

# Investigating the potential role of abrasion in the development of toe tip necrosis in beef cattle: an ex vivo study

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## Objective

To compare regional stiffness of the white line (objective 1) and image-based metrics of damage (objective 2) of control claws and claws subjected to an abrasion simulator mimicking animals abrading their claws against a concrete surface commonly found in feedlots.

## Methods

Sixteen ( $n = 16$ ) cadaveric bovine hind limbs were acquired from participating commercial feedlots and separated into different testing groups: lateral claws subjected to an abrasion simulation ( $n = 8$ ) and control claws manually rasped to the same level of wear found after the abrasion simulation ( $n = 8$ ). Claws were subjected to indentation testing along the white line to determine regional stiffness (control = 8; abraded = 8) and contrast-enhanced, high-resolution imaging (control = 6; abraded = 6) where mean image intensity was used to characterize damage. Analysis of variance was used to compare regional stiffness and image intensity of the different groups.

## Results

Lower stiffness of the white line along the apical region was noted in abraded claws versus control claws ( $P < .019$ ). Higher mean intensity (a measure of damage) was found in abraded claws versus control claws ( $P < .026$ ).

## Conclusions

Study findings indicate that abraded claws exhibited lower stiffness along the apical region of the white line relative to control claws. Also, analyses of contrast-enhanced, high-resolution imaging data suggested that pathways for foreign material to enter the claw may be present following abrasion.

## Clinical Relevance

These findings support the premise that abrasion may be involved in white line separation and toe tip necrosis pathogenesis. Alternative floorings that minimize abrasion may be beneficial for avoiding toe tip necrosis.

**Keywords:** toe tip necrosis syndrome, bovine claws, abrasion, white line, stiffness

**T**oe tip necrosis (TTN) most commonly presents as a hind limb lameness in feedlot cattle within weeks of arrival to the lot. Confusion exists regarding the disease's nomenclature, with it also referred to as toe abscesses, toe ulcers, third phalanx (P3) necrosis, toe necrosis, and TTN syndrome, among other names.<sup>1</sup> Toe tip necrosis is characterized by separation of the white line, abscess formation, necrosis of P3, and associated sequelae.<sup>2</sup>

Generally, morbid cattle in commercial feedlots are identified by active syndromic surveillance and then treated according to an algorithm generated by the consulting veterinarian. A putative diagnosis of TTN is based on identifying a lower hind limb lameness with no signs of lesions or swelling and occurring early in the feeding period. Most are administered parenteral antimicrobials without further examination. Feedlot cattle identified in the early stages of TTN are often treated successfully with parenteral antibiotics,<sup>1,3</sup> but debridement of necrotic horn tissue and claw amputation may be warranted.<sup>4</sup> Approximately 3% of lame cattle are diagnosed with TTN, with 70% dying or being

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prematurely slaughtered at a cost of approximately \$41 million annually in Western Canada.<sup>5</sup>

The etiopathogenesis of TTN is not fully understood; however, the “abrasion theory,” which is the focus of this research, is highly plausible.<sup>2</sup> Briefly, this theory posits that cattle incur excessive wear to the apex of the toes of the hind limbs during shipping and handling, particularly when exposed to abrasive flooring, such as concrete. This abnormal wear leads to a thinning of the sole, which, when combined with repetitive loading-unloading associated with ambulation, results in microfissures along the white line and diminished mechanical properties (eg, white line exhibits a lower stiffness as in it is more deformable or flexible). These microfissures become colonized by bacteria commonly found in the earthen floors of feedlot pens. Bacterial byproducts and the host’s immune response further degrade the soft white line, leading to an increase in white line separation. Presumably, lameness manifests once the infection penetrates the corium, which is a highly innervated and vascularized tissue that envelopes P3. At this point, antimicrobial therapy alone may be successful in arresting disease progression. If, however, the infection breaches the corium, then P3 necrosis and related sequela, such as deep digital sepsis, will require more aggressive intervention (claw amputation) or even euthanasia. Other sequelae include an ascending tenosynovitis of the hind limb and bacteremia, resulting in the seeding of bacteria into distant tissues and organs, which often results in death. Thus, TTN is best viewed as a disease continuum beginning with apical white line separation and, in some cases, ending in death due to bacteremia or an embolic event.

The abrasion theory is supported by both anecdotal evidence and clinical studies. Anecdotal evidence suggests that TTN is more common in fractious animals and those coming off wet pastures, with the pasture conditions resulting in softer horn tissue.<sup>1,3,6,7</sup> Presumably, fractious animals are more likely to abrade their claws, especially when crowded in chute systems where they push on the animals in front of them, resulting in a rasping motion of their hindfeet.<sup>2,5,8</sup> This is supported by the finding that the apical region of the toe of claws with TTN is thinner than that of healthy claws.<sup>9</sup> Further, *Escherichia coli* and *Trueperella pyogenes*, common bacterial inhabitants of the bovine gastrointestinal tract, were the most prevalent bacteria recovered from TTN lesions.<sup>9</sup> As an aside, it is known that the mineral composition of the hoof horn tissue is correlated to tensile properties; thus, nutrition may also be a component of this disease.<sup>10,11</sup> It is also salient that a large-scale descriptive epidemiological study reported that the mean number of days on feed until diagnosis was 19 (median, 12), with some animals arriving to the feedlot with TTN.<sup>1</sup> These findings underscore that TTN in feedlot cattle is initiated at the time of transportation and handling; it is rarely reported in pastured beef cattle. Hence, 1 of the first monikers for the disease was “transit-related lameness.”<sup>12</sup>

Although white line separation is considered a key step in the pathogenesis of TTN, it is nearly

imperceptible in the early stages of TTN<sup>2,8,13</sup> and only becomes readily apparent in animals that develop lameness. Gyan et al<sup>13</sup> reported that 100% of cattle with TTN had apical white line separation, whereas 97% of non-TTN cattle had no evidence of white line separation, confirming that the apical region of the white line is a key region of interest in the development of TTN.

This study expands upon previous mechanical testing research investigating the role of mechanical loading and damage on white line separation. Previously,<sup>14</sup> the influence of repetitive fatigue loading (simulating normal physiologic loading during walking) on white line separation was assessed. Toe tip necrosis was simulated by manually rasping healthy claws down to a similar level of wear observed clinically. No association was found between fatigue cycle count and white line separation for both healthy (unrasped) claws and manually rasped claws. In addition, contrast-enhanced, high-resolution peripheral quantitative CT (HR-pQCT) scans of healthy and manually rasped claws showed no evidence of contrast agent penetration through the white line. However, the damage arising with manually rasped claws may not mimic the in vivo condition. To address this issue, we developed a novel abrasion simulator that mimics animals abrading their claws against different surfaces in feedlots.<sup>15</sup> A comparison study indicated that claws subjected to abrasion exhibited lower overall stiffness (ie, deformation response to an applied load) relative to healthy control claws.<sup>16</sup> As well, claws subjected to abrasion exhibited similar overall stiffness as claws diagnosed with TTN,<sup>16</sup> suggesting that abrasion may be involved in TTN pathogenesis. However, it is unclear how abrasion may affect the white line, which (as noted above) is a key region of interest in TTN pathogenesis.

Using a novel abrasion simulator, mechanical indentation testing, and contrast-enhanced, HR-pQCT imaging, the overall aim of this study was to investigate the potential role of physiologically realistic abrasion on the development of white line separation and TTN. This aim was addressed by meeting the following objectives: (1) comparing regional stiffness of the white line of control claws and abraded claws and (2) comparing image-based metrics of damage of control claws and abraded claws. We hypothesized that abraded claws exhibited lower stiffness along the white line and higher damage than control claws. Together, these hypotheses would point to abrasion potentially being involved in white line separation and the development of TTN.

## Methods

### Samples

For this study, 16 hind limbs from feedlot calves were acquired from participating commercial feedlots in Alberta, Canada. All limbs were regarded as healthy, with the causes of death (eg, pneumonia, myocarditis) being unrelated to any foot issues. The average weight of cattle from which the hind limbs

were obtained was 241 kg (SD, 55.3 kg) and ranged from 160 to 342 kg. In cases where the animal mass was not recorded, a linear regression equation linking claw area (length X width) with body mass (**Supplementary Figure S1**) was used to estimate the body mass needed for deriving physiologic loading conditions. The specimens were sectioned at the fetlock joint. The specimens had undergone a freezing process at  $-20^{\circ}\text{C}$  prior to arriving at the University of Saskatchewan, which ensured that they were well preserved and suitable for experimentation. After being received, they were stored in a freezer at  $-20^{\circ}\text{C}$  prior to excising and potting.

The 16 hind limbs were organized into different groups for experimental testing and matched in terms of body mass. The testing groups consisted of claws subjected to an abrasion simulation ( $n = 8$ ) and control claws manually rasped to the same level of wear found after the abrasion simulation ( $n = 8$ ). Claws were subjected to indentation testing along the white line to determine regional stiffness (control = 8; abraded = 8) and contrast-enhanced, high-resolution imaging (control = 6; abraded = 6) to assess damage and pathway development.

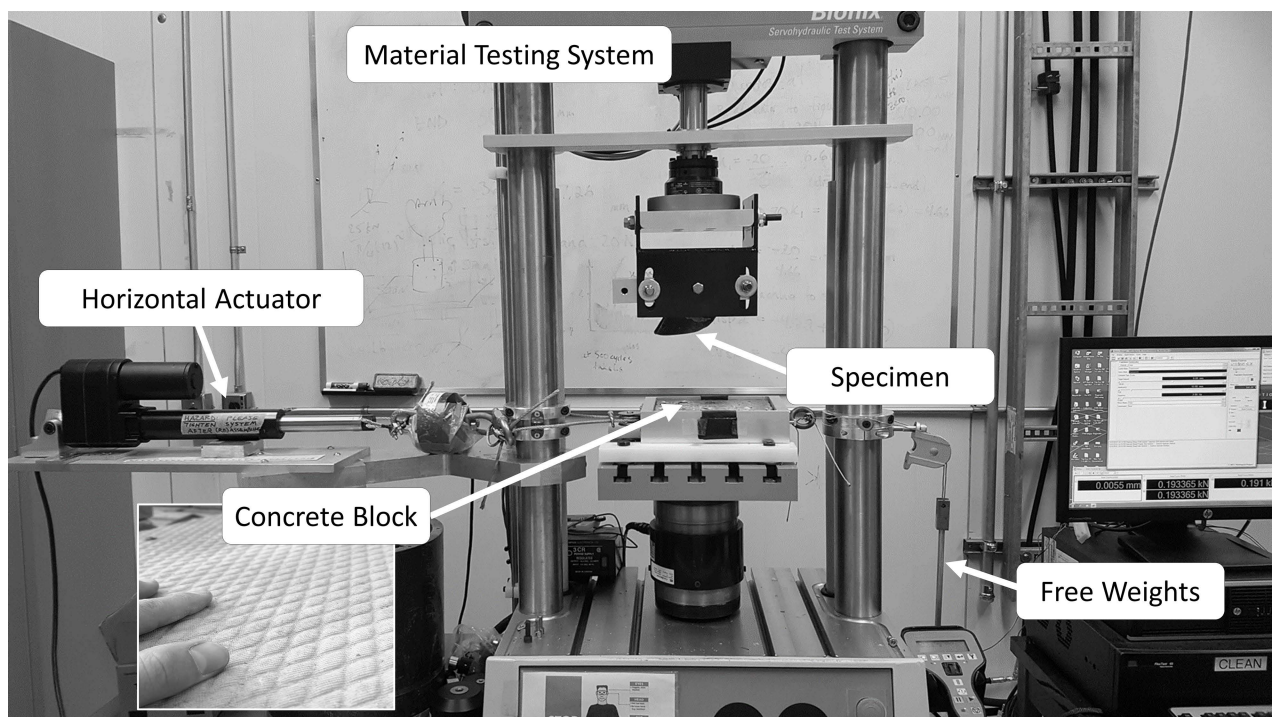
#### Specimen preparation

Specimens were thawed overnight, excised, and potted on the same day to minimize freeze-thaw cycles that could impact soft tissue mechanical properties<sup>17,18</sup> (previous research indicates that

freezing has no effect on bovine hoof mechanical properties, such as elasticity or stiffness<sup>19</sup>). Each specimen was sectioned horizontally at the second phalanx bone, then sectioned in the sagittal plane to obtain separate medial and lateral digits. Lateral digits, which support more load than medial digits,<sup>20</sup> were used in this study. Each lateral compartment was dissected down to the second phalanx bone, and all soft tissues were removed. Claws were then secured in a staged manner in a polyvinyl chloride pipe in an anatomical position. The staging consisted of first using polyester resin (Bondo Body Filler; 3M) and screws for initial alignment, followed by polymethylmethacrylate (Fastray; Bosworth) for rigid fixation. Claws were fixed with an angle of approximately  $10^{\circ}$  to the horizon to simulate the initial contact angle of the claws with the surface, with contact made by the apical region of the toe. The specimens were placed back in the freezer until mechanical testing.

#### Abrasion simulation

A novel abrasion simulator which mimicked animals abrading their claws against different surfaces in feedlots was used in this research (**Figure 1**). For this study, a concrete block representing common surfaces found in feedlots was used. The concrete block had a diamond pattern with grooves approximately 1.2 mm deep. For testing, a vertical servohydraulic material-testing system (Bionix 370.02 Axial/Torsional; MTS Systems Corp) applied



**Figure 1**—The abrasion simulation. Healthy claws were fixed in the testing system mimicking aggressive abrasion against a standard concrete block (diamond patterned). In this setup, a vertical servohydraulic material-testing system was employed to apply a compressive load while, simultaneously, a horizontal electromechanical actuator pulled the concrete block beneath the claw. This configuration replicated shear loading conditions for testing purposes. After each loading cycle, the compressive load was released, thereby unloading the claw, and free weights pulled the concrete block back to the original position (as shown).



a compressive load while a horizontal electromechanical actuator (Heavy Duty Linear Actuator; Progressive Automations) pulled a surface (in this case, the standard concrete block) underneath the claw, thereby simulating shear loading. For each cycle of loading, the claw was subjected to compressive and shear loading for approximately 20 seconds. Then, the compressive load was released, thereby unloading the claw, and free weights pulled the concrete block back to the original position. In terms of loading conditions, a pilot trial conducted by a coauthor (DM) involving accelerometers affixed to the hind legs of beef steers showed that cattle may experience loads up to 5.5 times their bodyweight when agitated. Accordingly, using a conservative estimate of 3X bodyweight,<sup>14</sup> with 40% of the load supported by the hind limbs<sup>21</sup> and 80% of that load carried by the lateral claw,<sup>20</sup> a compressive load of 0.48X bodyweight was applied to each lateral claw ( $3 \times 0.4 \times 0.8 / 2 = 0.48$ ). For shear loading, static and dynamic coefficients of friction, estimated from load cells attached to the vertical and horizontal actuator, were 0.84 and 0.73, respectively.

Claws from the abrasion group ( $n = 8$ ) were marked with a permanent marker to monitor wear, then subjected to the abrasion simulation for 800 cycles. The surface of the concrete was cleaned every 200 cycles. The abrasion simulation took approximately 10 hours for each claw. Following the abrasion simulation, the claws experienced 1 to 3 mm of wear.

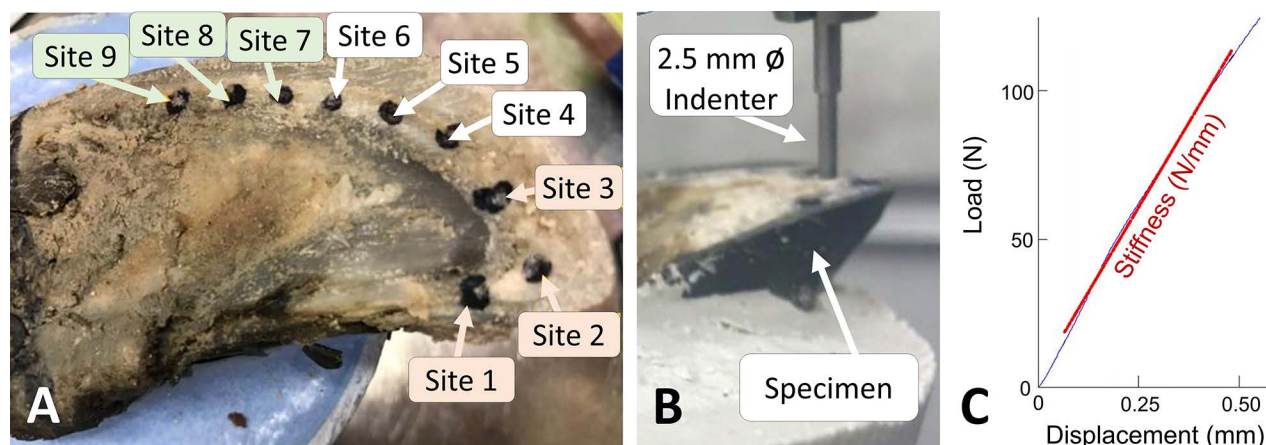
Control claws ( $n = 8$ ) were also marked with a permanent marker and then manually rasped by a single individual (NH) to mimic the wear pattern present with the matched abraded claws. A steel hand rasp was used to remove thin layers of material from the claw's surface until the same level of wear was achieved. This step was needed due to concerns that stiffness at the native, unrasped surface would be different than stiffness at the rasped surface, thereby influencing the comparison with abraded claws. Given that prior research indicated no difference in

white line separation between healthy (unrasped) claws and manually rasped claws,<sup>14</sup> we believe this approach is suitable for control purposes.

To verify that control claws exhibited the same level of wear as matched abraded claws, the thickness of the sole horn was assessed in a subset of the sample (abraded = 6; control = 6) via HR-pQCT (1st Generation XtremeCT; Scanco Medical AG) and commercial image-processing software (Analyze, version 14; Analyze Direct Inc; specific imaging details are discussed in more detail in the *Pathway determination* section below). The thickness of the sole horn was assessed via a line connecting the sole to the most anterior tip of P3, with the line being perpendicular to the sole. The distance between the sole and the dermis defined sole horn thickness. The thickness ranged from 1.8 to 4.4 mm, with no differences noted between the abraded and control specimens ( $P = .30$  via a Student  $t$  test).

#### Indentation stiffness testing of white line

Specimens (abraded = 8; control = 8) were subjected to mechanical indentation testing to determine the regional stiffness of the white line. Nine ( $n = 9$ ) equally spaced sites along the white line were marked with a permanent marker for indentation stiffness testing, with spacing being proportional to the size of the claw (**Figure 2**). Spacing was specifically set as the perimeter of the white line, measured from the apex (site 2) to the most peripheral aspect of the claw (site 9), divided by 7. The same spacing was used to set the position of site 1 relative to site 2. The final spacing between sites ranged from 6.5 to 9 mm, which met the minimum requirement of 3 mm spacing to ensure that indentation testing did not affect the mechanical properties of adjacent sites.<sup>22</sup> Sites were chosen based upon visual inspect of TTN specimens present within our laboratories, whereby sites 1 through 3 exhibited evidence of abrasion, whereas sites 7 through 9 exhibited no abrasion. Peripheral sites acted as internal validation



**Figure 2**—Marked indentation test sites along the white line, with spacing between adjacent sites being at least 3 mm. A—Sites 1 through 3 (orange) were along the apex of the claw, whereas sites 7 through 9 (green) were along the periphery. B—Indentation test setup, with the 2.5-mm-diameter flat indenter mounted to the material-testing system (not shown) for assessing stiffness. C—Plot of a typical load-displacement curve during indentation testing. The data within the linear region (red) were used to define the slope of the load-displacement curve for estimating stiffness.

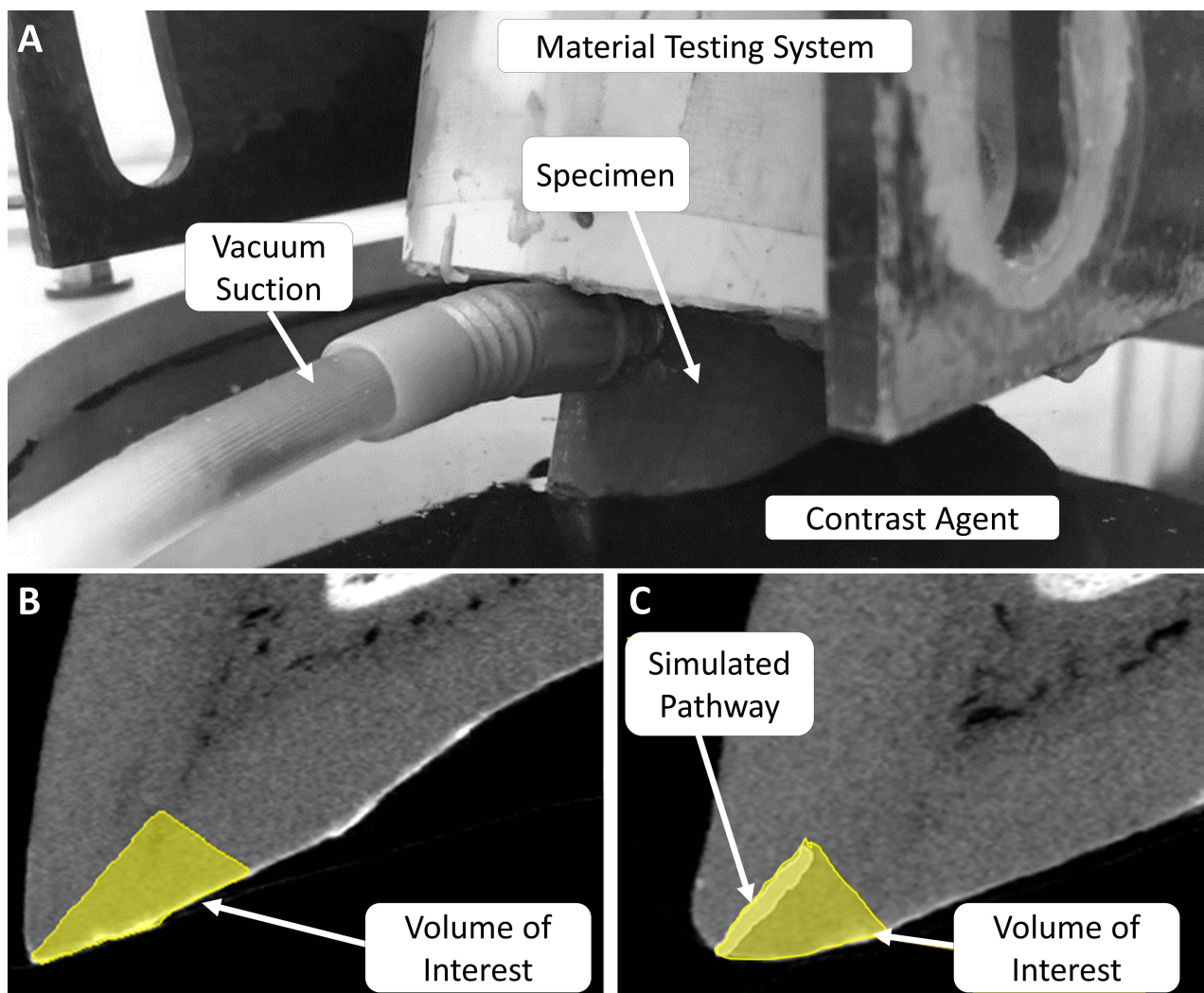
sites whereby similar peripheral stiffness between the groups would imply that the groups were reasonably matched, with observed differences at other sites being due to abrasion, not differences in physical size.

Before indentation testing, the potting system was reinforced with gypsum potting material (Denstone; Modern Materials Inc) to prevent any movement during the test. Approximately a 10-mm gap between the sole and the potting remained following reinforcement. Importantly, each claw was not fully emerged in potting material as that would overconstrain the specimen, leading to false, overestimated measures of stiffness.

Potted specimens were fixed within the servohydraulic material-testing system fitted with a 2.5-mm flat-shaped indenter used for assessing stiffness.

Specimens were aligned such that the claw surface was perpendicular to the axis of the indenter (Figure 2). Indentation testing was conducted at each of the 9 test sites, in random order, at 10 mm/min to a depth of 2 mm. The slope of the load-displacement curve in the linear region was used to characterize stiffness.

It is worthwhile noting that although elastic moduli can be estimated from measures of stiffness acquired via indentation testing (as per the Timoshenko-Goodier approach<sup>23,24</sup>), that approach is only valid for homogenous materials, whereas the white line and adjacent materials of the claw comprise a heterogenous material. Accordingly, the analysis was limited to measures of stiffness. It is also worthwhile noting that after stiffness testing, we attempted to determine the failure load and energy to



**Figure 3**—A—Suction test. Here, a vacuum fitting was affixed to the claw, after which the claw was reinserted into the material-testing system and immersed to a depth of 5 mm within an iodine-based contrast agent. To establish a vacuum, a vacuum pump was linked to the claw, which applied a suction at 80 kPa. The claw underwent cyclic compressive loading (ranging from 0.1 to 1 kN for 300 cycles at a frequency of 1 Hz) to simulate walking conditions. This methodology facilitated high-resolution peripheral quantitative CT (HR-pQCT) imaging of potential entry points developed during the abrasion simulation (where foreign materials could enter), wherein the contrast agent was drawn into these pathways. B—An example of a segmented volume of interest from an HR-pQCT scan of the apical toe. C—A pathway simulation on an HR-pQCT scan of a control claw.

**Table 1**—Mean and SD of regional stiffness measures from control claws (n = 8) versus abraded claws (n = 8) obtained at 9 different sites along the white line.

Site	Stiffness (N/mm)				P value
	Control claws		Abraded claws		
	Mean	SD	Mean	SD	
1	153.5	53.15	88.79	42.49	.018 <sup>a</sup>
2	189.9	64.83	99.24	49.19	.0071 <sup>a</sup>
3	148.9	60.75	83.50	33.57	.019 <sup>a</sup>
4	144.5	73.73	118.3	60.08	.45
5	116.6	57.31	114.9	46.18	.95
6	97.09	47.44	125.0	46.68	.25
7	110.9	43.63	103.6	44.45	.75
8	82.78	32.26	75.02	21.92	.58
9	68.41	27.33	54.05	13.04	.20

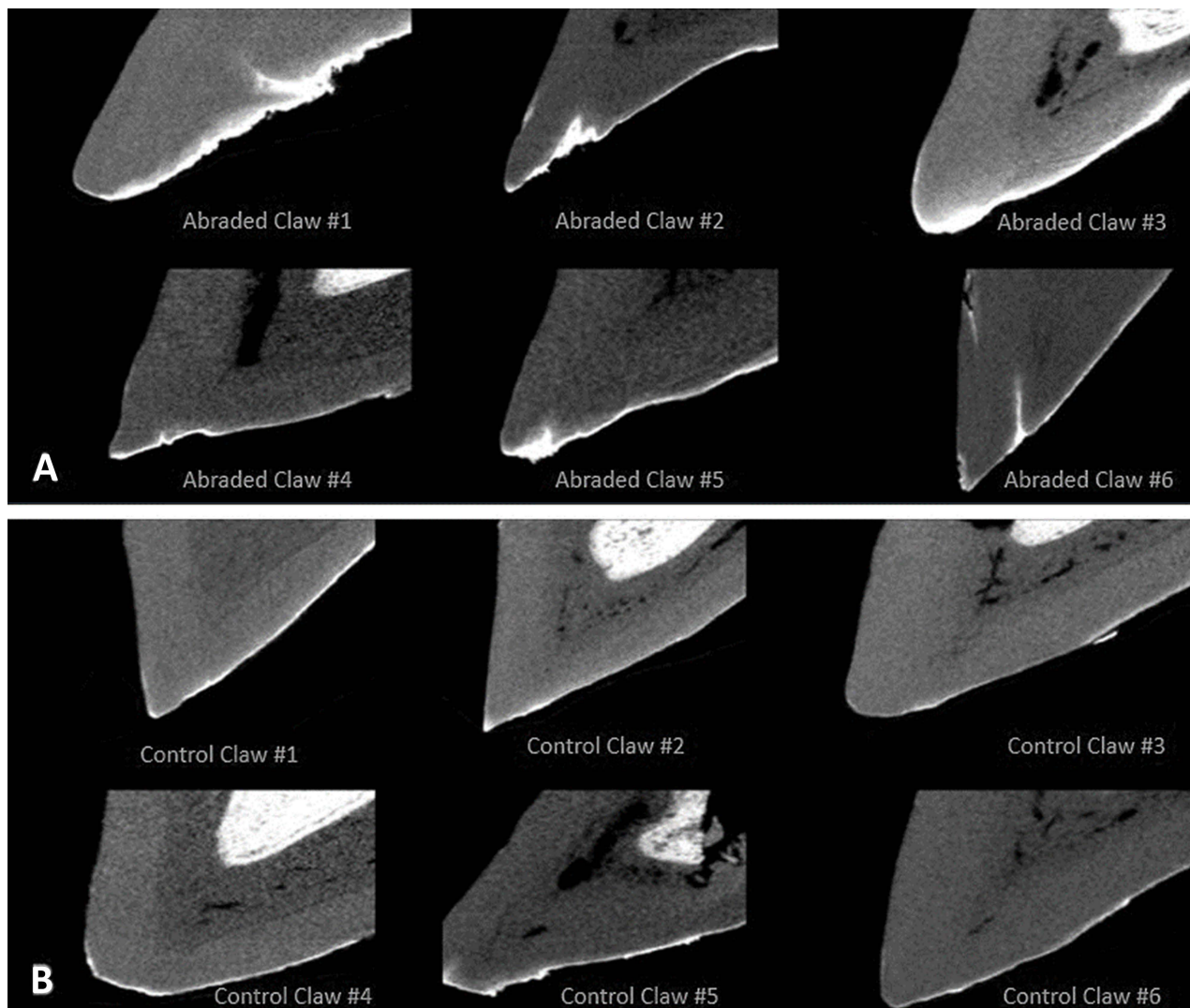
<sup>a</sup>Indicates a statistically significant difference.

Sites 1 through 3 were along the apex of the claw, whereas sites 7 through 9 were along the periphery. P values pertain to pairwise ANOVA comparisons of stiffness measurements.

failure leading to white line separation on 4 matched specimens (abraded = 2; control = 2). Unfortunately, the yield and failure points on the curve were not clear. These specimens were excluded from the following image analysis part of this research due to possible damage of the white line region. Failure testing was not performed on the remaining specimens (abraded = 6; control = 6).

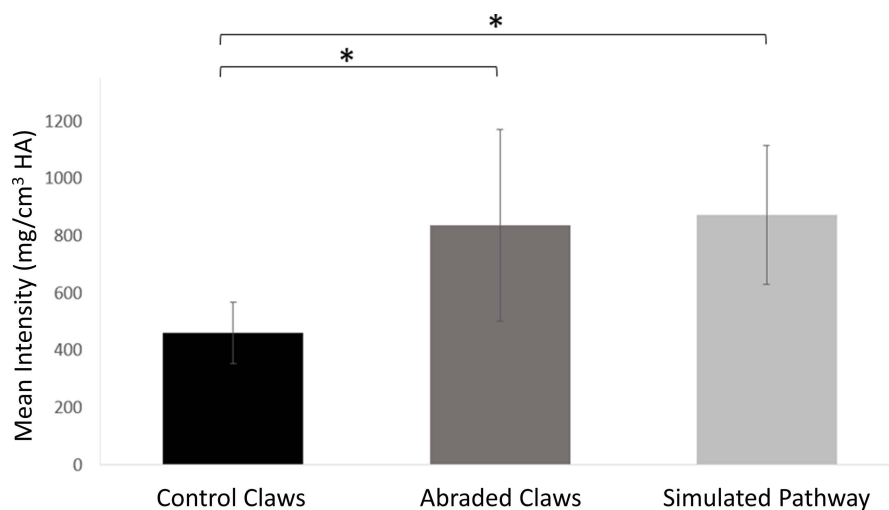
#### Pathway determination

Following mechanical testing, a subset of the sample (abraded = 6; control = 6) was assessed for pathway development. First, the extra gypsum potting material used for fixating the claws during indentation testing was removed, and a 10-mm diameter hole was drilled anterodorsally approximately 15 mm above the sole. A vacuum fitting was attached to the claw. The claw was then inserted back into the material-testing system and submerged to a depth of 5 mm in an iodine-based contrast agent (Cysto-Conray II;



**Figure 4**—A—Sagittal view from HR-pQCT scans of abraded claws with evidence of pathway development and local damage (characterized by high image intensity). B—High-resolution peripheral quantitative CT scans of sagittal view of control claws with minimal evidence of pathway development and local damage, apart from control claw #5.





**Figure 5**—Mean intensity of abraded claws, control claws, and simulated pathway. Error bars indicate SD for each group. \* $P < .05$ . HA = Hydroxyapatite.

Guerbet LLC). A vacuum pump (Schuco-Vac 130; Allied Healthcare) was connected to the claw, and 80 kPa suction was applied. The claw was then subjected to repetitive compressive loading (0.1 to 1 kN for 300 cycles at 1 Hz) simulating walking loads<sup>14</sup> (**Figure 3**). With this approach, potential pathways developed during the abrasion simulation (where foreign material could enter) would draw in the contrast agent, which could be visualized using HR-pQCT imaging. Following loading, specimens were placed back into the freezer and stored at  $-20^{\circ}\text{C}$  until undergoing HR-pQCT imaging.

Specimens were imaged using HR-pQCT with an isotropic voxel size of  $82\ \mu\text{m}$  across a 3.5-cm region of interest. This voxel size is the standard size used in HR-pQCT imaging and has been applied in previous clinical studies.<sup>25-27</sup> High-resolution peripheral quantitative CT image volumes were imported into commercial software (Analyze, version 14; Analyze Direct Inc) for image analysis. Images were first denoised using median filtering. The apex of the toe was then manually segmented via a touch-screen tablet (Cintiq 21UX; Wacom) in a region where damage is commonly found with TTN (**Figure 3**). The mean intensity (ie, luminance) for each segmented volume of interest was acquired, where a high mean image intensity indicated the presence of the contrast agent and possible pathway development.

Specimens with TTN within our laboratories have fully destroyed white line tissue. As a result, we were unable to include claws with TTN in this analysis as corresponding intensity measures would be excessively high as the volume of interest would primarily contain contrast agent. To overcome this limitation, we simulated a pathway on HR-pQCT images of control claws ( $n = 6$ ) that imitated patterns observed in claws with TTN using commercial software (Analyze, version 14; Analyze Direct Inc). A 0.8-mm-diameter pathway was simulated in the images of the control claws and filled with material having the same intensity as the contrast agent (**Figure 3**). The size of this pathway was estimated based upon the size of pathways observed in sectioned claws with TTN. The corresponding intensity of the volume of interest

represented the intensity of a claw with a definitive pathway for foreign material to enter.

### Statistical analysis

For comparing regional stiffness and mean image intensity, ANOVA was performed, followed by pairwise comparisons. The Fisher least significant difference procedure<sup>28</sup> was applied to account for multiple comparisons. All analyses were performed with commercially available statistical software (SPSS, version 22; IBM Corp). An  $\alpha$  level below 0.05 was considered statistically significant.

## Results

### Stiffness of white line

Analysis of variance indicated significant differences in stiffness at the apical region of the hooves (sites 1 through 3) between abraded claws and control claws ( $P < .019$ ; **Table 1**). No differences were noted any of the other sites ( $P > .199$ ).

### Pathway development and local damage

High-resolution peripheral quantitative CT images of abraded and control claws can be found in **Figure 4**. Analysis of variance indicated higher mean intensity in the abraded claws versus the control claws ( $P = .026$ ). Mean intensity of the abraded claws exhibited similar intensity as the simulated pathway reference ( $P = .83$ ; **Figure 5**).

## Discussion

The overall goal of this research was to investigate the effects of abrasion on white line separation at the toe and potential development of TTN. Our findings indicated that an abraded claw may be more damaged than a healthy claw as stiffness along the apical region was lower than that of control claws. Here, a lower stiffness may make the claws more vulnerable to local damage caused by gravel and rocks found in feedlots, possibly leading to white line separation and TTN. Also, analyses of HR-pQCT image data indicated that pathways for foreign material to enter the claw may be present following abrasion as

per image intensity mimicking a simulated pathway as well as higher intensity than the control claws. Altogether, these findings suggest that abrasion may be involved in white line separation and the pathogenesis of TTN.

In general, the findings of this study support a prevailing theory that mechanical loading is involved in the development of TTN.<sup>2,5,13</sup> Researchers have suggested that mechanical damage caused by abrasive surfaces found in feedlots, such as metal, concrete, and ice, leads to excessive wear, white line separation, and TTN.<sup>2,5,13</sup> Sick et al<sup>3</sup> were the first to correlate TTN with hyperexcitable cattle, a finding that has been corroborated in a well-documented outbreak of TTN in cattle sold through an auction in Western Canada.<sup>1</sup> A key component of the abrasion hypothesis is that fractious cattle will aggressively push on the animals ahead of them. In doing so, they generate large propulsive forces with their heavily muscled hind limbs. However, these forces eventually exceed the coefficient of friction, particularly when the floor is slick with urine and feces, and the loss of traction results in wearing of the apex of the hind toes on the concrete flooring. Thus, the inciting trauma that leads to TTN may occur within seconds when the cattle are scrambling to move ahead. This is exacerbated by poor cattle handling, which arises in part because the cattle handlers at the auction and feedlots are under deadlines to get cattle sorted and processed. It is noteworthy that new cases of TTN rarely develop after 30 days on feed, and there is no second peak of cases following reprocessing around day 80.<sup>2</sup> Therefore, TTN can be mitigated with better flooring, as has been demonstrated in the dairy industry, and improved cattle handling.

Mechanical damage risk factors are similar to those for thin soles and thin sole-induced toe ulcers of dairy cattle. Though, they are unlikely to be the same disease but in different cattle. The main risk factors for thin soles (sole horn < 5 mm in depth) in dairy cattle are exposure to abrasive flooring, such as concrete and mastic asphalt, and improper hoof trimming.<sup>29</sup> Furthermore, thin sole outbreaks in dairy cattle often coincide with an increase in sole, toe, and heel ulcers; white line disease; solar hemorrhage; and toe abscesses and necrosis.<sup>30,31</sup> Common environmental factors associated with these outbreaks include the feet being exposed to excessive moisture and concrete surfaces.<sup>30,31</sup> While it is tempting to equate thin sole-induced toe ulcers of dairy cattle to TTN of feedlot cattle, they present slightly differently.<sup>32</sup> In both instances, the infection can result in toe ulcers/abscesses, which may progress to P3 necrosis. However, thin sole-induced toe ulcers are usually preceded with solar hemorrhage, leading to separation of the sole away from the white line, followed by sub-solar abscess formation.<sup>32,33</sup> In contrast, TTN begins with separation along the apical white line. Although TTN of feedlot cattle and thin soles and thin sole-induced toe ulcers of dairy cattle share some risk factors, there are also important differences. Thin soles of dairy cattle are related to the cattle walking long distances on abrasive floors, such as concrete. Thus,

wear occurs continuously on a daily basis. In contrast, feedlot cattle are only exposed to abrasive surfaces for relatively short periods of time. Briefly, beef cattle are removed from pastures in the fall of the year and transported either directly to feedlots or sold through auctions. All auctions use concrete flooring, and the cattle are handled extensively as they are commonly led, sorted, and penned. Their time at the auction generally varies from 1 to 3 days before they are transported to feedlots. Upon arrival to the feedlots, they may be sorted again before being processed and then placed in earthen-floor pens for the duration of the feeding (fattening) period. Feedlots, like auctions, use concrete for the flooring in holding pens and alleyways and either concrete or metal in the chute systems. Whether the cattle are sold through an auction or directly into the feedlot, their time in contact with concrete flooring prior to developing TTN can be measured in hours to days versus weeks to months for dairy cattle with thin soles and thin sole-induced toe ulcers. There are, however, 2 important risk factors that are unique to TTN of feedlot cattle, namely cattle temperament and handling.

The results of our research differ from the previous static loading and fatigue study by Johnston et al,<sup>8</sup> where mechanical loading alone did not result in separation of the white line. The reason for this discrepancy may be that their study was solely limited to compressive loading and ignored the effects of horizontal shear loading present during physiologic activities.

This study has specific strengths and limitations that require consideration. With regard to strengths, first, we simulated an *in vivo* abrasion condition that models physiological loading more closely than pure compressive loading. Second, specimens were matched in terms of physical size and bodyweight, which helps to account for the confounding size factor influencing statistical comparisons (ie, differences in stiffness of the white line may be due to differences in physical size, not abrasion). Third, a multifaceted approach was used to evaluate the role of abrasion on white line separation, including stiffness testing as well as high-resolution, contrast-enhanced imaging. With regard to limitations, first, due to excess damage, we were not able to derive a representative estimate of white line stiffness for claws with TTN. The reason was that the white line region of claws with TTN was largely damaged, rendering stiffness results unreliable. Second, the claws with TTN were at an advanced stage of the syndrome. Corresponding intensity measures would then be excessively high as the volume of interest would primarily contain contrast agent (as per our previous research<sup>8</sup>). To address this issue, we simulated initial pathway development in control claws. The authors acknowledge, though, that this approach is not necessarily representative of the TTN condition. Third, the findings of this study are based on *in vitro* mechanical testing, which may not fully represent the complex and dynamic conditions of animal walking or other agitated behaviors *in vivo*. Future research will aim to test white line stiffness and pathway development of a large sample of claws at early stages of TTN.



In conclusion, our findings indicate that abraded claws exhibited lower stiffness along the apical region of the white line relative to control claws. Also, analyses of HR-pQCT image data suggested that pathways for foreign material to enter the claw may be present following abrasion as indicated by image intensity mimicking a simulated pathway as well as higher intensity than the control claws. These findings are consistent with the mechanical damage hypothesis. Although speculative, these results suggest that TTN may be initiated when cattle abrade their hooves on abrasive surfaces found in feedlots due to anxiety and aggressive behavior. The damage arising from this activity leads to a weakened structure vulnerable to damage caused by gravel and rocks found in feedlots, possibly leading to white line separation and TTN. Findings of this study point to the importance of floorings on TTN (eg, alternative floorings that minimize abrasion, such as rubber, may be beneficial for limiting TTN).

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The authors have nothing to disclose. No AI-assisted technologies were used in the composition of this manuscript.

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### Supplementary Materials

Supplementary materials are posted online at the journal website: [avmajournals.avma.org](http://avmajournals.avma.org).