York: Academic Press, Inc Harcourt Brace Jovanovich Publishers; 1993. pp. 61-63

[35] Russell AM, Rowlands GJ, Shaw SR, Weaver AD. Survey of lameness in British dairy cattle. Veterinary Record. 1982;**111**:155-160

[36] Tranter WP, Morris RS, Dohoo IR, Williamson NB. A case-control study of lameness in dairy cows. Preventive Veterinary Medicine. 1993;**15**:191-203

[37] Vermunt JJ, Leach DH. A scanning elcectron microscopic study of vascular system of the bovine hind limb claw. New Zealand Veterinary Journal. 1992;**40**:146-154

[38] Hirschberg R, Mülling C, Budras KD. Pododermal angioarchitecture of the bovine claw in relation to form and function of the papillary body. A scanning electron microscopic study. Microscopy Research and Technique. 2001;**54**:375-385

[39] Christmann U, Belknap EB, Lin HC, Belknap JK. Evaluation of hemodynamics in the normal and laminitic bovine digit. In: Proceedings of the 12th International Symposium on Lameness in Ruminants; 9-13 January 2002; Orlando Florida, U S A. pp. 165-166

[40] Mülling CKW, Grönemeyer D. Foot Anatomy: New Methods of Investigation and Therapeutic Implications. Paris/Frankreich: Société Francaise de Buiatrie; 2004, 2004. pp. 157-161

[41] Andersson L, Berman A. Pathology of bovine laminitis especially as regards vascular lesions. Acta Veterinaria Scandinavica. 1980;**21**:559

[42] Singh SS, Murray RD, Ward WR. Histopathological and morphometric studies on the hooves of dairy and beef cattle in relation to overgrown sole and laminitis. Journal of Comparative Pathology. 1992;**107**:319-328

[43] Bergsten C, Mülling C. Some reflections on research on bovine laminitis aspects of clinical and fundamental research. In: Proceedings of the 13th International Symposium and 5th Conference on Lameness in Ruminants; 11-15 February 2004; Maribor, Slovenija. p. 94

[44] Shearer JK, van Amstel S. Lameness in dairy cattle. In: Proceedings of 2000 Kentucky Dairy Conference. pp. 1-11

[45] Boosman R, Koeman J, Nap R. Histopatholgy of the bovine pododerm in relation to age and chronic laminitis. Zentralblatt für Veterinärmedizin. Reihe A. 1989;**36**:438-446

[46] Tarlton JF, Webster AJ. A biochemical and biomechanical basis for the pathogenesis of claw horn lesions. In: Shearer JK, editor. Proceedings of the 12th International Symposium on Lameness in Ruminants; 9-13 January 2002; Orlando Florida. pp. 395-398