Cartilage-derived biomarkers and lipid mediators of inflammation in horses with osteochondritis dissecans of the distal intermediate ridge of the tibia

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Objective—To assess whether reported alterations in metabolism of cartilage matrix in young (0 to 24 months old) horses with osteochondritis dissecans (OCD) may also be found in older (24 to 48 months old) horses with clinical signs of OCD and to investigate the role of eicosanoids in initiating these clinical signs.

Sample Population—Synovial fluid was collected from 38 tarsocrural joints of 24 warmblood horses with (22 joints of 16 horses) or without (16 joints of 8 horses) clinical signs and a radiographic diagnosis of OCD of the distal intermediate ridge of the tibia.

Procedures—Turnover of type II collagen was investigated by use of specific immunoassays for synthesis (carboxypropeptide of type II collagen [CPII]) and degradation (collagenase-cleaved fragments of type II collagen [C2C]) products. Furthermore, glycosaminoglycan (GAG), leukotriene (LT) B4, cysteinyl LTs, and prostaglandin (PG) E2 concentrations were determined, and concentrations in joints with OCD were compared with those in joints without OCD.

Results—Concentrations of CPII, C2C, and GAG did not differ significantly between affected and nonaffected joints. Fluid from joints with OCD had significantly higher LTB4 and PGE2 concentrations than did fluids from nonaffected joints.

Conclusions and Clinical Relevance—Altered collagen or proteoglycan turnover was not detected in 24- to 48-month-old horses at the time they developed clinical signs of OCD of the distal intermediate ridge of the tibia. However, increased concentrations of LTB4 and PGE2 in fluid of joints with OCD implicate these mediators in the initiation of clinical signs of OCD. (Am J Vet Res 2006;67:1156–1162)

Osteochondrosis, which is also termed OCD when there are loose osteochondral fragments, is a major developmental joint disease in horses and can be defined as a disruption of the endochondral ossification process in the epiphyseal cartilage. Because this process is typically only active during the first year after birth, osteochondrotic lesions, by definition, will only develop during this period. The tarsocrural joint is one of the joints most commonly affected by osteochondrosis or OCD in warmblood horses,1,2 and lesions at this location often do not clinically manifest until later in life when training commences and young horses are first subjected to athletic challenges.3

In the tarsocrural joint, lesions characteristically develop at the cranial end of the intermediate ridge of the tibia as well as at the lateral trochlear ridge of the talus.4 In longitudinal experimental studies5,a conducted by our research group, the radiographic appearance of tarsocrural OCD lesions was highly dynamic in foals between 1 and 5 months of age, after which lesions remained permanent and radiographically stable until foals were 24 months old.

The underlying molecular changes in osteochondrotic cartilage have been the subject of many investigations. Studies5,13 involving markers of cartilage turnover in synovial fluid or serum have revealed significant differences in metabolism of collagen and proteoglycan between OCD-affected foals and healthy, age-matched control foals. Evidence5,13 points toward an early involvement of the collagen network in the pathogenesis of lesions (ie, during the first year after birth). However, the eventual clinical expression of OCD is believed to be the outcome of 2 consecutive events: the initial onset of lesions and the ensuing repair process.5,2 Little is known about cartilage metabolism in established OCD lesions after horses exceed 1 year of age. The fact that tarsocrural OCD lesions stabilize radiographically after horses are 3 months old, whereas clinical disease often does not become apparent until after horses are 24 months old, raises the issue as to whether affected joints may remain metabolically aberrant despite being radiographically stable and whether such metabolic aberrations may contribute to clinical manifestation of lesions.

Another poorly understood aspect of clinical expression of tarsocrural OCD is the pathophysiologic...
background of its most common signs (ie, prominent joint effusion with minimal disturbance of motion).24 Synovial membranes release vasoactive mediators, notably E-series PGs, when irritated by cartilage detritus and osteochondral fragments within the joint cavity.25,27 and this may partially account for the effusion. However, PGs are also believed to be intimately linked with joint pain and lameness in dogs and horses.25,26 Because lameness is an inconsistent finding in horses with OCD of the distal intermediate ridge of the tibia,24 we hypothesized that in clinically affected animals with joint effusion but no notable lameness, other inflammatory mediators may dominate to initiate the clinical signs. The LTs, which are also members of the eicosanoid family of inflammatory mediators, are potent vasoactive mediators that may be released by neutrophils and resident articular cells, such as synoviocytes, chondrocytes, and osteoblasts.22,23 but bear no direct relation to joint nociception. Leukotriene B4 and CysLTs are prime mediators of plasma extravasation and joint swelling in the stifles joints of guinea pigs22 and rats.23 So far, little is known about the involvement of the various LTs in joint disorders of horses, although LTB4 was considered a good indicator of experimentally induced synovitis in 1 study.25

In the study reported here, the underlying molecular mechanisms governing clinical signs of disease were examined in 24- to 48-month-old horses with OCD of the distal intermediate ridge of the tibia. The study was intended to serve 2 purposes. First, we investigated whether changes in metabolism of articular cartilage matrix, which have been reported in other studies8-13 in OCD-affected foals during the first year after birth, are also detectable in 24- to 48-month-old horses by the time that clinical signs become evident. Second, we tested the hypothesis that LTs (rather than PGs) may be involved in horses with joint effusion without associated lameness, which is typical of horses with OCD of the distal intermediate ridge of the tibia.

Materials and Methods

Sample population—Synovial fluid was collected from 38 tarsocural joints of 24 warmblood horses. Samples of synovial fluid were obtained from 16 horses with joint effusion and OCD of the distal intermediate ridge of the tibia and from 8 clinically normal control horses. The study was approved by the Utrecht University Ethical Committee.

Samples from joints with OCD were obtained from 22 joints of 16 client-owned warmblood horses evaluated to determine the cause of prominent joint effusion of 1 or both tarsocural joints. Because these were samples that would otherwise have been discarded during surgery, informed consent of the owners was not obtained. All selected horses were between 24 and 48 months of age at the time of examination. All horses were subjected to a full lameness examination, but lameness was not detected in any horse. Diagnosis of OCD of the tarsocural joint was confirmed by standard radiographic examination. Only fluids from joints in which the OCD lesion was limited to the distal intermediate ridge of the tibia and that had no additional signs of degenerative changes at arthroscopy were included in the study.

Eight control horses, maintained as part of a university research herd, were matched on the basis of age with the OCD-affected horses. Each control horse underwent a full clinical lameness examination, and all were deemed free of musculoskeletal disease. Tarsocural joints of control horses were considered free of OCD on the basis of examination of 3 standard radiographic views (dorsosomedial-plantarolateral oblique, lateral-medial, and craniol-caudal) of each tarsocural joint.

Sample collection—Horses with OCD of the distal intermediate ridge of the tibia were admitted for arthroscopic removal of fragments. Before surgical exploration of a joint, synovial fluid was aspirated into a sterile 20-mL syringe. Within 1 hour after collection, part of the sample was submitted for a cell count, whereas the remainder was transferred to polypropylene tubes and centrifuged at 10,000 g for 10 minutes. Synovial fluid was then divided into aliquots and stored at –80°C until further analysis.

For control horses, aseptic arthrocentesis of both tarsocural joints was performed via a dorsomedial approach by use of an 18-gauge, 1.5-inch needle connected to a sterile 20-mL sterile syringe. When needed, a nose twitch or mild sedation achieved by the administration of detomidine (0.01 mg/kg, IV) were used to ensure proper immobilization of horses during the procedure. Processing and storage of synovial fluid were conducted in accordance with the same protocol described for the samples from joints with OCD.

CPII assay—A commercially available competitive ELISA’ for CPII was used. In the course of fibril formation, CPII is released into synovial fluid, where its concentration reflects synthesis of type II collagen.2 The assay included a bovine CPII standard and rabbit polyclonal capture antibody. Although this antibody was raised against human CPII, studies22,27 have established cross-reactivity as well as parallelism and effective spike recovery in samples obtained from horses. A preliminary study was conducted to assess dilution and digestion of samples for optimal assay performance. The preliminary study revealed that digestion of synovial fluid with testicular hyaluronidase (0.05 mg/mL) for 30 minutes at 37°C, followed by a 1:1 dilution of the digested synovial fluid, resulted in values within the optimal concentration range for the assay (50 to 2,000 ng/mL). Absorbance was measured at 450 nm.

C2C assay—A commercially available competitive ELISA’ was used for the detection of C2C. This assay included a mouse primary C2C antibody (formerly referred to as col2-3/4Clong) that detects the neoeptope located at the C terminus of the third-fourth length cleavage product of type II collagen fibrils. This antibody was raised against a synthetic peptide representing the neoeptope (used as the assay standard) and can successfully detect cleavage fragments carrying this neoeptope in synovial fluid obtained from dogs and humans.26,28 Because the structure of collagen type II and collagenase cleavage sites are highly conserved,30,31 there was ample reason to assume that the assay would be cross-reactive in samples obtained from horses.

Proper assay performance was confirmed in a preliminary study of equine synovial fluid. Linearity was assessed by assay of serial dilution of samples with known concentrations of C2C, which yielded a correlation coefficient of 0.99 between the measured and expected concentration at each dilution. In addition, effective spike recovery in synovial fluid was observed (mean recovery, 95.3% range, 92.6% to 105.8%). This preliminary study further revealed that after digestion with testicular hyaluronidase (0.05 mg/mL) for 30 minutes at 37°C, no dilution of synovial fluid samples was necessary to yield values within the dynamic range of the assay (1 μg/mL to 10 ng/mL). Absorbance was measured at 450 nm.

GAG assay—Proteoglycan content of synovial fluid was estimated by measuring sulfated GAG concentrations by use of the 1,9-dimethylbenzene blue metachromatic dye
assay, which was modified for use in microtiter plates. Eighty microliters of buffered hyaluronidase (500 μg of testicular hyaluronidase/mL in 50 mM sodium acetate [pH, 5.2]) was added to 20 μL of synovial fluid; the mixture was allowed to digest for 30 minutes at 37°C. Then, 40 μL of the mixture was transferred to a well of a microtiter plate, and 200 μL of Farndale reagent (464 μM 1.9-dimethylmethylenedioxyl blue, 40 mM glycine, and 42 mM NaCl, adjusted to pH 3.0 by the addition of hydrogen chloride) was added. The plate was incubated for 15 minutes, and absorbance was then measured at 525 nm. Shark cartilage chondroitin sulfate was used as the assay standard.

Eicosanoid immunoassays—Commercial competitive ELISA kits were used to determine synovial fluid concentrations of LTB4, CysLTs1, and PGE2. Each kit was used in accordance with the manufacturer’s instructions. The kits used rabbit polyclonal antibodies raised against the synthetic eicosanoids. Although these kits were intended for use in human samples, there are no species differences in the structure of arachidonic acid derivatives among species, so the assays can also be applied to samples obtained from horses.3 Samples were extracted before analysis by use of the immunoassays. In brief, 200 μL of synovial fluid and 10 μL of testicular hyaluronidase (10 mg/mL) were allowed to digest for 30 minutes at 37°C. Then, 100 μL of 0.1% formic acid was added, and the solution was vortexed, after which samples were centrifuged for 10 minutes at 13,000 × g. Supernatant was harvested and applied to an RP-18 extraction column that had been conditioned with 1 mL of acetone followed by 1 mL of water. The column was then flushed with 1 mL of water, 1 mL of 5% ethanol, and 2 applications of hexane (1 mL of each application). Eicosanoids were eluted from the column by use of 500 μL of ethylacetate, dried under a stream of nitrogen gas, and stored under nitrogen at –80°C until analysis.

All samples were analyzed within 1 week after sample extraction. At the time of assay, samples were reconstituted by the addition of 250 μL of assay buffer and analyzed in accordance with the manufacturer’s instructions. Absorbance was measured at 405 nm, with a reference wavelength of 595 nm. Absorbance values were assumed in statistical testing (Welch correction). Computer software was used, and a value of P < 0.05 was considered significant for all statistical analyses.

Results

Animals—Mean age of horses for the OCD and control (16 joints) groups. Data were assessed for normality of distribution, and differences in synovial fluid variables between groups were tested by use of an unpaired t test. When data from the 2 groups failed the Levene test for equality of variance, unequal variances were assumed in statistical testing (Welch correction). Computer software was used, and a value of P < 0.05 was considered significant for all statistical analyses.

Analysis of synovial fluid—All data for the variables were normally distributed. Hence, no data transformations were performed.

WBC counts—Mean ± SEM WBC counts did not differ significantly between synovial fluid obtained from joints with OCD (0.73 × 109 ± 0.03 × 109 cells/L) and normal joints (0.71 × 109 ± 0.05 × 109 cells/L; Table 1).

Markers of metabolism of cartilage matrix—No significant differences in CPII (type II collagen synthesis marker), C2C (type II collagen degradation marker), and GAG (estimate of proteoglycan degradation) concentrations were found between synovial fluid obtained from OCD-affected joints and normal tarsocrural joints (Table 1).

Table 1—Mean ± SEM concentrations of cartilage-derived markers and eicosanoids in synovial fluid obtained from 22 OCD-affected and 16 normal (control) tarsocrural joints of horses.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control joints</th>
<th>OCD-affected joints</th>
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<tbody>
<tr>
<td>CPII (μg/mL)</td>
<td>528.0 ± 29.5</td>
<td>740.2 ± 130.5</td>
</tr>
<tr>
<td>C2C (μg/mL)</td>
<td>170.9 ± 8.5</td>
<td>192.8 ± 10.2</td>
</tr>
<tr>
<td>GAG (μg/mL)</td>
<td>13.9 ± 2.1</td>
<td>12.8 ± 1.5</td>
</tr>
<tr>
<td>CysLT (pg/mL)</td>
<td>416.5 ± 38.3</td>
<td>400.5 ± 20.6</td>
</tr>
<tr>
<td>LTB4 (pg/mL)</td>
<td>538.9 ± 35.8</td>
<td>534.5 ± 30.0</td>
</tr>
<tr>
<td>PGE2 (pg/mL)</td>
<td>127.3 ± 33.6</td>
<td>362.3 ± 93.6</td>
</tr>
<tr>
<td>WBCs (× 109 cells/L)</td>
<td>0.71 ± 0.05</td>
<td>0.73 ± 0.03</td>
</tr>
</tbody>
</table>

*Means differed significantly (P < 0.001) between groups. †Means and variances differed significantly (means, P < 0.001; variances, P < 0.05) between groups.

Figure 1—Mean ± SEM LTB4 concentrations in synovial fluid obtained from 22 OCD-affected and 16 normal (control) tarsocrural joints of horses. The mean value differed significantly (P < 0.001) between the groups.

Figure 2—Mean ± SEM PGE2 concentrations in synovial fluid obtained from 22 OCD-affected and 16 normal (control) tarsocrural joints of horses. The mean value differed significantly (P < 0.001) between the groups.
Eicosanoid concentrations—A significant ($P < 0.001$) increase in the LTB4 concentration was found in synovial fluid obtained from OCD-affected joints, compared with concentrations in synovial fluid obtained from control joints of age-matched horses (Figure 1). Mean ± SEM value for the OCD-affected horses (934.5 ± 60.0 pg/mL) was nearly twice that for the control horses (568.9 ± 33.9 pg/mL). Similarly, mean PGE$_2$ concentration for the OCD-affected horses (362.3 ± 93.6 pg/mL) was significantly higher than the concentration for the control horses (127.3 ± 33.6 pg/mL; Figure 2). Concentrations of LTB4 and PGE$_2$ had significantly ($P = 0.005$ and $P < 0.001$, respectively) greater variance for the OCD-affected horses than for the control horses. Concentrations of CysLTs were detectable in all synovial fluid samples, but we did not detect significant differences between mean concentrations for OCD-affected (400.4 ± 20.6 pg/mL) and control (416.9 ± 38.5 pg/mL) joints.

Discussion

Osteochondrosis and OCD affect the young of numerous species, including humans, and the underlying molecular mechanisms of OCD have been the subject of many scientific investigations. Alterations in turnover of articular cartilage matrix have been reported for foals with OCD, and these changes have been associated with pathogenesis of the disease condition.

It has been asserted that because lesions only develop within the first year after birth, studies of OCD in older animals may reflect only the secondary reparative processes. However, evidence has revealed that most radiographic lesions in foals heal uneventfully within the first year after birth. Hence, it would be of value to know whether cartilage metabolism in horses that do not have proper healing of defects and subsequently develop clinical disease differ from those of healthy age-matched control horses.

Alterations in the collagen network and, in particular, changes in metabolism of type II collagen have been implicated in the OCD disease process. In the young horses reported here, we found no differences in synthesis of type II collagen, on the basis of synovial fluid concentrations of CPII, between OCD-affected and normal joints. Absolute concentrations of CPII in the synovial fluid reported here are 10-fold higher than the control horses (568.9 ± 33.9 pg/mL). Similarly, mean PGE$_2$ concentration for the OCD-affected horses (362.3 ± 93.6 pg/mL) was significantly higher than the concentration for the control horses (127.3 ± 33.6 pg/mL; Figure 2). Concentrations of LTB4 and PGE$_2$ had significantly ($P = 0.005$ and $P < 0.001$, respectively) greater variance for the OCD-affected horses than for the control horses. Concentrations of CysLTs were detectable in all synovial fluid samples, but we did not detect significant differences between mean concentrations for OCD-affected (400.4 ± 20.6 pg/mL) and control (416.9 ± 38.5 pg/mL) joints.

Lack of alterations in concentrations of markers of collagen metabolism, synovial fluid concentrations of GAG in the study reported here were independent of OCD status of the joint from which the sample was collected and corresponded closely with concentrations for normal tarsocural joints obtained in other studies. There are contradictory results with regard to aberrations in proteoglycan metabolism in osteochondrotic horses. Investigators in 1 study found low GAG contents and altered proteoglycan composition of OCD-affected cartilage of the distal intermediate ridge of the tibia, and investigators in another study in vivo study found significant increases in synovial fluid concentrations of GAGs in OCD-affected horses. In 2 other studies, investigators found no difference in GAG release from osteochondrotic or normal cartilage explants. Because the aforementioned in vivo study does not provide information about the age of the horses or whether there were degenerative changes in the osteochondrotic joints, it is hard to compare results from that study and the study reported here. Investigators in yet another study found a decrease in synthesis of proteoglycans in 9- to 18-month-old horses with OCD of the tarsocural joint but not in 24- to 48-month-old horses with OCD of the tarsocural joint, which suggests that this aspect of proteoglycan metabolism was no longer aberrant by the time horses were > 2 years old, and this is in agreement with data for proteoglycan degradation reported here.

Analysis of our results for concentrations of metabolic markers in synovial fluid points toward a stabilization in turnover of type II collagen as well as proteoglycan degradation in OCD-affected horses at 29 to 48 months of age. This supports the idea that OCD lesions in the tarsocural joint become both radiographically and metabolically stable over time.

Lack of alterations in concentrations of markers of cartilage matrix metabolism at the time of clinical manifestation reinforces the idea that other factors are involved, with synovitis appearing to be a likely candidate. However, synovial inflammation as a cause of
clinical signs of OCD has received relatively little attention from researchers. In 1 study on the involvement of PGE\(_2\) in joint disease of horses, the authors included a subgroup of 8 horses with OCD. In that study, no increase in PGE\(_2\) concentrations was detected in synovial fluid from OCD-affected joints, compared with concentrations in normal joints. The authors conjectured that because OCD is commonly associated with minimal lameness, it could be expected that PGE\(_2\) concentrations would be low in OCD-affected joints. Unfortunately, no information was supplied in their report on the type of joints from which samples were obtained or the exact location of lesions within affected joints. The study reported here involved 22 narrowly defined clinical cases of OCD of the distal intermediate ridge of the tibia. We detected a significant increase in PGE\(_2\) concentration in synovial fluid obtained from OCD-affected joints, despite the fact that none of the horses were lame. In accordance with our findings, investigators in another study also detected higher concentrations of PGE\(_2\) in synovial fluid collected from all 7 horses affected by OCD in their study; however, they partially explained this by reference to the more clinically active nature of the disease in their horses, all of which were lame at the time of sample collection.

Our finding of increased PGE\(_2\) concentrations in synovial fluid of OCD-affected joints despite the fact that horses were not lame raises questions about the supposed intimate link between joint pain and concentrations of PGE\(_2\) in synovial fluid. Although the high PGE\(_2\) concentrations in OCD-affected joints are perhaps unexpected with regard to the lack of lameness, they are not illogical. Bone fragments cause release of PGE\(_2\) from equine synoviocytes, and PGE\(_2\) can influence the diameter and permeability of synovial vessels, thus contributing to joint effusion. Because joint effusion was the primary clinical sign in the horses of our report, it may have been expected that we would detect increased PGE\(_2\) concentrations.

The actions of LTs, another member of the eicosanoid group of inflammatory mediators, have been relatively overlooked in joint disease. Synoviocytes and subchondral osteoblasts are both capable of production and release of LTs in response to various stimuli, whereas chondrocytes are believed to release LTs following interactions with granulocytes. Increased concentrations of LT\(_B_4\) in synovial fluid have been associated with arthritic conditions and can be significantly correlated with severity of synovitis in equids. In the study reported here, a significant (\(P < 0.001\)) increase in LT\(_B_4\) concentrations was found in synovial fluid obtained from osteochondrotic joints, compared with concentrations in control joints.

Although increases in LT\(_B_4\) concentrations in synovial fluid reportedly correlate with increases in WBCs, this was not the situation in our study. The WBC counts were comparably low for both the OCD-affected and control joints. It is hard to explain the reason that increases in LT\(_B_4\) concentrations in synovial fluid were not accompanied by increased infiltration by polymorphonuclear neutrophils in these joints. Possibly, the powerful chemotactic actions of LT\(_B_4\) in joints with infectious, rheumatoid, or chemically induced arthritis (conditions in which many more mediators are liberated that may contribute to leukocyte attraction into the joint) may not be as pronounced in the relatively calm OCD-affected joints. Alternatively, the LT\(_B_4\) concentrations found in our study may have been too low to cause a substantial influx of leukocytes into the joint cavity, given that picomolar or nanomolar concentrations of LT\(_B_4\) are needed in humans to induce an influx, and equine leukocytes seem less responsive to LT\(_B_4\) than their human counterparts. Even so, an in vivo study of humans with rheumatoid arthritis also did not yield consistent correlations between LT\(_B_4\) concentrations and WBC counts, despite the known chemotactic properties of LT\(_B_4\) and leukocyte involvement in this disease.

Because leukocytes are apparently not the major source of LT\(_B_4\) in OCD, 2 other likely candidates remain (ie, the synovial membrane and subchondral bone). The synovial membrane is likely to be irritated by cartilage and bone detritus in animals with OCD, and synoviocytes are believed to be major contributors to eicosanoid concentrations in synovial fluid. Interestingly, osteoblasts in subchondral bone can also be apt producers of LT\(_B_4\) in vivo, and in the subchondral bone compartment, LT\(_B_4\) is an important regulator of the extracellular matrix. Because in osteochondrosis, especially the OCD form, the subchondral bone is exposed and may communicate with the synovial cavity, we speculate that the increased concentrations of LT\(_B_4\) in synovial fluid may partially originate from subchondral bone. The finding of increased LT\(_B_4\) concentrations in osteochondrotic joints of older horses raises the issue of whether LT\(_B_4\) is also involved in the earlier stages of the disease. In this respect, it is interesting to mention that LT\(_B_4\) can stimulate osteoclast differentiation and bone resorption and affect bone remodeling during endochondral ossification. Moreover, LT\(_B_4\) can cause upregulation of cartilage catabolic factors, such as matrix metalloproteinases, and could thus hypothetically play a role in the disturbance of normal chondrocyte maturation in animals with OCD. Additional investigations into the involvement of LT\(_B_4\) in osteochondrosis in horses, especially young horses in which lesions are developing, would seem warranted.

To our knowledge, the study reported here is the first to provide concentrations of CysLTs in synovial fluids obtained from horses. Although CysLTs were detectable in substantial amounts in samples from all osteochondrotic joints, there were no significant differences from concentrations detected in control joints. Hence, despite the fact that CysLTs are involved in synovial effusion in rats, it appears that they are unlikely to be involved in the synovial effusion seen in horses with OCD.

The usual caveats associated with analysis of mediator or marker concentrations in synovial fluid should be considered when interpreting results from the study reported here. Most importantly, the difficulty in comparing concentrations of analytes between joints with and without prominent joint effusion should be appre-
diated. In addition, concentrations of metabolic markers only allow indirect investigation of tissue metabolism. Many factors other than those involved with actual rate of synthesis or degradation of matrix components may affect synovial clearance rates and thus marker concentrations in synovial fluid. Among these, the amount of active inflammation may be an important determinant, which could not be corrected for.

Analysis of our findings supports the idea that alterations in cartilage metabolism cannot account for the observed clinical signs in horses between 24 and 48 months of age with OCD of the distal intermediate ridge of the tibia. This study extends the findings from radiographic studies, which revealed that tarsocrural OCD lesions remained radiographically stable in affected horses from 5 to 24 months of age. We can conclude that in addition to being radiographically stable, these lesions also appear to be metabolically stable. Furthermore, we established the local involvement of various eicosanoids in joints of horses with OCD of the distal intermediate ridge of the tibia and found that PGE₂ and LTB₄, but not CysLTs, are likely to play a role in the prominent joint effusion that is the most common clinical sign.

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