

DISSERTATION

PRESALE DIAGNOSTIC IMAGING IN THOROUGHBRED HORSES:  
THE PREVALENCE AND PROGRESSION OF RADIOLOGICAL AND ULTRASONOGRAPHICAL  
FINDINGS AND THEIR ASSOCIATIONS WITH RACING PERFORMANCE

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## ABSTRACT

### PRESALE DIAGNOSTIC IMAGING IN THOROUGHBRED HORSES: THE PREVALENCE AND PROGRESSION OF RADIOLOGICAL AND ULTRASONOGRAPHICAL FINDINGS AND THEIR ASSOCIATIONS WITH RACING PERFORMANCE

#### *Background*

The sale of young horses at bloodstock auctions plays an important role in the success of the Thoroughbred industry worldwide. The Keeneland September Yearling Sale in Lexington, Kentucky, is the largest sale of Thoroughbred yearlings in the world. Conducted over 12 days and presenting between 2500 and 3000 horses through the auction ring, the sale now records gross receipts exceeding 400 million United States Dollars (USD) per year. Veterinarians at the sale perform presale inspections on future racing prospects and consult with prospective purchasers regarding a horse's suitability for its intended use. Radiography and ultrasonography are used in presale examinations to identify orthopaedic issues that may affect soundness during athletic training and racing. Modern diagnostic imaging technology produces high quality images that have enabled the detection of a number of presale findings of unknown significance in young horses. Variable interpretation of these findings and conflicting assignments of potential clinical importance have become a source of contention in the Thoroughbred industry and this requires resolution.

Specific imaging findings for which further scientific evidence is needed include radiological changes in the equine medial femoral condyles (MFCs) and proximal sesamoid bones (sesamoids) and ultrasonographic findings in the medial and lateral branches of the suspensory ligament (branches). Changes in the sesamoid and the insertional region of the adjacent suspensory branch are of particular importance in young Thoroughbreds, due to the potential for catastrophic injury to the suspensory apparatus in which they are an integral structure. Little is known about the prevalence of concurrent ultrasonographic branch change relative to the various grades of radiological sesamoid appearance seen in horses. Objective

scientific data would enable prepurchase and training management decisions to be made that are in the best interests of the horse and its connections and the wider industry.

### *Objectives*

In the equine stifle (femorotibial joint) and fetlock (metacarpo/metatarsophalangeal joint), the objectives of this doctoral research were firstly to identify the prevalence of subchondral lucencies (SCLs) in the distal aspect of the MFC and the prevalence of various sesamoid changes on sales repository radiographs in yearling and 2-year-old Thoroughbreds, and to identify the prevalence of ultrasonographic suspensory branch changes in the same population of horses. Secondly, the studies aimed to monitor changes in MFC, sesamoid and suspensory branch grades between yearling and 2-year-old sales in horses that presented for sale at both ages. Thirdly, the research aimed to determine any associations between grade of MFC, sesamoid or suspensory branch findings and future racing performance.

Lastly, the research was designed to examine the existence of concurrent radiological and ultrasonographic findings in individual sesamoid-branch units in sales horses; to determine whether there are any radiological findings that are consistently accompanied by a particular degree of insertional branch change and to provide practical recommendations as to when suspensory branch ultrasonography may be warranted in the sales environment.

The overriding objective was to provide an evidence-based determination of which presale imaging findings should be regarded as an acceptable appearance at a given age in sales horses and which findings constitute a risk to future performance.

### *Methods*

The research was performed via prospective cohort studies using enrolled samples. Sales repository radiographs were obtained with consignor permission from the 2016 Keeneland September Yearling Sale and the five major North American 2-year-old sales in 2017 run by Fasig-Tipton Company and Ocala Breeders' Sales Company. Ultrasonography was performed immediately prior to the sales on the forelimb suspensory branches of horses with consignor permission.

Stifle and fetlock radiographs were evaluated for MFC and sesamoid changes, respectively. MFC SCL were graded on a scale of 0-3 according to radiological size and axial MFC lucencies were recorded separately. Sesamoid findings relating to vascular channel appearance (0-3), abaxial contour changes and apical and abaxial fragments were graded according to a grading system established for the purposes of this study. Ultrasonographic findings relating to suspensory branch size, fibrillar pattern, the presence of hyperechoic foci, periligamentar tissue thickness and the adjacent proximal sesamoid bone surface were recorded during post-sale image evaluation.

Racing performance was assessed for all study horses until the end of their 4-year-old racing season and performance data was obtained from Equibase Company LLC. Racing performance was measured via eight outcome variables: whether the horse started at least one race by the end of their 4-year-old year, age at first race start, total number of race starts, total prizemoney earned, earnings per start, class of career best start achieved, weighted Listed and Group race starts and Class Performance Index. Clinical follow-up was sought to ascertain why horses that did not race never started.

Distributions of imaging findings were examined using descriptive statistics at the individual bone and branch level and at the horse level. Associations between imaging findings and racing performance from 2 to 4 years of age were examined using multivariate regression analyses, controlling for horse sex. Analysis was via logistic, negative binomial or linear regression as appropriate, with the threshold for significance set at  $\alpha=0.05$ .

### *Results*

Sales radiographs from 2,508 yearlings and 436 2-year-olds were included. This sample represented 11% of the annual US Thoroughbred foal crop. It comprised 36% of all yearlings sold at auction in North America in 2016 and 20% of all 2-year-olds sold at auction in North America in 2017.

Radiographs of 5,016 yearling stifles and 872 2-year-old stifles were evaluated. MFC SCLs of Grades 1-3 were observed in 242 yearlings (9.7%) and 49 2-year-olds (11.2%). Bilateral MFC SCLs of Grades 1-3 were observed in 54 yearlings (2.2%) and 12 2-year-olds (2.8%). Yearling Grade 1 MFC SCLs had either resolved (11/31), remained unchanged (14/31) or progressed to a Grade 2 (6/31) by 2-year-old

sales. Yearling Grade 2 MFC SCLs had either improved to a Grade 1 (2/10), remained unchanged (6/10) or progressed to a Grade 3 (2/10) by 2-year-old sales. Yearlings with a Grade 3 MFC SCL had a 78% probability of starting a race (95% Confidence Interval (CI): 58.2-89.6%), compared to 84% for MFC Grade 0 yearlings (95% CI: 82.7-85.8%). Six of the seven yearlings with axial MFC lucencies raced.

Radiographs of 20,064 yearling sesamoids and 3,488 2-year-old sesamoids were evaluated. Interobserver agreement using the new radiological grading system was substantial. Yearling findings associated with a significantly reduced probability of starting a race were: Grade 3 vascular channels in forelimb sesamoids (0.52,  $P < 0.001$ , 95% CI: 0.37-0.67), abaxial new bone in forelimb sesamoids (0.62,  $P = 0.01$ , 95% CI: 0.49-0.73), apical or abaxial fragments in forelimb sesamoids (0.55,  $P = 0.005$ , 95% CI: 0.37-0.72). For affected horses that did race, Grade 3 vascular channels in forelimb sesamoids were associated with fewer race starts (9.9 starts,  $P = 0.03$ , 95% CI: 8.0-12.2) and Grade 3 vascular channels in hindlimb sesamoids were associated with a delayed start to racing careers (54 days,  $P = 0.01$ , 95% CI: 20-89). Abaxial new bone in forelimb sesamoids was associated with a 54% reduction in total earnings ( $P = 0.003$ , 95% CI: 24-72) and a 46% reduction in earnings per start ( $P = 0.002$ , 95% CI: 21-64). Abaxial concavity occurred predominantly in yearling medial forelimb sesamoids, had no impact on racing performance and mostly resolved by two-year-old sale.

A total of 593 sales yearlings and 367 2-year-olds had ultrasonography performed on all four forelimb suspensory branches per horse. Grade  $\geq 2$  fibrillar branch change was present in 8.9% of yearlings and 14.4% of 2-year-olds. A 0.25cm increase in branch width was associated with a 49-day delayed start to racing careers ( $P < 0.001$ , 95% CI: 21-77 days). The presence of Grade 2 hyperechoic foci was associated with significantly lower total earnings ( $P = 0.01$ , 95% CI: \$2,000-\$16,022) and lower earnings per start ( $P = 0.003$ , 95% CI: \$349-\$1,718) in USD. Grade 3 fibrillar branch change had clinically important reductions in the probability of racing, calibre of racing performance and earnings. Grade 1 fibrillar pattern was associated with significantly higher earnings per start ( $P = 0.004$ , 95% CI: \$2,641-\$5,759).

A total of 2,204 yearling forelimb sesamoid-branch units and 1,336 2-year-old forelimb sesamoid-branch units were available for evaluation of concurrent imaging findings. The proportion of yearling

sesamoids with Grade  $\leq 1$  vascular channels that had adjacent Grade  $\geq 2$  fibrillar branch change was 1.2%. The same proportion for 2-year-olds was 3.8%, with medial forelimb sesamoids with Grade 1 vascular channels overrepresented in 2-year-olds. In yearlings, 31% of sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change and 59% of sesamoids with Grade 3 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change. In 2-year-olds, 47% of sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change and 67% of sesamoids with Grade 3 vascular channels had the same. Only 1 yearling and 1 2-year-old sesamoid with radiological abaxial concavity had Grade  $\geq 2$  fibrillar branch change.

### *Limitations*

The samples used in this research are representative of the population of interest at Thoroughbred sales but may underestimate the prevalence of severe lesions in non-sale horses. The study design could not address exclusions prior to sale. The findings are applicable to horses prepared for public auction and deemed fit to be entered for sale by consignors and their veterinarians. Clinical examinations were not performed for the purposes of this research.

### *Conclusions*

Regarding stifle lucencies, Grade 1 MFC SCLs were the most common type seen in yearling and 2-year-old sales horses. The majority of yearling Grade 1 MFC SCLs resolved or remained unchanged by 2-year-old sales. It was also possible for Grade 2 and 3 MFC SCLs to improve one grade between sales. Fewer sales yearlings with a Grade 3 MFC SCL raced, but in those that did race there was no evidence of worse performance compared to unaffected peers. Axial MFC lucencies did not affect racing performance.

For sesamoids, Grade 3 vascular channels, forelimb sesamoid abaxial new bone and forelimb sesamoid fragments are important findings in sales repository radiology. The new grading scale assigns a numerical grade for vascular channel appearance that matches the number of enlarged vascular channels evident in a given sesamoid. Abaxial contour changes, when present in sesamoids that are Grade 0 for

vascular channels, are noted separately as either abaxial new bone or abaxial concavity. Fragments are also noted and interpreted separately.

Reference values specific to young Thoroughbreds have been established for suspensory branch ultrasonography. Grade 1 fibrillar suspensory branch change should be regarded as an acceptable appearance in sales yearlings and 2-year-olds. Approximately one third of Grade 2 yearling branches progressed to a Grade 3 lesion. Evidence of enlarged branch width and Grade 2 hyperechoic foci at 2-year-old sales constitute a risk to racing performance.

The existence and prevalence of concurrent radiological and ultrasonographic findings in the proximal sesamoid bones and adjacent suspensory ligament branches has been established in yearling and 2-year-old Thoroughbred sales horses. General recommendations have been made for selective branch ultrasonography on the basis of sesamoid radiological appearance. The results support a separate aetiology for radiological sesamoid abaxial concavity that does not primarily involve the suspensory branch insertion.

This research provides veterinarians and the wider Thoroughbred industry with evidence-based determinations of the importance or otherwise of the various presale imaging findings seen in the MFCs, sesamoids and suspensory branches of yearlings and 2-year-olds. Many findings can be regarded as an acceptable appearance in yearlings and 2-year-olds. For those findings that are associated with reduced performance, sale and management decisions can be made based on quantitative evaluations of risk that are in the best interests of the horse.

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## DEDICATION

To John H. Peat and Ronald J. Flay. Thank you for your love and example.

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## CHAPTER 1 – INTRODUCTION

Radiograph repositories were introduced to Thoroughbred auction houses in the 1990s to centralise veterinary information on sale horses, as a service to sellers and buyers. A repository was first implemented at Thoroughbred sales in Kentucky, USA, in 1996 and has since become accepted practice at bloodstock auctions worldwide. A standardised protocol for each sale requires radiographs of certain limb joints to be acquired in a defined time period prior to the sale. Those radiographs are then lodged with the sales company and made available for veterinary inspection. The system was designed to allow buyers to consult with their veterinarian to make more informed decisions about a purchase, particularly with regard to the likelihood of findings affecting a horse's future soundness and athletic performance, alongside the individual buyer's risk tolerance for potential issues <sup>1</sup>. Veterinarians are tasked with providing this assessment of risk based on their own evaluation of diagnostic images. The current body of equine veterinary literature has led to a degree of uniformity in repository radiology, particularly regarding the interpretation of lesions of osteochondrosis and osteochondritis dissecans. However, the superior image quality now afforded by digital radiographs enables the detection of subtle changes previously under-reported or undetected with analogue film-screen radiography. With this increased clarity comes the potential for over-interpretation of certain findings. There are radiological changes that are commonly identified in the equine medial femoral condyle (MFC) and proximal sesamoid bone (sesamoid) for which veterinarians currently have limited scientific evidence to make objective assessments of the true significance or associated risk of future disease. Many recommendations to buyers are based on small datasets and personal experiences, causing discrepancies in veterinary interpretation and the subsequent advice that is relayed to vendors and prospective buyers. This is increasingly becoming a source of frustration within the Thoroughbred industry and has led some consignors, purchasers and trainers to question the usefulness of the repository system.

In the equine MFC, condylar flattening, subchondral lucencies (SCLs) of varying depths, cystic lesions adjacent to the central distal articular surface of the MFC and axially located cysts are known to

behave differently, but are often categorized together, making the significance of each lesion difficult to understand<sup>2-8</sup>. The prevalence of such lesions in the MFC ranges from 0.8% to 16% depending on how a lesion is defined<sup>2-8</sup>. Radiological abnormalities of the stifle can change with age in young horses and SCLs most commonly occur in the MFC within the first 18 months of life<sup>9-11</sup>. Both substantial radiological improvement and worsening of MFC SCLs have been documented in growing horses<sup>4,9</sup>. However, there is limited scientific evidence regarding the frequency of progression of SCLs in sales yearlings and 2-year-olds and their impact on racing careers<sup>12,13</sup>. Consequently, the interpretation of shallow MFC lucencies on presale radiographs is particularly problematic.

The sesamoid bones in the metacarpophalangeal and metatarsophalangeal joints are critical anatomical structures in the suspensory apparatus of the horse. Injuries to the suspensory apparatus may impair athletic ability and can have significant consequences in the horse<sup>14,15</sup>. The suspensory ligament (tendon of the third interosseous muscle) bifurcates in the distal metacarpal and metatarsal regions, giving rise to medial and lateral ligament branches<sup>16,17</sup>. The medial and lateral branches of the suspensory ligament insert abaxially onto the respective medial and lateral sesamoids<sup>16,17</sup>. Given their anatomical relationship and clinical importance, a link between pathological change in the sesamoids and suspensory branches has been discussed for over 50 years<sup>18-21</sup>. Variability in the radiological appearance of bony canals or channels within the sesamoids has been termed “sesamoiditis”<sup>18,19</sup>. This use of this term in reference to subclinical presale radiological findings in untrained Thoroughbreds strays from its original intended use as a diagnosis of clinical injury involving sesamoid changes and associated lameness<sup>19</sup>. Much like with MFC findings, inconsistent interpretation of sesamoid findings on presale radiographs and their impact or otherwise on future racing success has been controversial<sup>12,13,22,23</sup>.

Ultrasonography is not currently included in sales repository imaging protocols. However, ultrasonography of the suspensory branches is now frequently performed by veterinarians presale, as consignors and prospective purchasers sensitised to the issue of sesamoid changes seek to gather more information on which to base their sale and management decisions. Ultrasonography is being performed on yearlings with radiological sesamoid changes, as well as on valuable yearlings that have sesamoid

radiographs considered to be within normal limits. The ultrasonographic appearance of the suspensory ligament branches has never been studied at Thoroughbred sales and there is no published reference data for this population of young Thoroughbreds. As presale suspensory branch ultrasonography is becoming commonplace, it is critical that evidence-based research be performed to provide objective data to explain the long-term significance of varying degrees of ultrasonographic findings, as well as their relationship to radiological sesamoid findings.

The goal of the Orthopaedic Research Center at Colorado State University's C. Wayne McIlwraith Translational Medicine Institute is to improve the diagnosis and treatment of musculoskeletal diseases in athletic horses. Researchers strive to identify features via diagnostic imaging that may predispose the horse to injury during training and racing. To answer the current research questions surrounding presale imaging findings in the sesamoids, suspensory branches and MFCs, a large-scale scientific investigation in Thoroughbred sales horses is needed.

The research presented in this dissertation began with the hypothesis that specific radiological characteristics of the sesamoids would be associated with measured ultrasonographic findings in the suspensory branches in yearling and 2-year-old sales horses and with racing performance. It was also hypothesized that the severity of subchondral lucencies in yearling MFCs would dictate the risk of progression to a cystic lesion.

The specific aims of this work were 1) to objectively evaluate the MFCs and sesamoids on repository radiographs of sales yearlings and the concurrent ultrasonographic appearance of the adjacent suspensory branches at yearling sales, 2) to repeat radiological and ultrasonographical evaluations at 2-year-old in-training sales for included yearlings that presented for sale again as 2-year-olds and 3) to collect racing data until the end of the 4-year-old racing season, including follow-up information for any horses that did not race. The aims for data analysis were to analyse the distribution of each finding in yearlings and 2-year-olds, objectively evaluate paired yearling and 2-year-old sales radiographs to track the progression, regression or unchanged nature of findings between sales, and to determine statistical associations between presale imaging findings and racing performance outcomes.

The goal is to improve the Thoroughbred industry's understanding of MFC, sesamoid and suspensory branch changes in yearlings and 2-year-olds; to define the range of findings that might be considered insignificant within the normal population of sales horses, and to determine what level of change carries an increased risk of detrimental effects on future racing performance or clinical disease.

The radiograph repository system at sales should benefit individual horses by enabling appropriate management tailored to any abnormalities that carry an increased risk of injury during future training. The repository system should also benefit the Thoroughbred industry as a whole through increased knowledge and confidence in decision making surrounding bloodstock purchases.

The ultimate objective of this research is to provide a strong scientific evidence base to enable bloodstock sale and training management decisions that are based on quantitative evaluations of risk. This will increase industry confidence in the sales repository system and in the veterinarian's role in the management and care of the Thoroughbred horse.

## CHAPTER 2 – LITERATURE REVIEW

A review of the existing scientific literature surrounding presale radiology in Thoroughbred horses and ultrasonographic suspensory branch findings is set out below. The foundational sales radiology studies are discussed and specific evidence is presented as it relates to radiological changes seen in the medial femoral condyles (MFC), proximal sesamoid bones (sesamoids) and suspensory ligament branches (suspensory branches) of young Thoroughbreds. This is followed by a discussion of areas in which further research is required to build on the existing scientific knowledge, with the aim of advancing the evidence base utilised by equine veterinarians and the wider Thoroughbred industry at bloodstock sales.

### **2.1 Sales radiology research**

The work of Kane, Park, McIlwraith et al<sup>12,13</sup> examining various presale radiographic changes in Thoroughbred yearlings in North America remains the most comprehensive sales radiology study to-date. The prevalence and distribution of radiographic findings on presale yearling radiographs acquired between 1993-1996 were analysed, followed by their associations with 2 and 3-year-old racing performance. In this foundational work, the authors showed that reduced odds of starting a race as a 2 or 3-year-old were found in association with moderate or extreme palmar supracondylar lysis of the third metacarpus, enthesophyte formation on the proximal sesamoid bones, dorsal medial intercarpal joint disease and dorsoproximal first phalanx fragmentation in the hind fetlock. No association with reduced racing performance was found for sagittal ridge flattening of distal third metacarpus, circular lucencies in ulnar carpal bone or circular lucencies and vascular channels in proximal sesamoid bones. Much of this objective information continues to be used by sales veterinarians when evaluating radiographs in the repository setting today. One exception is the finding relating to vascular channels in the proximal sesamoid bones, for which different findings in subsequent research are discussed below. Medial femoral condyle lucencies were not addressed by Kane, Park, McIlwraith et al because of the limited availability of stifle radiographs at the time.

In the same journal issue as the above work, the first sales radiology study solely focused on sesamoid findings was published by Spike-Pierce and Bramlage (2003), involving Thoroughbred sales radiographs acquired from 1991-1994. The results of this study are discussed in the sesamoid section below. In addition to these two foundational articles, a number of subsequent sales radiology studies have been conducted and their methodologies are outlined in Table 2.1. Results of these studies that relate specifically to MFC lucencies and sesamoid changes are discussed in the following relevant sections of this chapter.

## **2.2 Medial femoral condyles**

Subchondral lucencies in the distal aspect of the MFC are a contentious presale finding on stifle radiographs in Thoroughbreds. Particular uncertainty surrounds the significance of such lucencies in clinically unaffected yearlings, which are yet to undergo substantial athletic training. The risk of progression to cystic lesions has not been quantified and the effect on future racing performance is not well understood.

Clinical and radiological aspects of stifle bone cysts in the horse were reported in the equine veterinary literature by Jeffcott and Kold in 1982<sup>3</sup>. The report detailed clinical cases in 33 horses, 28 of which were young Thoroughbreds, involving intermittent hindlimb lameness of varying degrees. Large, circular or dome-shaped cysts in the central aspect of the distal MFC with a distinct communication with the medial femorotibial joint were most common (28/33). Five cases involved cysts that were much more variable in shape and size and were located either in the axial aspect of the MFC, adjacent to the intercondylar fossa, or in the proximal tibia distal to the tibial spine. Seven of 13 cases with follow-up examination showed mild improvement in cyst radiodensity. The authors remarked that a prominent zone of sclerosis on follow-up radiographs was a feature in horses that fared well in subsequent racing performance.

In the 1990s, radiographic surveys conducted as part of the presale examination of young Thoroughbreds began to include the stifle region. Initially, the sites of most interest within the stifle were trochlear ridges of the distal femur affected by osteochondrosis lesions. In the foundational sales radiology research by Kane, Park, McIlwraith et al<sup>13</sup>, limited radiographic projections of the stifle were available and

TABLE 2.1. Published scientific literature involving sales radiology findings in horses

Primary Author	Publication	Study period	Study Population	Sampling Method	Number of Horses	Radiographic findings examined	Investigative Focus	Radiograph Type	Follow-Up Performed
<b>Sales radiograph studies investigating radiographic changes in multiple anatomic locations (in order of study size):</b>									
Kane	Equine Veterinary Journal	1993 - 1996	Keeneland & Fasig-Tipton Thoroughbred (TB) yearling sales, Kentucky USA	All yearling sales radiographs from one private practice in central Kentucky	1162	Front feet, fetlocks, carpi, hocks, stifles	Prevalence & distribution, association with 2 & 3yo racing performance	Analogue	2 & 3yo racing performance: number of race starts, percent of starts placed, money earned, earnings per start
Meagher	Journal of the American Veterinary Medical Association	1997 - 2001	2yo TB in-training sales in Pomona, California USA	All radiographs lodged at sale each year	953	Fetlocks, carpi, hocks, stifles	Prevalence & distribution, 1 furlong presale breeze times; association with entire career racing performance	Analogue	Time from sale date to first start; 2 & 3yo starts, earnings; career length, starts, earnings, number of stakes races run, highest purse
Cohen	Journal of Equine Veterinary Science	2002 - 2003	Fasig-Tipton Summer TB Yearling Sale, Texas USA	All radiographs lodged at sale each year	348	Fetlocks, carpi, hocks, stifles	Prevalence, association with sale price; association with 2 & 3yo performance	Digital	2 & 3yo performance: Number of starts, wins, placings, graded stakes races, earnings
Contino	Equine Veterinary Journal	2003 - 2006	Quarter horse yearlings and 2yos, National Cutting Horse Association (NCHA), USA	All radiographs from NCHA sales and survey radiographs from one private cutting horse ranch	458 (278 yearlings, 180 2-year-olds)	Fetlocks, carpi, hocks, stifles	Prevalence & distribution in yearlings & 2yos	Digital	None
Oliver	New Zealand Veterinary Journal	2003 - 2006	National Yearling TB Sales, New Zealand	Contacted all vendors for permission, obtained for 47% of yearlings with radiographs	1505	Hocks, stifles	Prevalence & distribution	Analogue (digital photographs of films used by 1 of 2 evaluators)	None
Preston	Journal of the American Veterinary Medical Association	2006	Keeneland September TB yearling sale, Kentucky USA	Random selection of all horses offered for sale	397	Fetlocks, carpi, hocks, stifles; history of arthroscopy	Prevalence, association with sale price; association with 2yo performance	Digital	2yo performance: having started a race, whether placed, having at least one placing, money earned, earnings per start
Furniss	Journal of the South African Veterinary Association	2008	National TB Yearling Sale, South Africa	All radiographs lodged at sale	269	Front feet, fetlocks, carpi, hocks, stifles	Prevalence & distribution	Digital	None

Sales radiograph studies investigating radiographic change involving one specific location:									
Spike-Pierce	Equine Veterinary Journal	1991 - 1994	TB Yearling sales, USA	Pre- and post-sale survey radiographs. Sampling not described	487	Proximal sesamoid bones	Categorisation of changes, association with 2 & 3yo racing performance	Analogue	2 & 3yo number of races started, earnings
Whitman	American Association of Equine Practitioners Convention Proceedings	1998 - 2003	Keeneland September TB yearling sales, Kentucky USA	Repository reports from 3 veterinarians with medial femoral condyle (MFC) lucencies $\geq 15\text{mm}^2$	52	Medial femoral condyle lucencies	Correlation of MFC lucency size to sale price & racing performance	Analogue	Race start as 2yo, number of 2yo starts, win percentage as 2yo, total starts and wins from 2 – 4 years-old, earnings
McLellan	Equine Veterinary Journal	2009 - 2010	TB horses at a single training centre in Ocala, Florida USA	All in-training medical records prior to first race, retrospective evaluation of yearling sale radiographs	291 (29 sesamoiditis cases, 262 controls)	Proximal sesamoid bones	Categorisation of changes using 3 different scales, association with SLBI in first year of race training	Digital	Clinical signs of SLBI in first year race training, with diagnosis confirmed ultrasonographically

no MFC lucencies or cystic lesions were reported. From the 600 yearlings with stifle radiographs included in the study, the MFC could only be clearly visualized in 170 lateromedial radiographs. No radiographs of the caudolateral-craniomedial oblique stifle projection were included.

With the growing utilisation of radiograph repositories at bloodstock sales came the increasingly topical discussion of survey radiology findings. Whitman, Morehead, Pritchard et al presented a conference abstract in 2006 <sup>8</sup> that correlated MFC lucency size to sale price and racing performance in 52 Thoroughbreds with a mean age of 18.1 months old. Cases were derived from searching 2915 Thoroughbred yearling sales reports from a 5-year period. The authors categorised MFC lucencies into two groups: cysts (25 horses), i.e. rounded lucencies within the condyle that usually communicate with the medial femorotibial joint, and subchondral lucencies (27 horses), i.e. shallow crescent-shaped lucencies at the distal aspect of the condyle that have a greater width (lateral-medial) than height (proximal-distal). There was a prevalence of 0.93% for MFC subchondral lucencies and 0.86% for MFC cysts, with a 1.8% overall prevalence of MFC lucencies of at least 15 mm<sup>2</sup> in size. Lucencies under 15 mm<sup>2</sup> in area were not included. The area of the decreased radiodensity in the MFC was determined by multiplying its largest proximal-distal dimension by its largest lateral-medial dimension, on either caudocranial or 20° caudolateral craniomedial oblique radiograph projections. In unilaterally affected horses, the MFC lucency was significantly more likely to occur in the right stifle than the left. Significantly fewer yearlings with MFC cysts sold at auction, with a 60% clearance rate, compared to yearlings with MFC subchondral lucencies and unaffected yearlings, of which 89% and 83% were sold respectively. The median sale prices for yearlings with MFC cysts and subchondral lucencies were significantly lower than the median sale price for yearlings with no MFC lucencies (\$32,000 and \$34,000 respectively, compared to \$95,000). The authors concluded that subchondral lucencies of the MFC present no threat to a horse's racing ability.

A radiology study performed on Thoroughbred yearlings at New Zealand sales by Oliver, Baird, Baird et al <sup>5</sup>, reported the prevalence and distribution of stifle lesions using only the caudocranial radiographic projection. A minimum width of 5 mm was required for inclusion of an MFC lucency. Under this definition, 16% of 1505 yearlings had a lucency; 11% 1-2 mm deep, 4% 3-5 mm deep and 1.7% had a

cyst-like lesion >5 mm deep. 2.5% of yearlings had bilateral lucencies. A significantly higher proportion of MFC lucencies were seen in the right hindlimb compared to the left hindlimb (172 yearlings, 96 yearlings respectively). Analysis showed no difference in the proportion of colts or fillies affected. This refuted the clinical impression that colts were more frequently affected, due to their often larger size and faster growth. The images used in this study were analogue film-screen radiographs, plus digitally converted film-screen radiographs. The authors noted some loss of image detail due to this method of conversion.

Flattening of the MFC contour on radiographs, in the absence of a lucency, has been a source of much debate, both in terms of whether it is real and whether it is important. Oliver, Baird, Baird et al defined subjective flattening as any change in the contour of the distal MFC <1 mm deep from the articular surface. MFCs from 52% of 1505 yearlings met this definition for flattening in their study. The authors considered MFC flattening to be a normal variation in yearlings and unlikely to be of clinical significance. It was also mentioned that the higher prevalence of flattening and lucencies in right stifles could be positioning artefact and the result of altered obliquity when the radiograph is taken. Projection angle, radiographic technique and hindlimb stance affect MFC shape on the resulting images.

Preston, Brown, Trumble et al <sup>24</sup> examined associations between yearling sales radiographic findings and 2-year-old racing performance, though only 6 radiographic findings were recorded and lesions within certain joints were combined. The data analysis grouped lateral and medial trochlear ridge defects and MFC cysts together, however these stifle findings are distinct entities that may differ in their clinical importance.

In 2012, Contino, Park and McIlwraith published a study of sales radiographic changes in Quarter Horses intended for cutting <sup>25</sup>. These authors used a grading scale for MFC lucencies based on their radiological appearance in both the caudocranial and oblique radiographic projections. This grading scale is equally applicable to the radiological appearance of MFC lucencies in Thoroughbreds. Subsequent follow-up data on this group of Quarter Horses found no statistically significant association between radiographic lesions of the MFC and subsequent cutting horse competition performance <sup>26</sup>.

A recent retrospective cohort study by Perez-Nogues et al examined the change in size and morphology of shallow MFC lucencies seen on presale screening radiographs of weanling and yearling Thoroughbreds, with mean ages of 10 and 17 months old (7-12 months old and 14-19 months old, respectively) <sup>6</sup>. The objective was to address the fact that it is unclear whether shallow MFC lucencies are precursors to larger cystic lesions or fully developed defects that remodel into a flattened condyle. The study aimed to determine whether radiologically diagnosed shallow MFC lucencies progress, regress or remain static in size between weanling and yearling screening radiographs. Shallow lucencies were defined as  $\leq 3$  mm in height from the articular surface, measured on the caudo-cranial radiographic projection. This type of shallow MFC lucency was present in 6.4% of horses with paired weanling and yearling radiology reports (248 of 3874 horses). A right-sided predilection was seen, with 74% of the shallow MFC lucencies involving the right stifle and 26% involving the left stifle. The mean percentage width of the MFC affected by a shallow lucency ranged from 14% to 21%. Lucencies in the left MFC were significantly smaller than those in the right stifle. The authors hypothesized that larger (deeper or wider) lucencies were more likely to increase in size or progress to cystic lesions. The presence of subchondral bone sclerosis surrounding shallow MFC lucencies was found to be inconsistent by other authors and thus was not used in the definition of lesion characterisation in this study. In addressing the question of whether shallow MFC lucencies are distinct developmental defects that may remodel with time, or whether they are precursors to cystic lesions, the authors found that 41% of lucencies remained unchanged in size from weanling to yearling age, 24% became smaller, 6% resolved, 8% increased in size and 4% developed into cysts. Also of note is that 13% of the MFC lucencies seen in yearlings arose from MFCs that did not have an apparent radiolucency at weanling age. The authors considered whether MFC lucencies may be a late-developing lesion, similar to femoral trochlear ridge OCD in the stifle (Dik, Enzerink 1999). The study concluded that most shallow radiographic MFC lucencies remain a similar size between weanling and yearling age, however a small proportion have the potential to resolve (6%) or progress to a cystic lesion (4%). The possible changing nature of MFC lucencies beyond yearling age was outside the scope of this study. Regarding impact on racing performance, 182 of 248 horses with a shallow MFC lucency had racing data available. In these 182

horses, those with a shallow MFC lucency had significantly fewer 2-year-old race starts compared to their unaffected maternal siblings. The authors concluded that shallow MFC lucencies were associated with a later start to racing careers but did not significantly impact performance once racing (when performance is assessed by number of starts, earnings and earnings per start). Colts and geldings earned significantly more money as 3-year-olds than fillies, suggesting horse sex should be taken into consideration when analysing performance.

### **2.3 Proximal sesamoid bones**

Changes in the radiological appearance of the sesamoids have been discussed in the equine veterinary literature for over 50 years. Published in 1971, Fraser's work entitled "Some conditions of the proximal sesamoid bones in the horse"<sup>18</sup> remarked on "sesamoiditis or inflammation of the sesamoid bones" and described a distinct form of pathological change involving the external or non-articular surface, whereby "an initial periostitis of the abaxial surface of the bone [is seen] due to tearing of the ligamentous attachments." Fraser concluded that many sesamoid lesions seen on radiological examination are difficult to interpret and most cases of sesamoid lameness are related to previous primary soft tissue damage.

Later the same year, a radiographic study into sesamoid changes in racing Thoroughbreds with lameness was produced by O'Brien, Morgan, Wheat et al<sup>19</sup>. The authors aimed to more clearly define "sesamoiditis" as a term used to describe injury of the sesamoids in Thoroughbreds characterized by lameness. Radiographs from 100 racing Thoroughbreds with suspected sesamoid lameness were studied and additional postmortem and radiographic examinations were made. Using analogue film-screen radiography, vascular channels were observed in 50% of fetlock radiographs (77/154). The appearance of the normal sesamoid in mature racing Thoroughbreds was defined as two or fewer sharply marginated vascular channels and well-defined abaxial, flexor and articular surfaces without radiodense buildup. The authors proposed that the term "sesamoiditis" be used to describe primary disease of the suspensory apparatus involving bony change on the sesamoid abaxial surface or basilar area with increased radiodense buildup, increased number and irregularity of the vascular channels and increased coarseness and mottling

of the bone trabeculation. It was concluded that “large vascular channels in sesamoids are radiographic findings of immense importance” and that “these diagnoses present a diagnostic challenge to the clinician and radiologist.”

In the early 1990s, research into the adaptive response of the equine proximal sesamoid bones to exercise was performed on a small group of trained and untrained Thoroughbred racehorses<sup>27</sup>. Training stimuli resulted in a significant stress-adaptive response and regional morphological variations in the cancellous bone. A separate study into the clinical relevance of the microvasculature of the equine proximal sesamoid bone was also published<sup>20</sup>. This showed that the sesamoids were supplied by multiple branches of the medial and lateral palmar digital arteries, which enter the proximal half of the bone on its abaxial surface and arborize into smaller branches that supply the rest of the bone. The major branches reside in bony canals that match the radiological lucencies seen in horses with “sesamoiditis”.

Radiography became an increasingly important part of the presale veterinary examination of horses during the 1990s. Particularly with the establishment of radiograph repositories at Thoroughbred auction houses, this led to increased scrutiny of radiological sesamoid findings, now in the absence of clinical disease, in untrained yearlings and unraced 2-year-olds. The condition of “sesamoiditis”, previously diagnosed in clinical cases, was being applied to radiological findings on survey and presale radiographs of Thoroughbred yearlings. This was addressed by two studies published in 2003 that retrospectively examined yearling sales radiographs from the early-mid 1990s.

Kane, Park, McIlwraith et al<sup>13</sup> performed the largest study to-date on radiographic changes in 1162 Thoroughbred yearlings, from Kentucky sales radiographs obtained from one private practice. This represented a 7% sample of all yearlings sold at these sales (1162/16577). Specific to sesamoid findings, 98% of yearlings had radiolucent vascular channels evident in at least one forelimb sesamoid. Irregular vascular channels were present in 79%, when defined as channels “greater than 2 mm wide or with non-parallel sides for any portion of their length”. To be categorized as normal, vascular channels had to have parallel sides for their entire length. Based on this classification, irregular vascular channels were more common than regular/normal vascular channels. Analysis of associations between radiological findings and

racing performance at 2 and 3 years of age found no association between racing performance and vascular channel appearance <sup>12</sup>. Significantly reduced odds of racing was found for enthesiophyte formation on forelimb sesamoids (57% started, 8/14), where enthesiophytes were defined as bone production within the suspensory ligament branch or distal sesamoidean ligament attachments, i.e. abaxial or basilar new bone. Hind sesamoid enthesiophyte formation was associated with reduced earnings, earnings per start and percent of starts placed.

Another foundational sales radiology study on sesamoids, published the same year by Spike-Pierce and Bramlage <sup>22</sup>, reached a different conclusion regarding the significance of vascular channels. This study involved 487 Kentucky sales yearlings and did not regard radiologically evident vascular channels as incidental. Rather, analysis of future racing performance showed a significant decrease in the mean number of starts and lower total earnings at 2 and 3 years of age for Thoroughbred yearlings with 3 or more abnormally shaped vascular channels in a sesamoid (10/487 horses). For yearlings with 1 or 2 abnormally shaped vascular channels, fewer starts and reduced earnings were seen at 2 years of age only (76/487 horses). Changes in the abaxial contour of sesamoids, in the absence of abnormal vascular channels, were not associated with reduced performance. It was concluded that horses with enlarged vascular channels as yearlings start fewer races and earn less prize money.

The discrepancy in the conclusions drawn from each study can be explained in part by differences in the classification criteria used to grade the radiological appearance of the vascular channels. By the definition of Kane, Park, McIlwraith et al, 32% of yearlings (366/1127) had 3 or more enlarged vascular channels, whereas the definition of Spike-Pierce and Bramlage classified 2% of yearlings (10/487) as having 3 or more enlarged vascular channels. The latter study allowed for a conical opening at the abaxial entrance of the vascular channel to be considered within normal limits, whereas formerly any deviation from parallel sides was considered abnormal even if only at the abaxial end of the channel.

Building on these two foundational studies, further research specific to sesamoid findings was published by McLellan and Plevin in 2014 <sup>23</sup>, involving 291 yearlings with sales radiographs from 2009-2010. The authors recognized the importance of the radiological grading scale used to define vascular

channel appearance and used a retrospective case control method to show that significant sesamoid change at yearling sale time, when identified using a modified Spike-Pierce grading scale, was associated with an increased likelihood of clinical suspensory ligament branch injury in the first year of race training. The study concluded that sesamoids with one or more irregular vascular channels wider than 2 mm were 5 times more likely to develop clinical signs of suspensory ligament branch injury within the first year of race training. This was the first published evidence of a predisposition to subsequent suspensory ligament branch injury for horses with radiological sesamoid change at yearling sales.

## **2.4 Suspensory ligament branches**

Prepurchase ultrasonography of the suspensory branches has become commonplace in Thoroughbred sales, as prospective purchasers sensitized to the issue of sesamoid changes and potential branch injury seek to gather more information on which to base their decision-making. The detection of ultrasonographic lesions can have implications for the market value and career management of the horse<sup>28</sup>. However, to-date, no research has been published on the ultrasonographic appearance of the suspensory ligament branches in yearlings and 2-year-olds at Thoroughbred sales. Relevant work has been conducted in young Thoroughbreds outside of the sales environment, investigating subclinical and clinical suspensory branch changes.

Ultrasonographic branch abnormalities were examined in a survey of 60 Thoroughbred racehorses from a training yard in the United Kingdom by Ramzan, Palmer, Dallas et al in 2013<sup>28</sup>. The ultrasonographic appearance of the forelimb suspensory branches was graded from 0-3 on an ordinal scale for echogenicity and fibrillar pattern disruption. If present, evidence of sesamoid bone margin disruption at the branch insertion was also recorded. All horses were historically and clinically free of desmopathy, thus this was the first report of subclinical ultrasonographic branch abnormalities in Thoroughbred racehorses. Mild fibrillar pattern change (grade 1) was seen in 26-28% of horses. Moderate severity branch changes (grade  $\geq 2$ ) were found in 6.7% of horses (4/60). The majority of abnormalities were present in the distal (insertional) third of the branch and, specifically, the palmar or abaxial margin of the insertion. Medial

branches were overrepresented. The authors concluded that ultrasonographic branch abnormalities are present in a proportion of normal racehorses in training and may not always be related to clinical desmopathy. Further longitudinal studies were needed to determine whether subclinical findings were predictive of future injury.

Evidence of subclinical suspensory branch abnormalities has also been published for disciplines other than flat Thoroughbred racing. Fairburn, Busschers and Barr reported on the ultrasonographic findings in National Hunt (jumps) racehorses in 2017 <sup>29</sup>, identifying grade 2 branch change in 31% of horses in one training yard (19/62). Medial branches were overrepresented and the medial branch insertional cross-sectional area was significantly larger than that of the lateral branch. In 60 elite showjumping Warmblood horses, Read, Boys-Smith and Bathe <sup>30</sup> also reported a high prevalence of subclinical grade 2 branch change. Periligamentar fibrosis was overrepresented in hindlimbs in the latter study and is a known poor prognostic indicator of clinical branch injury in sport horses <sup>31</sup>.

The suspensory branches were identified by O'Brien and Smith as one of the structures most commonly affected by tendon/ligament mineralization in the horses, in a review of cases presented to the Royal Veterinary College Hospital, London, between 1999 and 2015 <sup>32</sup>. The results indicated that mineralization can sometimes be an incidental finding in equine tendons and ligaments. Dystrophic mineralization was also a recognised feature in some cases of clinical suspensory branch injury reported by Marneris and Dyson <sup>31</sup>. Little is known about the importance of subclinical suspensory branch mineralization specifically in young Thoroughbreds when it occurs prior to racing.

The current evidence regarding suspensory branch ultrasonography specific to young Thoroughbreds comes from the work of Plevin and McLellan. In 2014, these authors published a study into the effects of clinical insertional branch desmitis on future racing performance in juvenile Thoroughbreds <sup>33</sup>. The aim was to examine the prognosis for 2-year-olds that injure the suspensory branch insertion prior to their first race. Cases were identified by clinical signs including reaction to branch palpation and visible swelling, then confirmed with ultrasonography. From 896 Thoroughbreds in their first year of training at a training centre in Florida, the prevalence of clinical branch injury was 9.5% (85/896). Analysis of racing

performance concluded that mildly affected horses (grade 1) had a reduced number of starts and lower earnings per start as a 2-year-old then performed similarly to cohorts by their 3-year-old season. Grade 2 cases continued to have fewer starts and lower earnings at 3 years of age. There were insufficient numbers of Grade 3 branch lesions to further analyse the racing performance of this group.

In 2016, Plevin, McLellan and O’Keeffe published the first investigation into the association between the presence of sesamoiditis and subclinical suspensory branch change in untrained (non-sale) yearlings<sup>34</sup>. Fifty horses were followed prospectively during the first 9 months of their training careers and the odds of developing clinical suspensory branch injury during this time were determined. By design, this study did not include horses that had been through bloodstock auctions, instead only selected homebred horses so as to remove variables related to horses undergoing a yearling sales preparation. The results found a significant relationship between radiological sesamoid change and subclinical suspensory branch change in a population of untrained Thoroughbreds, and an association with subsequent clinical branch injury. The findings supported the belief that sesamoid change and suspensory branch injury are aetiologically related and the authors recommended further work using larger populations to provide further insight into these findings.

## **2.5 Conclusions**

The studies described above form the basis of the evidence currently used by veterinarians evaluating presale radiographs in Thoroughbreds, in conjunction with shared clinical experience and anecdotal reports. Recommendations from the previous authors and discussions within the Thoroughbred industry have highlighted areas in which further research is needed, to build on the work already performed.

A study involving large numbers of high-quality repository radiographs is warranted<sup>5</sup>. Relatively low numbers have been a practical limitation in previous studies and have, at times, limited the conclusions that may be drawn. Determination of associations with racing performance has been most difficult for the less common radiological findings. Even in the largest study to-date, which included 1162 yearlings<sup>12</sup>, the statistical models for some radiological findings (percent of starts placed, total money earned and average

earnings per start) were unstable due to the small number of yearlings affected within the sample. Early studies were also limited by lower quality images, at times inconsistent radiographic technique, and single projections per joint. The image resolution now obtained with digital radiography is substantially greater than that of analogue film-screen radiographs. Inclusion criteria that samples from the entire population of yearling radiographs available at a sale would ensure horses are not enrolled from only a subset of consignors associated with one veterinary practice or one training centre <sup>12,13,22,23,33</sup>.

A new grading system for radiological sesamoid findings is needed and it is critical that such a grading scale is concise, easily understood and able to be applied consistently among veterinarians in clinical practice. The grading scale must also allow for the identification and analysis of the importance, or otherwise, of a single enlarged vascular channel, as distinct from “one or more” enlarged vascular channels that have been grouped together in recent studies <sup>22,34</sup>. Personal communication with the authors of the foundational sales radiology sesamoid studies has confirmed that separate analysis of a single enlarged vascular channel would enhance understanding of the information provided by their work <sup>13,22,34</sup>. Enrolling horses and then prospectively evaluating all images specifically for a research study would enable new grading scales to be used, in a way that is not possible when retrospectively gathering data from previous sales radiology reports without re-evaluating the images <sup>6</sup>.

There is currently no research that examines both yearling and two-year-old sales radiographs from individual horses. Paired radiographs would enable the progression, regression or unchanged nature of certain radiological findings to be studied, under the sales preparation conditions in which these trade horses are managed. Given the spectrum of sesamoid change that is seen in yearlings, there is currently insufficient evidence for veterinarians to objectively advise what degree of sesamoid change constitutes an unsuitable risk for a 2-year-old in-training sales prospect. This knowledge is needed so that horses with an increased risk of sesamoid injury or insertional desmopathy are managed appropriately and, conversely, yearlings with mild sesamoid changes associated with no increased risk are not unnecessarily ruled out as trade prospects.

For the same reason, paired yearling and two-year-old radiographs need to be studied to follow lucencies in the medial femoral condyle. Sales repositories now prescribe the inclusion of caudocranial and caudolateral-craniomedial oblique radiographic projections of the stifle. The inclusion of the oblique view minimizes misinterpretation of MFC contour abnormalities due to positioning artifacts resulting from variations in the proximal-distal angle of the X-ray beam on the caudo-cranial projection. Specifically, over-interpretation of condyle flattening can be avoided and true lucencies should be identifiable with the use of orthogonal projections. Unknown rates of progression from subchondral lucencies to cystic lesions requires that evidence be drawn prospectively from a population of sales horses as they progress into training and racing. Despite showing a reduced sale price and reduced likelihood of sale at auction, the existing literature does not suggest that MFC lucencies significantly affect horses' long-term racing ability by the performance measures used. Performance outcome measures commonly included number of starts, wins and percent of starts placed from 2-4-years-old, plus earnings per start and total earnings from 2-4-years-old. A peer-reviewed study involving a large sample size of sales horses, the use of orthogonal radiographic projections of the MFC and radiographic follow-up from yearlings to two-year-olds, combined with prospective racing performance data, is needed to address the questions that remain about MFC lucencies.

The prevalence of ultrasonographic findings in the suspensory branches of sales yearlings and 2-year-olds has not yet been studied and no reference data exists for ultrasonographic measurements in this population of horses<sup>28</sup>. The impact of the varying degrees of subclinical ultrasonographic findings at sales age on future racing performance is unknown. Features of suspensory branch injury that occur in sport horses, such as periligamentar fibrosis and dystrophic mineralisation<sup>31,32</sup>, have not been studied in sales Thoroughbreds and so their importance in this group of horses is yet to be determined. Authors of previous equine ultrasonography research have identified limitations associated with the evaluation of static ultrasound images at a later date after ultrasonography<sup>34</sup>. Ideally, future studies would include collection of dynamic ultrasonographic cine loops (short videos) in addition to still images, to enable accurate evaluation by a blinded observer post-acquisition. Authors have also called for further work correlating

suspensory branch findings with the radiological appearance of the sesamoids in large numbers of horses  
28,34 .

Evidence specific to horses that have undergone preparation for sale at public auction is needed. The environment in which yearlings are prepared for bloodstock auctions has the potential to alter their musculoskeletal development, in comparison to homebred horses that are not under the same pressure to achieve the desired phenotype for success at a public sale. Subjectively, this includes the market demand for a physically strong, well-conditioned appearance in sales yearlings. A rising plane of nutrition during sales preparation and daily exercise differs from the management of homebred horses that are not being prepared for sale as a yearling. Yearlings then purchased with the intention of resale at a 2-year-old in-training sale have a short period in which to be broken to saddle and commence athletic training. A successful selling price at today's 2-year-old sales often depends in-part on a fast breeze (gallop) time. The intensity of the 2-year-old sales preparation is at the forefront of buyers' minds when selecting suitable resale prospects as yearlings. Thus, research directly applicable to sales horses will enhance the evidence base utilized by equine veterinarians and the wider Thoroughbred industry in addressing the above research problems.

CHAPTER 3 – SUBCHONDRAL LUCENCIES IN THE MEDIAL FEMORAL CONDYLE OF  
YEARLING AND 2-YEAR-OLD THOROUGHBRED SALES HORSES: PREVALENCE,  
PROGRESSION AND ASSOCIATIONS WITH RACING PERFORMANCE<sup>a</sup>

### 3.1 Summary

Subchondral lucencies (SCLs) in the distal aspect of the medial femoral condyle (MFC) of young Thoroughbred horses are a source of controversy on presale radiographs. There is limited scientific evidence regarding the risk of progression and impact on future racing performance.

This was a prospective cohort study using an enrolled sample. The objectives were to 1) define the prevalence of MFC SCLs on sales repository radiographs in yearling and 2-year-old Thoroughbreds, 2) identify any associations between grade of MFC SCL and future racing performance and 3) monitor changes in MFC SCL grades between yearling and 2-year-old sales in horses that present for sale at both ages.

Radiographs were obtained with consignor permission from a 2016 yearling sale and five 2017 2-year-old sales. Stifle radiographs were evaluated and MFC SCL graded on a scale of 0-3. Axial MFC lucencies were recorded separately. Maximum MFC grades per horse were analysed for associations with racing performance outcomes, adjusted for sex, to the end of the horses' 4-year-old racing year. Analysis was via logistic, negative binomial or linear regression as appropriate, with the threshold for significance set at  $\alpha = 0.05$ .

Stifle radiographs for 2508 yearlings (5,016 Stifles) and 436 2-year-olds (872 Stifles) were included in the study. MFC SCLs of Grades 1-3 were observed in 242 (9.65%) yearlings and of 49 (11.2%) 2-year-olds. Bilateral MFC SCLs of Grades 1-3 were observed in 54 (2.2%) of yearlings and 12 (2.8%) of 2-year-olds. Grade 1 MFC SCL in yearlings either remained static (14/31), progressed to a Grade 2 (6/31) or

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<sup>a</sup> Peat, F. J., Kawcak, C. E., McIlwraith, C. W., Keenan, D. P., Berk, J. T., & Mork, D. S. (2024). Subchondral lucencies of the medial femoral condyle in yearling and 2-year-old Thoroughbred sales horses: Prevalence, progression and associations with racing performance. *Equine Veterinary Journal*, 56(1), 99–109. <https://doi.org/10.1111/evj.13945>. *Included with permission of journal Editor-In-Chief.*

resolved (11/31). Grade 2 MFC SCLs in yearlings remained static (6/10), progressed to a Grade 3 (2/10) or improved to a Grade 1 (2/10) by the 2-year-old sale. Yearlings with a Grade 3 MFC SCL had a 78% probability of starting a race (95% confidence interval (CI) 58.2 – 89.6%), compared to 84% for MFC Grade 0 yearlings (95% CI 82.7 – 85.8%). Six of the seven yearlings with axial MFC lucencies raced.

Grade 1 MFC SCLs are the most common type seen in yearling and 2-year-old sales horses. The majority of yearling Grade 1 MFC SCLs resolved or remained static by 2-year-old sales. It was also possible for Grade 2 and 3 MFC SCLs to improve one grade between sales. Fewer sales yearlings with a Grade 3 MFC SCL raced, but in those that did race there was no evidence of worse performance compared to unaffected peers. Axial MFC lucencies did not affect racing performance.

### **3.2 Introduction**

The significance of radiological findings on presale examinations is a concern to the Thoroughbred industry,<sup>1</sup> and the radiological appearance of subchondral lucencies (SCLs) in the equine medial femoral condyle (MFC) and their potential impact on athletic performance has been a source of controversy.<sup>2-8</sup> The incidence of lesions in the MFC ranges from 0.8% to 16% depending on how a lesion is defined.<sup>2-8</sup> Subchondral lucencies (which may be shallow in nature), condylar flattening, cysts in the axial MFC and cysts on the central MFC surface are known to behave differently, but are often categorized together, making the significance of each lesion difficult to understand.<sup>2-8</sup> Radiological abnormalities of the stifle can change with age in young horses, and SCLs most commonly occur in the MFC within the first 18 months of life.<sup>9-11</sup> Both substantial radiological improvement and worsening of MFC SCLs have been documented in growing horses.<sup>3,9</sup> However, there is limited scientific evidence regarding the frequency of progression of SCLs in sales yearlings and 2-year-olds, and their impact on racing careers.<sup>12,13</sup>

This study aimed to objectively evaluate yearling and 2-year-old Thoroughbred sales repository radiographs from a large percentage of the US population, to clearly define the prevalence and lesion characteristics of MFC SCLs and to analyse associations of the lesion grades with racing performance. In horses presented at both sales, the study aimed to identify differences in grade of MFC SCL between one

and two years of age. It was hypothesized that yearlings with Grade 3 lesions would have reduced starts and that there would be no progression of lesion severity with age.

### **3.3 Methods**

#### *3.3.1 Study design*

A prospective cohort study was conducted using an enrolled sample of yearling Thoroughbred horses. Researchers sought consent for study inclusion from consignors of all horses at the 2016 Keeneland September Yearling Sale (Lexington, KY, USA), and from the five major subsequent 2-year-old sales in 2017: Fasig-Tipton Gulfstream, Ocala Breeders' Sales (OBS) March, OBS Spring, Fasig-Tipton Maryland and OBS June (Ocala, FL, USA) (Appendix 1). Horses for which study consent was granted at the yearling sale required new research consent from their respective 2-year-old consignors to be eligible for 2-year-old sale radiograph inclusion (Appendix 2). Radiographs for which permission was granted were downloaded in DICOM format after the completion of each sale for confidential evaluation.

#### *3.3.2 Radiological evaluation*

Six radiographs acquired as part of the standard sales repository protocol were evaluated for each horse: the lateromedial (LM), caudocranial elevated 10-20° proximodistal (CdCr) and caudolateral 30°-craniomedial oblique (CdLCrMO) projections of the left and right stifles. The radiological appearance of each MFC was categorised into one of four grades as depicted by the example radiographs in Figures 3.1-3.4. Grades were recorded for the MFC appearance on CdCr and CdLCrMO images and the highest grade was taken to represent that MFC. Grade definitions are tabulated in Table 3.1.

TABLE 3.1 Grading scale for the radiological appearance of a subchondral lucency (SCL) in the equine medial femoral condyle

MFC SCL radiological appearance	Definition
Grade 0	Continuously convex or flattened appearance of the distal articular surface of the condyle with no radiolucency
Grade 1	Mild, shallow, crescent-shaped lucency with proximodistal depth $\leq 3$ mm and width greater than depth
Grade 2	Moderate, dome-shaped lucency that extends through the subchondral bone with depth approaching width
Grade 3	Large, spherical or ovoid cystic lesion that communicates with the central MFC articular surface

Lucencies located within the axial aspect of the MFC adjacent to the medial intercondylar eminence of the tibia, as depicted in Figure 3.5, were recorded separately from the MFC SCL grading system. MFCs with an axial lucency were graded according to the appearance of the central distal articular portion of the condyle, as for all other stifles. An additional record was made of the presence and size of the axial lucency.

The majority of radiographs were read by two veterinarians with 18 and 10 years' experience in repository radiology, respectively (DK, FP). Prior to evaluating the study radiographs, all four involved veterinarians underwent a period of training during which they discussed the grading system and applied group consensus to a sample of images. Each veterinarian then independently evaluated the same sets of CdCr and CdLCrMO stifle radiographs for ten MFCs, to test interobserver agreement. During evaluation of the study images by individual veterinarians, questionable findings were graded via consensus when

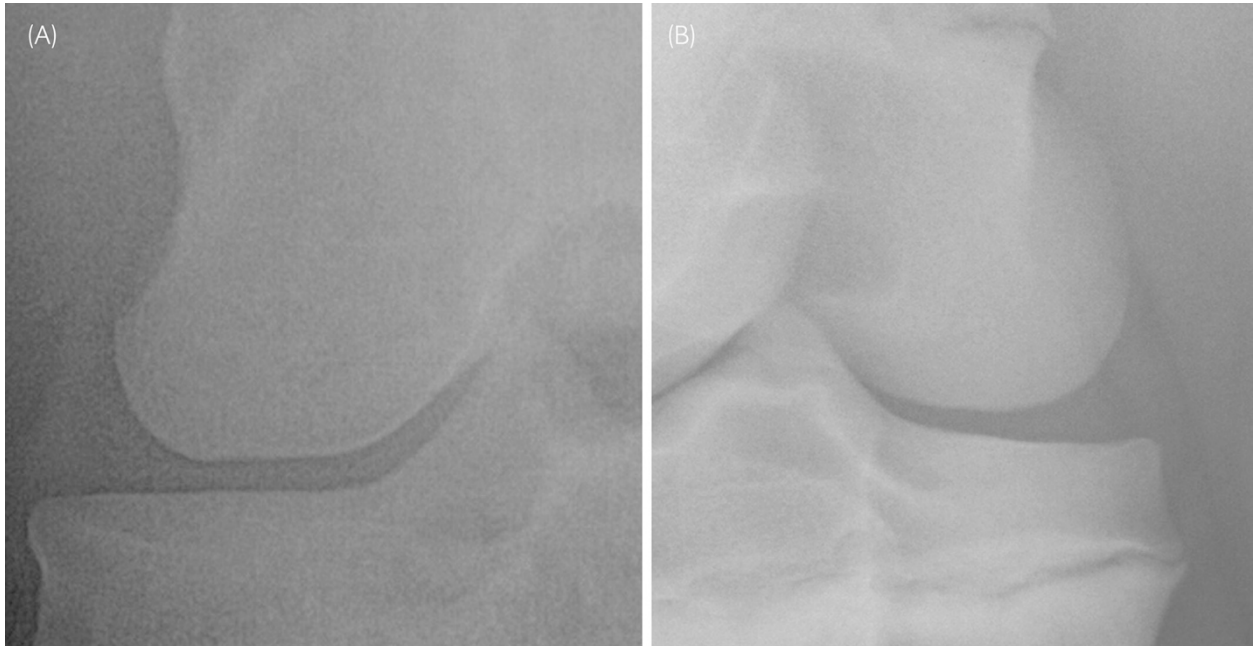


FIGURE 3.1. Radiographs showing the appearance of a Grade 0 medial femoral condyle (MFC) in a Thoroughbred yearling on CdCr (a) and CdLCrMO (b) projections. The Grade 0 MFC has no subchondral lucency. Flattening of the distal articular contour of the condyle is allowed.

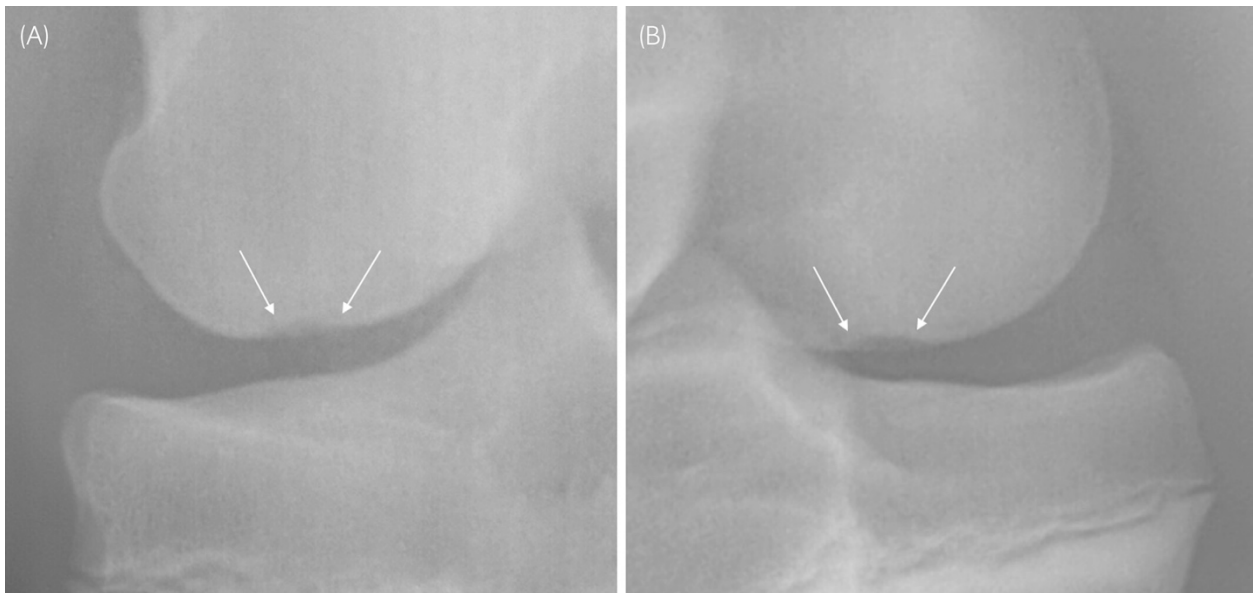


FIGURE 3.2. Radiographs showing the appearance of a Grade 1 subchondral lucency (SCL, white arrows) in the distal aspect of the equine medial femoral condyle (MFC) in a Thoroughbred yearling on CdCr (a) and CdLCrMO (b) projections. The Grade 1 MFC SCL is a mild, shallow, crescent-shaped lucency with a proximodistal depth  $\leq 3$ mm and with an axial-abaxial width greater than its depth.

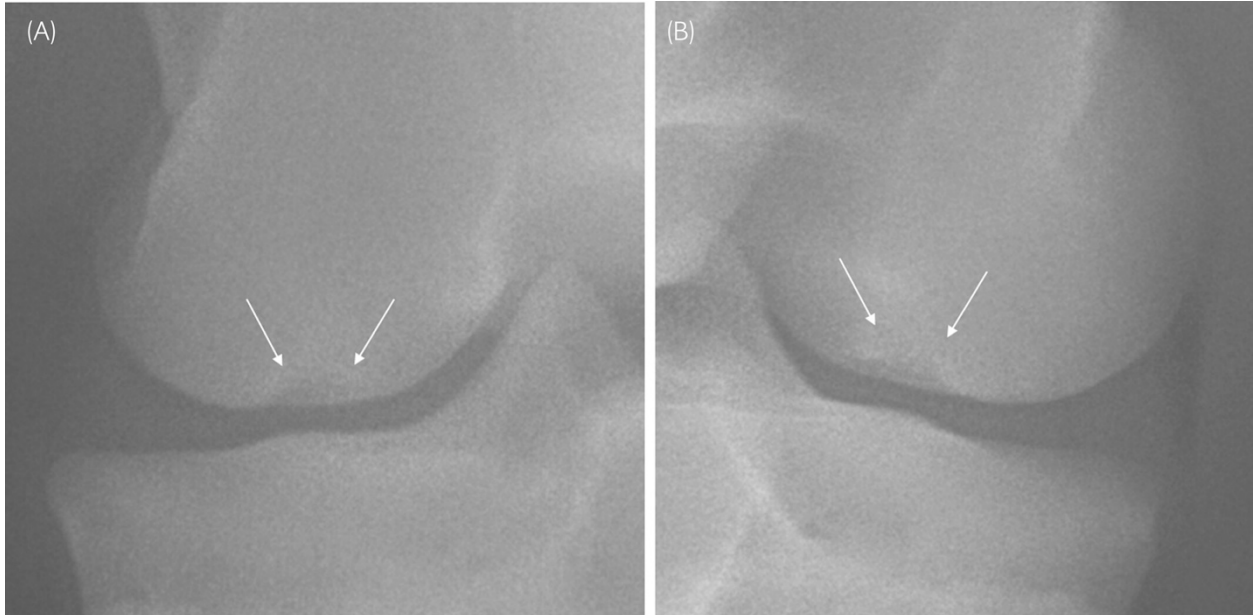


FIGURE 3.3. Radiographs showing the appearance of a Grade 2 subchondral lucency (SCL, white arrows) in the distal aspect of the equine medial femoral condyle (MFC) in a Thoroughbred yearling on CdCr (a) and CdLrCrMO (b) projections. The SCL is a moderate, dome-shaped lucency that extends through the subchondral bone with a depth approaching that of its width (i.e. Grade 2 MFC SCL).

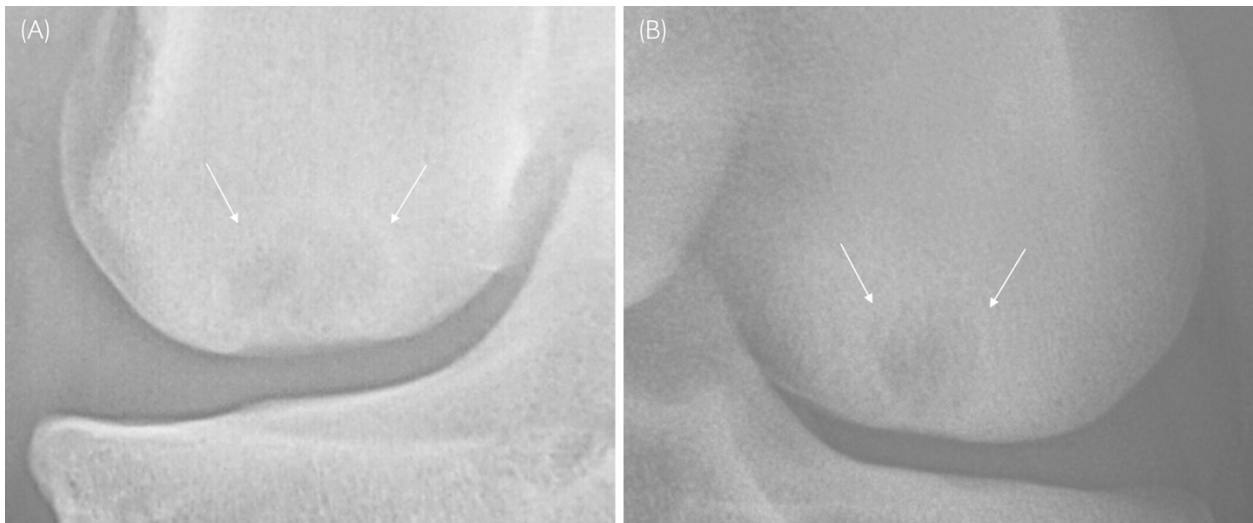


FIGURE 3.4. Radiographs showing the appearance of a Grade 3 subchondral lucency (SCL, white arrows) in the distal aspect of the equine medial femoral condyle (MFC) in a Thoroughbred yearling on CdCr (a) and CdLrCrMO (b) projections. The SCL is a large, spherical or ovoid cystic lesion that communicates with the central articular surface of the condyle (i.e. Grade 3 MFC SCL).

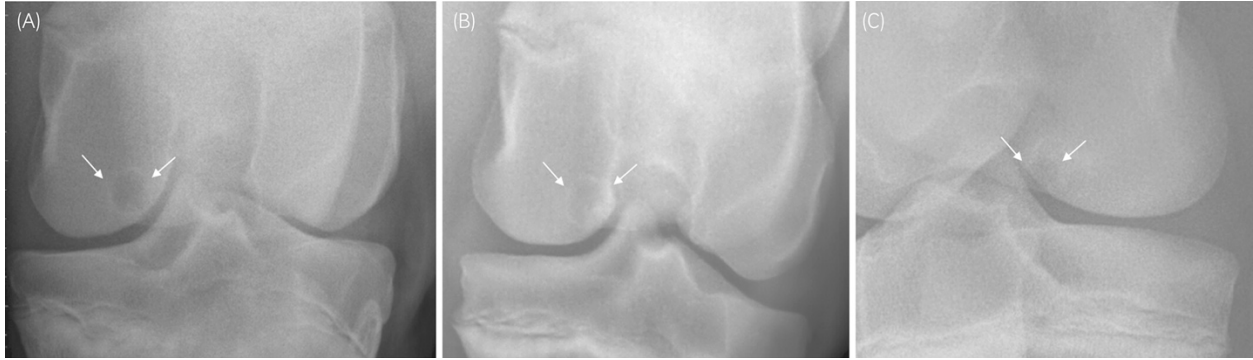


FIGURE 3.5. Radiographs showing spherical or ovoid lucencies in the axial aspect of the equine medial femoral condyle (MFC), adjacent to the medial intercondylar eminence of the tibia, in three different 2-year-old Thoroughbred horses on CdCr (a, b) and CdLCrMO (c) projections. Axial MFC lucencies were recorded and analysed separately from the MFC subchondral lucency grading system because they do not involve the central weightbearing portion of the distal MFC. All three of these horses are Grade 0 MFC SCL for the central distal articular portion of the condyle.

necessary. Radiographs were viewed using DICOM viewing software OsiriX MD and Horos. Observers were blinded to sale and race results during radiograph evaluation.

### 3.3.3 Racing performance

Racing performance data collection continued until the end of the horses' 4-year-old racing season. Racing data was obtained from the Equibase Company LLC. Racing performance was measured via eight outcome variables<sup>14</sup>. Variables relating to race starts included; whether the horse started at least one race by the end of the 4-year-old racing year<sup>8,12</sup>, age at first start and total number of starts<sup>14,15</sup>. Variables relating to prizemoney included total earnings and earnings per start, in United States Dollars (USD)<sup>8,16</sup>. International earnings were converted to USD using the exchange rate on the date of racing. Variables related to calibre of racing performance achieved included; career best start, weighted Listed and Group race starts and Class Performance Index<sup>12</sup>. Best start was a binary outcome for whether a horse started at least one race at Listed or Group level. Weighted Listed and Group race starts was the sum total of weighted starts, including each Listed race (1 point), Group 3 race (2 points), Group 2 race (3 points) and Group 1 race (4 points), respectively. Class Performance Index was a lifetime ratio based on an individual horse's average earnings per start, divided by the average earnings per start of all other horses of the same sex and birth year.

### 3.3.4 *Non-starter follow-up information*

Any horse that did not start at least one race was considered a non-starter. Follow-up information for non-starters was sought via email and telephone communication with the connections of each horse, using a flow-chart conversation framework that was designed to obtain increasing levels of detail while minimising bias (Appendix 3).

### 3.3.5 *Data analysis*

Data analysis was performed via consultation with statisticians using the open-source statistical software package R, version 4.0.2 by RStudio<sup>b</sup>. All reported confidence intervals are 95% and  $\alpha=0.05$  was used as a threshold for significance across all tests. No adjustments to reported p-values were made for multiple tests performed across outcomes.

Agreement between all pairs of observers was assessed using Cohen's kappa with a squared weighting, reported as a group range<sup>17,18</sup>. Krippendorff's alpha for multiple observers with an ordinal method was also used, which is equivalent to the mean of Cohen's kappa between pairs. Unweighted Fleiss' kappa for multiple observers was used to calculate group agreement within each grade of lesion<sup>19</sup>.

Radiological findings were summarised using descriptive statistics, with frequency distributions at the stifle-level by age, by sex and by left and right hindlimb. Radiological findings were summarised at the horse-level by age, using maximum MFC grade per horse. The proportion of horses with Grade >1 MFC SCLs at yearling age compared to the same proportion at two years of age was analysed using McNemar's Chi-squared test with a continuity correction for low frequencies.

Associations between yearling radiological findings and future racing performance were analysed, with each yearling assigned a horse-level categorical variable in the form of "maximum MFC grade". This was the most severe MFC SCL appearance the horse had as a yearling, in either the left or right stifle. To test for a significant relationship between "maximum MFC grade" and the racing outcome; an ANOVA F-

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test (for continuous outcomes) or a likelihood ratio test (for binary and count outcomes) was performed and associated test statistics and p-values were reported.

The associations between “maximum MFC grade” and each racing performance outcome were examined in regression analyses. Specifically, each racing outcome was regressed on the categorical radiological variable “maximum MFC grade” while controlling for horse sex, to adjust for differences in racing opportunities. Binary outcomes, “Started a race” and “Best start” were analysed using logistic regression. The count outcome “Number of starts”, was analysed using negative binomial regression. The remaining race performance variables were analysed using linear regression. The reference value for “maximum MFC grade” in all regressions was set to zero. For the racing outcome variable “started at least one race”, all available horses were used. For other racing performance measures, only horses that started a race were included in the analysis.

Data transformations were used when necessary to better satisfy model assumptions, e.g. normally distributed errors. Earnings related measures required log transformation, with all \$0 earnings and \$0 earnings per start set to \$1. Class Performance Index required log transformation with all CPI of zero set to 0.05, which is half the minimum performance value of 0.1. Weighted Listed and Group starts required square root transformation.

Model estimates and confidence intervals produced by single variable regression analyses were back-transformed to provide estimates that pertained to the original scale of the racing performance data, describing the average value of the outcome for each level of max MFC score.

## **3.4 Results**

### *3.4.1 Study enrolment*

Repository radiographs for 2508 yearlings were used (Appendix 4), which represented 10.9% of the annual foal crop and 36.0% of the 6,963 North American yearlings sold at auction in 2016 (Jockey Club Information Systems Inc.). The age of the study yearlings on the first day of the sale ranged from 458 to

614 days old, with a median of 535 days old. Repository radiographs for 436 2-year-olds were used (Appendix 5), representing 19.7% of the 2-year-olds sold at auction in North America in 2017.

### 3.4.2 Interobserver agreement

Overall ordinal agreement between the four veterinarians evaluating stifle radiographs was  $K\alpha = 0.94$ , (group range weighted  $Ck = 0.88$ – $1.00$ ,  $P 0.004$ – $0.002$ ). Fleiss' kappa for group agreement within each grade of MFC lesion was  $k = 1.00$  for Grade 0 and Grade 3,  $k = 0.57$  for Grade 1 and  $k = 0.62$  for Grade 2.

### 3.4.3 Distribution of radiological findings

MFC findings summarised at the horse-level are presented in Table 3.2. Tables 3.3-3.5 contain the individual MFC findings for all 5016 yearling stifles and 872 2-year-old stifles, the frequency distribution of MFC findings by left and right hindlimbs in each age group, and MFC findings by horse sex (colts/geldings, fillies) in each age group, respectively.

TABLE 3.2. Distribution of the most severe radiological medial femoral condyle subchondral lucency (MFC SCL) grade in the left or right stifle per horse in Thoroughbred sales yearlings and 2-year-olds, where N represents individual horses.

MFC SCL grade	Number of yearlings, N (%)	Number of 2-year-olds, N (%)
Grade 0	2266 (90.4%)	387 (88.8%)
Grade 1	171 (6.8%)	30 (6.9%)
Grade 2	45 (1.8%)	14 (3.2%)
Grade 3	26 (1.0%)	5 (1.1%)
Total	2508	436

TABLE 3.3. Distribution of medial femoral condyle subchondral lucency (MFC SCL) grades in all stifles of yearling and 2-year-old Thoroughbred sales horses, where N represents individual stifles.

MFC grade	Number of yearling stifles, N (%)	Number of two-year-old stifles, N (%)
Grade 0	4720 (94.1%)	811 (93.0%)
Grade 1	214 (4.3%)	39 (4.5%)
Grade 2	55 (1.1%)	17 (1.9%)
Grade 3	27 (0.5%)	5 (0.6%)
Total	5016	872

TABLE 3.4. Distribution of medial femoral condyle subchondral lucency (MFC SCL) grades by left and right hindlimbs in yearlings and 2-year-old Thoroughbred sales horses, where *N* represents individual stifles and the percentage shown is the left vs right hindlimb distribution for each grade.

MFC grade	Number of yearling stifles, N (% left vs right)		Number of two-year-old stifles, N (% left vs right)	
	Left	Right	Left	Right
Grade 0	2388 (50.6%)	2332 (49.4%)	412 (50.8%)	399 (49.2%)
Grade 1	92 (43.0%)	122 (57.0%)	20 (51.3%)	19 (48.7%)
Grade 2	19 (34.5%)	36 (65.5%)	3 (17.6%)	14 (82.4%)
Grade 3	9 (33.3%)	18 (66.7%)	1 (20.0%)	4 (80.0%)
Total	2508	2508	436	436

TABLE 3.5. Distribution of radiological medial femoral condyle subchondral lucency (MFC SCL) grades in the stifles of yearling and 2-year-old Thoroughbred sales horses by colts/geldings versus fillies, where *N* represents individual stifles.

MFC grade	Yearling stifles, N (%)		Two-year-old stifles, N (%)	
	Colts/Geldings	Fillies	Colts/Geldings	Fillies
0	2405 (94.5%)	2315 (93.7%)	426 (93.8%)	385 (92.1%)
1	106 (4.2%)	108 (4.4%)	21 (4.6%)	18 (4.3%)
2	25 (1.0%)	30 (1.2%)	5 (1.1%)	12 (2.9%)
3	10 (0.4%)	17 (0.7%)	2 (0.4%)	3 (0.7%)
Total	2546 (100%)	2470 (100%)	454 (100%)	418 (100%)

#### 3.4.4 Paired yearling and 2-year-old sales radiographs

Radiographs were available at both the yearling sale and a 2-year-old sale for 422 horses, enabling assessment of any change in the appearance of MFC SCLs between one and two years of age. Results are presented in Table 3.6.

Of the yearling Grade 1 MFC SCLs, by two years of age 35.5% were a Grade 0 and 45.2% were a Grade 1 (11/31 and 14/31, respectively). Thus, 80.7% of yearling Grade 1 MFC SCLs had either resolved or remained static at 2-year-old sale presentation. Overall, 19.4% of yearling Grade 1 MFC SCLs progressed to a Grade 2, i.e. one in five yearlings with a Grade 1 MFCL (6 of 31). When analysed by hindlimb side, 14% of left stifle and 24% of right stifle Grade 1 MFC SCLs progressed to a Grade 2 (2/14

and 4/17, respectively). For yearlings with Grade 2 lesions, 2 (20%) improved to Grade 1, 6 (60%) remained static and 2 (20%) progressed to a Grade 3. For yearlings with Grade 3 lesions, 2 (66%) improved to a Grade 2 and 1 (33%) remained static. McNemar’s test did not indicate a significant difference in the proportion of yearling and 2-year-old horses with at least one non-normal MFC ( $P = 0.1$ ).

TABLE 3.6. Distribution of medial femoral condyle subchondral lucency (MFC SCL) grades per stifle at 2-year-old sales (columns), relative to each stifle’s respective MFC SCL grade at the yearling sale (rows), for 422 horses with paired yearling and 2-year-old Thoroughbred sales radiographs. Shaded values represent grades that remained unchanged between sales.

MFC grade at yearling sale	MFC Grade at 2-year-old sale, N (%)				Total stifles n=844
	Grade 0	Grade 1	Grade 2	Grade 3	
Grade 0	775 (96.9%)	21 (2.6%)	2 (0.2%)	2 (0.2%)	800 (100%)
Grade 1	11 (35.5%)	14 (45.2%)	6 (19.4%)	0 (0%)	31 (100%)
Grade 2	0 (0%)	2 (20%)	6 (60%)	2 (20%)	10 (100%)
Grade 3	0 (0%)	0 (0%)	2 (66.7%)	1 (33.3%)	3 (100%)
Total stifles	786	37	16	5	844

### 3.4.5 Axial MFC lucencies

Seven yearlings had lucencies located in the axial aspect of the MFC, adjacent to the medial intercondylar eminence of the tibia (7/2508, 0.3%). These ranged in appearance from irregular lucencies to well-defined spherical or ovoid cystic lesions. One yearling was bilaterally affected. In all 7 yearlings, the affected stifles had a Grade 0 appearance for the central articular aspect of the distal MFC. One yearling had a Grade 1 MFC SCL in the contralateral stifle, the rest had Grade 0 central MFCs bilaterally. Two of these yearlings presented again at 2-year-old sales and the axial MFC lucencies persisted in both cases. No new axial MFC lucencies were seen in 2-year-olds that did not have them as yearlings.

### 3.4.6 Associations with racing performance

#### *i. Started at least one race*

Overall, 85% of study yearlings started at least one race (2119/2508) and 15 % had not raced by the end of their 4-year-old year (389/2508). The probability of yearlings with a Grade 3 MFC SCL starting a race was lower than the probability of yearlings with Grade 0-2 MFCs starting a race (77.6% vs.

84.3-91.3%) (Table 3.7). However, logistic regression analysis showed no statistically significant difference in the probability of starting a race for any category of MFC SCL ( $P = 0.3$ ).

TABLE 3.7. The distribution of radiological medial femoral condyle subchondral lucency (MFC SCL) grades for all study horses at the Thoroughbred yearling sales, by most severe finding per horse, with the accompanying proportion of horses that started at least one race by the end of their 4-year-old racing season and the probability of starting a race via logistic regression analysis controlling for horse sex (colt/gelding, filly).

Yearling MFC grade	Proportion raced	Probability of starting a race, controlled for horse sex	SE for probability	95% Confidence limits	P value	Number raced	Total yearlings, N=2508
0	84.2%	0.843	0.008	0.827 – 0.858	NA	1909	2266
1	87.1%	0.872	0.026	0.813 – 0.914	0.314	149	171
2	91.1%	0.913	0.042	0.790 – 0.967	0.203	41	45
3	76.9%	0.776	0.081	0.582 – 0.896	0.352	20	26

Of the entire registered North American foal crop born in 2015, 68.4% (15,770/23,043) had started a race by the end of their 4-year-old year and an additional 0.7% had their first race start aged five years or older (159/23,043) (Jockey Club Information Systems Inc.).

*ii. Follow-up of non-starters*

Follow-up information was obtained for 166 of 389 horses that never started a race (43% of non-starters). For 84 non-starters, the follow-up was partial, i.e. confirmation was given that the horse never raced but the reason for not starting was unknown, or confirmation was given that the horse was a broodmare but the reason for never racing was unknown, or the horse was identified as exported from the United States and the racing history was unknown and not accessible. Follow-up information was complete for 82 non-starters. Of these 82 horses; 66 did not make a race start for reasons related to performance, 10 did not race due to fatal or near fatal accidents unrelated to performance, 5 did not race due to medical events requiring euthanasia and 1 was purchased from the yearling sale for a non-racing discipline.

Of the 66 non-starters with follow-up that indicated a performance-related reason for not racing: 45 involved lameness, 14 were deemed to have insufficient athletic talent, 3 had upper respiratory tract

dysfunction, 1 did not start for behavioural reasons, 1 had severe exertional rhabdomyolysis, 1 had cervical vertebral malformation (wobbler) and 1 had recurrent exercise-induced pulmonary haemorrhage.

Of the 45 non-starters that did not race due to an identified lameness, the clinical diagnosis involved the stifle region in 4 horses: in 3 cases the specific structure affected within the stifle was unknown and 1 horse was kicked in the stifle and had a persistent swelling at the site of external trauma. None of these 4 cases mentioned MFC SCL as a suspected cause of lameness. All MFCs in these 4 horses had a Grade 0 appearance on yearling radiographs and none had 2-year-old sales radiographs. In all 45 non-starters that did not race due to known lameness, there was 1 horse with a Grade 2 MFC lucency on its yearling and 2-year-old radiographs. This horse did not race due to a sesamoid fracture.

In the 14 non-starters that did not race because they were deemed too slow, 1 had a Grade 3 MFC SCL as a yearling. However, the owner specifically mentioned that this cystic lesion had been monitored during training and it had improved radiologically and never caused a clinical problem, although this was not confirmed by the authors. The other 13 horses that did not start due to lack of ability all had Grade 0 MFCs.

Of the 84 non-starters with partial follow-up where the reason for not racing was unknown and the 223 non-starters with no follow-up information, 5 had a Grade 3 MFC SCL and 3 had a Grade 2 MFC SCL. The overall distribution of MFC findings at the horse level in horses that raced compared to non-starters is presented in Table 3.8.

TABLE 3.8. Comparison of the number of horses that raced versus the number of horses that did not start at least one race by the end of their 4-year-old season, for each radiological grade of medial femoral condyle subchondral lucency (MFC SCL) in Thoroughbred sales horses as yearlings, by most severe finding per horse.

Yearling MFC grade	Number of horses that raced	Number of horses that did not start a race
0	1909	357
1	149	22
2	41	4
3	20	6
Total	2119	389

*iii. Age at first race start*

Of the 2119 yearlings that raced, the mean age at first start was 1006 days old, i.e. 2 years and 9 months (median 969 days, range 687-1725 days, lower quartile (Q1) 882 days, upper quartile (Q3) 1102 days). There was no significant difference in the average age at first start between horses with any grade of MFC SCL ( $P = 0.3$ ) (Table 3.9).

TABLE 3.9. Mean age at first race start in days old for horses with each radiological grade of medial femoral condyle subchondral lucency (MFC SCL) at Thoroughbred sales as yearlings, by most severe finding per horse, controlling for sex of the horse.

Yearling MFC grade	Mean number of days old at first start	SE	Confidence limits
0	1004	3.831	997-1012
1	1021	13.700	995-1048
2	981	26.116	930-1033
3	1056	37.400	982-1129

*iv. Number of race starts*

The horses that raced had an average of 12.3 starts by the end of the study period (median 11 starts, range 1-49 starts, Q1 6, Q3 17). There was no significant difference in the average number of starts between horses with any MFC grade ( $P = 0.1$ ) (Table 3.10).

TABLE 3.10. Mean number of race starts to the end of the 4-year-old racing season for horses with each radiological grade of medial femoral condyle subchondral lucency (MFC SCL) at Thoroughbred sales as yearlings, by most severe finding per horse via negative binomial regression analysis controlling for horse sex (colt/gelding, filly).

Yearling MFC grade	Mean number of starts at 2-4yo	SE	Confidence limits
0	12.28	0.194	11.91-12.67
1	11.31	0.645	10.12-12.65
2	14.90	1.584	12.10-18.35
3	11.71	1.818	8.64-15.87

*v. Earnings*

Median total earnings were \$32,253 per horse (mean \$69,445, range \$0-\$3,750,000, interquartile range (IQR) \$9,166-\$73,535) for the 2119 horses that raced. Median earnings per start for the same horses were \$2,674 (mean \$5856, range \$0-\$268,614, IQR \$1,112-\$5,901). There was no significant association

with total earnings ( $P = 0.8$ ), or earnings per start ( $P = 0.9$ ), for any MFC grade. Back-transformed estimates and confidence intervals for average earnings and earnings per start are provided in Tables 3.11 and 3.12.

TABLE 3.11. Back-transformed mean total earnings in United States Dollars to the end of the 4-year-old racing season for horses with each radiological grade of medial femoral condyle subchondral lucency (MFC SCL) at Thoroughbred sales as yearlings, by most severe finding per horse via linear regression analysis with log transformation and controlling for horse sex (colt/gelding, filly).

Yearling grade	MFC	Estimated average total earnings (USD)	SE	Confidence limits
0		\$15,311	985	13497-17369
1		\$11,935	2745	7602-18737
2		\$16,679	7313	7059-39410
3		\$13,513	8485	3944-46293

TABLE 3.12. Back-transformed mean total earnings per start in United States Dollars to the end of the 4-year-old racing season for horses with each radiological grade of medial femoral condyle subchondral lucency (MFC SCL) at Thoroughbred sales as yearlings, by most severe finding per horse.

Yearling grade	MFC	Estimated average earnings per start (USD)	SE	Confidence limits
0		\$1,803	90	1634-1989
1		\$1,597	286	1124-2270
2		\$1,566	535	801-3061
3		\$1,665	815	638-4348

#### vi. Race caliber

The distribution of Career Best Starts for all 2119 horses that raced was as follows: 84% raced but did not reach Listed or Group level. 4.8% had their highest start in a Listed race. 4.2% had their highest start in a Group 3 race. 3.1% had their highest start in a Group 2 race. 4.3% started a Group 1 race. No significant associations were found between any MFC grade and Career Best Start ( $P = 0.5$ ), nor Weighted Listed and Group Starts ( $P = 0.1$ ). For Class Performance Index, the median CPI in 1,996 horses for which it was available was 0.70 (mean 1.47, range 0-88). There were no significant associations between CPI and any MFC grade ( $P = 0.3$ ).

*vii. Racing performance for axial MFC lucencies*

Of the 7 yearling sale horses with axial MFC lucencies, 6 started at least one race and all 6 placed, including 4 who won at least one race. 5 first raced as 2-year-olds and 1 was exported as a 2-year-old then subsequently raced. The age at first race ranged from 812-1003 days. The total number of starts ranged from 6 – 21 races. Total earnings ranged from \$6,330-\$126,826 and earnings per start ranged from \$1,055-\$14,092. 4 had available CPIs, ranging from 0.23-3.42. Follow-up communication for the 1 horse with an axial MFC lucency that did not race revealed that the filly was involved in a fatal accident during breaking in.

*viii. Racing performance for 2-year-old Grade 3 MFC SCLs*

The above racing performance outcomes refer to yearling sales radiograph findings. The 5 horses with Grade 3 MFC SCLs at 2-year-old sales all went on to start at least one race. Their average number of starts was 15.2, average number of wins was 2.2, median earnings per start were \$1717 (mean \$2928) and median total earnings were \$36,064 (mean \$35,442).

### **3.5 Discussion**

This study has documented the prevalence of MFC SCLs in a convenience sample of yearling and 2-year-old Thoroughbreds presented for sale at public auctions. It is part of the largest sales radiology study to date and the first time MFC SCLs have been studied longitudinally in horses presenting for sale at one and two years of age.

This is the first study to sample from all repository radiographs using consignors' permission for enrolment and to subsequently analyse associations between radiological findings and racing performance. Obtaining research consent for 74% of yearlings with repository radiographs and 78% of eligible 2-year-olds exceeded expectations and reflected a strong desire within the Thoroughbred industry to add to the evidence base regarding the significance or otherwise of MFC SCLs in sales horses. Previous sales studies have sampled up to 47% of radiographs<sup>4-6</sup>. The majority of previous sales radiology research has retrospectively utilised sales radiographs acquired by a limited number of veterinary practices and thus a

select sample of consignors<sup>8,12,13</sup>. The present study enrolled horses from 71 different yearling consignors and 45 2-year-old consignors, thereby sampling a range of management practices and sales preparation systems. Earlier studies did not all include the minimum three radiographic projections that are necessary for MFC evaluation, which were included in this study<sup>4,5,13,20</sup>.

The prevalence of MFC SCLs is similar to values reported in previous Thoroughbred sales radiology work, bearing in mind differences in the classification of lucency size and appearance. Grade 3 MFC SCLs, i.e. large cystic lesions, are reported in 1.0% of yearlings and 1.1% of 2-year-olds, which is consistent with previous reports of 0.86%<sup>8</sup> to 1.7%<sup>5</sup> in yearlings and 0.84% in 2-year-olds<sup>6</sup>. Previously reported prevalences of MFC SCLs most equivalent to the Grade 2 defined in this study range from 0.93%<sup>8</sup> to 3.9%<sup>4</sup> in yearlings; Grade 2 MFC SCLs are reported here in 1.8% of yearlings and 3.2% of 2-year-olds. MFC SCLs most equivalent to the Grade 1 defined in this study were previously reported in 6.4%<sup>5</sup> and 10.7% of yearlings<sup>4</sup>. The current results show Grade 1 MFC SCLs in 6.8% of sales yearlings and 6.9% of sales 2-year-olds.

The right hindlimb predilection for Grade 2 and 3 MFC SCLs in both yearlings and 2-year-olds is consistent with previous reports<sup>3,4,5,8,21,22</sup>. This study shows approximately two-thirds of yearling Grade 2 and Grade 3 MFC SCLs were in the right stifle (65.5% and 66.7%, respectively). In 2-year-olds, closer to 80% of Grade 2 and 3 MFC SCLs were in the right stifle (82.4% and 80.0%, respectively). The reasons for this predisposition are unknown, but it may be due to some form of low-grade stress to that particular joint surface related to horses' direction of movement.

Results show similar percentages of each grade of MFC SCL between colts or geldings and fillies, both in yearlings and 2-year-olds, as reported previously<sup>14</sup>. This study controlled for horse sex in all regression analyses of racing performance because there are differences in racing performance and racing opportunities between colts, fillies and geldings; whereby horses with breeding value are often retired earlier than their gelding counterparts.

The sample of yearlings studied represents 10.9% of all registered North American foals born in 2015 and 36% of all yearlings sold at auction in North America in 2016. The prevalence of MFC findings

refers to the sales population. The prevalence in the entire population is likely to be greater due to sales selection pressure. This is supported by studies including both sale and non-sale yearling radiographs that report a prevalence of 4.3-7.8% for MFC SCLs <sup>3,21</sup>.

The grading scale used in this study was modified from previously published systems <sup>22,23</sup>. A flattened radiological appearance of the MFC, both in terms of whether it is real and whether it is important, has been debated. Projection angle, radiographic technique and hindlimb stance affect MFC shape on resulting images <sup>19</sup>. Flattening of the distal contour of the MFC was previously recorded in 52% of 1505 Thoroughbred sales yearlings <sup>4</sup>, suggesting a finding affecting approximately half the population is unlikely to be pathological. A recent study considered MFC flattening without deeper sclerosis to be normal and did not include it as an abnormality <sup>3</sup>. In the current study, flattening of the distal articular contour of the MFC, in the absence of radiolucent subchondral change, was considered to be within normal limits (Grade 0). Of 800 Grade 0 yearling MFCs, 97% remained Grade 0 as 2-year-olds.

Education in application of the radiological grading system was important. Following a period of training in the use of the grading scale, its application was shown to have excellent interobserver agreement ( $Ka=0.94$ ). Analysis demonstrated perfect agreement ( $k=1.00$ ) in identifying Grade 0 MFCs and Grade 3 MFC SCLs and occasional differences in categorising Grade 1 and 2 MFC SCLs ( $k=0.57$  and  $k=0.62$ , respectively) <sup>24</sup>. Variation for Grades 1 and 2 may arise from the use of a 3mm depth threshold, where borderline lucencies are classified according to an observer's opinion of the overall appearance as either a shallow, crescent-shaped or a moderate, dome-shaped lucency, in addition to the exact depth. Variability in radiopacity or sclerosis surrounding SCLs was intentionally not included in this grading system <sup>3</sup>. The image zoom function was used to increase measurement accuracy using DICOM viewing software tools. Horses were all Thoroughbreds of similar age and size; as opposed to different breeds, or the inclusion of foals or weanlings, where it becomes necessary to use a ratio of affected MFC distance rather than absolute measurements <sup>5</sup>.

Lucencies located in the axial aspect of the MFC, adjacent to the medial intercondylar eminence of the tibia, were categorised separately from SCLs in the central, weightbearing portion of the MFC because

in the authors' experience these have a different risk profile. These were referred to as lucencies rather than cystic lesions because there tends not to be an obvious communication with the joint and the histological features are unknown. Axial lucencies are reported to occur at an earlier age than central MFC lucencies and their documented resolution in 8 of 8 Thoroughbred foals by 12 months of age suggests that many may resolve prior to yearling sales<sup>3</sup>. The current results support evidence that some do persist and present as spherical or ovoid axial lucencies in yearlings and 2-year-olds<sup>14</sup>. No new axial MFC lucencies developed in 2-year-olds that were not already present at yearling age in the horses studied. None of the 7 affected horses in this study had reduced racing performance because of an axial lucency. It is recognised that axial lucencies within the femoral condyles can occur in association with proximal tibial SCLs<sup>25</sup>, but on their own they do not appear to be a clinical concern.

The time period between North American yearling and 2-year-old sales ranges from 6-9 months. Although numbers of pinhooked yearlings with Grade 2 and 3 MFC SCLs are small, the results document evidence of a static or one grade improvement in 8 of 10 yearling Grade 2 MFC SCLs by 2-year-old sales, and a one grade improvement in 2 of 3 yearling Grade 3 MFC SCLs. The study could not address whether any yearlings with Grade 3 MFC SCLs may have had surgical intervention post-sale that contributed to their ability to race<sup>11</sup>.

There are two scenarios in which a horse intended for sale may be subject to negative selection pressure for reasons related to MFC SCLs. The first is the onset of clinical lameness, either between birth and yearling age, or during yearling or 2-year-old sale preparation. It is recommended practice that horses intended for sale are withdrawn if they develop an overt lameness. Horses are assessed at the walk at yearling sales, whereas 2-year-olds undergo a timed presale gallop after a minimum of 90 days of training. Analgesic use could not be accounted for in this study.

The second scenario occurs after acquisition of screening or repository radiographs, when the consignor receives a radiology report. Severe lesions that may affect sale value can result in withdrawal from sale prior to submission of radiographs. The study results reflect the distribution of lesions only in horses entered for sale and may underestimate the prevalence of severe lesions in non-sale Thoroughbreds.

The associations with racing performance reported in this study should be applied to horses presented for sale at auction and are not applicable to horses with clinical lameness at yearling or 2-year-old age attributed to MFC lesions.

The number of yearlings with a Grade 3 MFC SCL is relatively small at 26 horses. However, this equals the highest number of MFC cystic lesions studied previously<sup>4</sup>. The current results show that fewer yearlings with Grade 3 SCLs go on to race, but in those that do race there is no evidence of worse performance compared to unaffected peers. Despite not meeting the threshold for statistical significance, there is a clinically relevant difference in that Grade 3 yearlings had a 78% probability of racing (20 of 26), which was lower than the overall proportion of 85% yearlings that raced (2119 of 2508) and lower than the 84% probability of horses with a Grade 0 MFC SCL racing (1909 of 2266).

The goal of this research was to translate results into practical advice that veterinarians can give to consignors and prospective purchasers. Veterinary advice must consider a client's particular needs, including his or her intentions for a given horse and personal level of risk tolerance, because individual client's needs will never be identical and nor will be the decisions different clients make based on certain radiological findings. The veterinarian's role is to accurately identify lesions and provide evidence-based information as to the associated risk. The provision of objective data will increase consignors' and purchasers' ability to make evidence-based decisions in sales horses. For example, if a prospective buyer understands that the presence of a Grade 3 MFC SCL in a sales yearling increases the horse's probability of not making it to the races from 16% to 22%, i.e. from a 1 in 7 to a 1 in 5 chance of not racing, and it may or may not need treatment to do so, it is then up to the client to decide the weight of importance that finding carries for a given purchase.

Of arguably greater importance than the identification of severe lesions is the documentation of common, mild findings that have no detrimental impact on racing performance and a low incidence of progression. This applies strongly to the Grade 1 MFC SCL results. The least positive outcome of the repository system is when minor radiological findings of no consequence to the horse result in failed sales, or sale prices that do not reflect the horse's value, when the horse may go on to perform exceptionally well.

The intention of undertaking a large, sales-based study was to provide veterinarians with objective evidence that frees them from being unnecessarily critical about certain radiological findings in young Thoroughbreds.

The main limitations of this study arise from its use of convenience sampling of horses presented for sale at public auction. Results are likely to underestimate the prevalence of severe, clinical MFC lesions in the non-sale population. The study design could not address unknown losses between weanling screening and yearling age, and between yearling and 2-year-old age in horses intended for resale. However, this also ensures that the findings are specifically applicable in the repository environment to future populations of horses that have made it to the sales. Some effects of sex may be missed by not analysing the performance of entire males and geldings separately. Grouping the male subsets together was necessary due to gelding occurring at a range of ages after commencement of racing.

### **3.6 Conclusions**

Overall, MFC SCLs of varying grades were observed in 9.6% of Thoroughbred yearlings and of 11.2% of 2-year-olds. Shallow, crescent-shaped lucencies (Grade 1) were the most common type of MFC SCL in both age groups. The majority of Grade 1 MFC SCLs seen in sales yearlings were no worse at 2-year-old sale presentation; 19.4% of yearling Grade 1 MFC SCLs progressed to a Grade 2 deeper, dome-shaped lucency by two years of age and the rest remained static or resolved. It was also possible for Grade 2 and 3 MFC SCLs to improve one grade in appearance between yearling and 2-year-old sales.

Although fewer sales yearlings with a Grade 3 MFC SCL made it to the races, no statistically significant difference was found. If a sales yearling with a Grade 3 MFC SCL did race, there was no evidence of worse racing performance compared to unaffected peers. All horses that presented at a 2-year-old sale with a Grade 3 MFC SCL raced, though this sample size was small. Axial MFC lucencies did not affect racing performance.

CHAPTER 4 – RADIOLOGICAL FINDINGS IN THE PROXIMAL SESAMOID BONES OF  
YEARLING AND 2-YEAR-OLD THOROUGHBRED SALES HORSES: PREVALENCE,  
PROGRESSION AND ASSOCIATIONS WITH RACING PERFORMANCE<sup>°</sup>

#### 4.1 Summary

Radiological findings in the proximal sesamoid bones (sesamoids) are a persistent source of controversy at Thoroughbred sales, due to inconsistent classification and conflicting assignment of potential clinical importance.

This was a prospective cohort study using an enrolled sample. The objectives were to 1) define the prevalence of sesamoid findings on sales repository radiographs in yearling and 2-year-old Thoroughbreds, 2) identify any associations between sesamoid findings and future racing performance and 3) monitor changes in sesamoid findings between yearling and 2-year-old sales in horses that present for sale at both ages.

Horses were enrolled from a 2016 yearling sale and five 2017 two-year-old sales with consignor permission. Radiological findings relating to sesamoid vascular channel appearance, abaxial contour changes and sesamoid fragments were examined. Associations between sesamoid findings and racing performance from two to four years of age were examined using multivariate regression analyses. Clinical follow-up was sought to ascertain why horses that did not race never started.

A total of 2508 yearlings and 436 two-year-olds were included for evaluation. Interobserver agreement using the new grading system was substantial. Yearling findings associated with a significantly reduced probability of starting a race were: Grade 3 vascular channels in forelimb sesamoids (0.52,  $P <$

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<sup>°</sup> Peat, F. J., Kawcak, C. E., McIlwraith, C. W., Keenan, D. P., Berk, J. T., & Mork, D. S. (2024). Radiological findings in the proximal sesamoid bones of yearling and 2-year-old Thoroughbred sales horses: Prevalence, progression and associations with racing performance. *Equine Veterinary Journal*, (Online). <https://doi.org/https://doi.org/10.1111/evj.14051>. *Included with permission of journal Editor-In-Chief.*

0.001, 95% Confidence Interval (CI) 0.37-0.67), abaxial new bone in forelimb sesamoids (0.62,  $P = 0.01$ , 95% CI 0.49-0.73), apical or abaxial fragments in forelimb sesamoids (0.55,  $P = 0.005$ , 95% CI 0.37-0.72). For affected horses that did race, Grade 3 vascular channels in forelimb sesamoids were associated with fewer race starts (9.9 starts,  $P = 0.03$ , 95% CI 8.0-12.2) and Grade 3 vascular channels in hindlimb sesamoids were associated with a delayed start to racing careers (54 days,  $P = 0.01$ , 95% CI 20-89). Abaxial new bone in forelimb sesamoids was associated with a 54% reduction in total earnings ( $P = 0.003$ , 95% CI 24-72) and a 46% reduction in earnings per start ( $P = 0.002$ , 95% CI 21-64). Abaxial concavity occurred predominantly in yearling medial forelimb sesamoids, had no impact on racing performance and mostly resolved by two-year-old sale.

Grade 3 vascular channels, forelimb sesamoid abaxial new bone and forelimb sesamoid fragments are important findings in sales repository radiology. The new scale assigns a numerical grade for vascular channel appearance that matches the number of enlarged vascular channels evident in a given sesamoid. Abaxial contour changes, when present in sesamoids that are Grade 0 for vascular channels, are noted separately as either abaxial new bone or abaxial concavity. Fragments are also noted and interpreted separately.

## **4.2 Introduction**

Variability in the radiological appearance of bony canals or channels within the equine proximal sesamoid bones (sesamoids) has been termed “sesamoiditis”<sup>18,19</sup>. These channels carry branches of the palmar or plantar digital arteries<sup>20</sup> and alterations in vascular channel number, size and shape are considered possible indicators of pathological change to the bone itself or to the adjacent suspensory ligament branch<sup>34,35</sup>. Variable radiological grading schemes and the enhanced resolution afforded by digital radiography have led to inconsistent interpretation of sesamoid findings on presale radiographs and their impact on future racing success has been controversial<sup>12,13,22,23</sup>. Previous research concluded that the appearance of some radiological sesamoid changes in sales yearlings warranted further investigation if sufficient numbers of affected yearlings could be studied<sup>12</sup>.

The aim of this study was to objectively evaluate the radiological appearance of the sesamoids in a large number of sales yearlings and two-year-olds; to define the range and prevalence of sesamoid findings on repository radiographs and to analyse associations with racing performance. In horses with paired sales radiographs, the study was designed to track the changes in sesamoid findings between one and two years of age in horses that presented for sale at both ages. The intention was to determine which sesamoid findings should be regarded as an acceptable appearance at a given age in sales horses, and to determine which sesamoid findings are associated with an evidence-based risk of reduced performance.

### **4.3 Methods**

#### *4.3.1 Study design*

A prospective cohort study was conducted using an enrolled sample of yearling Thoroughbred horses. Researchers sought consent for study inclusion from consignors of all horses at the 2016 Keeneland September Yearling Sale in Kentucky and from the five major subsequent North American two-year-old Thoroughbred sales in 2017: Fasig-Tipton Gulfstream, Fasig-Tipton Maryland, Ocala Breeders' Sales (OBS) March, OBS Spring and OBS June. Horses for which study consent was granted at the yearling sale required new research consent from their respective two-year-old consignors to be eligible for two-year-old sale radiograph inclusion. Enrolled horses' radiographs were downloaded in DICOM format after the completion of each sale for confidential evaluation. For any horse that presented at more than one two-year-old sale, radiographs from the latest sale were used as this was considered the eventual point of two-year-old sale. Veterinary letters containing mandatory presale surgery declarations were collected alongside radiographs when present.

#### *4.3.2 Radiological evaluation*

Images evaluated consisted of four fetlock radiographs for each of four limbs per horse, acquired as part of the sales repository protocol: dorso15°-palmar/plantar (DP), dorso15°medial30°-palmarolateral oblique (DMPLO), dorso15°lateral30°-palmaromedial oblique (DLPMO) and lateromedial (flexed or standing LM) projections. The radiological appearance of each of the eight proximal sesamoid bones per

horse was assigned one of six categories according to the definitions in Table 4.1. Example radiographs are depicted in Figure 4.1 and Figure 4.2.

TABLE 4.1. Grading system for the radiological appearance of vascular channels and other abaxial border changes in the proximal sesamoid bones. †Grade 2 can include one very enlarged vascular channel with a width equivalent to the combined width of two enlarged vascular channels, i.e.  $\geq 4\text{mm}$  total width. ‡Grade 3 can include one or two very enlarged vascular channels with a total width equivalent to the combined width of three enlarged vascular channels, i.e.  $\geq 6\text{mm}$  total enlarged channel width.

Radiological appearance of the proximal sesamoid bone	Definition
Grade 0 vascular channels and no abaxial contour change	The sesamoid contains any number of vascular channels that are less than 2mm wide for at least two-thirds of their visible length. The sesamoid also has no abaxial new bone or abaxial margin concavity.
Grade 1 vascular channels	One vascular channel that is $\geq 2\text{mm}$ wide for more than one third of its visible length. Any number of vascular channels $<2\text{mm}$ wide.
Grade 2 vascular channels	Two vascular channels that are $\geq 2\text{mm}$ wide for more than one third of their visible length <sup>†</sup> . Any number of vascular channels $<2\text{mm}$ wide.
Grade 3 vascular channels	Three or more vascular channels that are $\geq 2\text{mm}$ wide for more than one third of their visible length <sup>‡</sup> . Any number of vascular channels $<2\text{mm}$ wide.
Abaxial new bone only	New bone formation on the abaxial margin of the sesamoid, in the absence of any enlarged vascular channels.
Abaxial margin concavity only	A distinct radiolucent concave defect on the abaxial margin of the sesamoid, in the absence of any enlarged vascular channels.

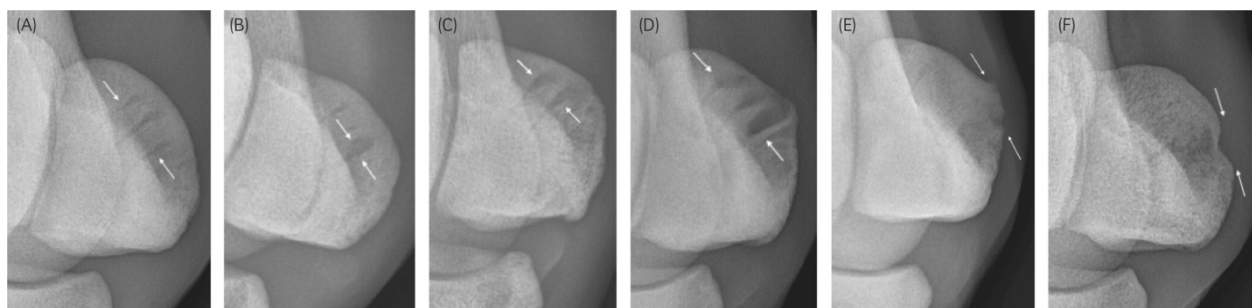


FIGURE 4.1. (A) Grade 0 vascular channel appearance: no enlarged vascular channels and no abaxial contour change. (B) Grade 1 vascular channel appearance: one enlarged vascular channel. (C) Grade 2 vascular channel appearance: two enlarged vascular channels. (D) Grade 3 vascular channel appearance: three or more enlarged vascular channels. The presence of enlarged vascular channels supersedes abaxial new bone for grading purposes. (E) Abaxial new bone, in the absence of any enlarged vascular channels. (F) Abaxial margin concavity, in the absence of any enlarged vascular channels.

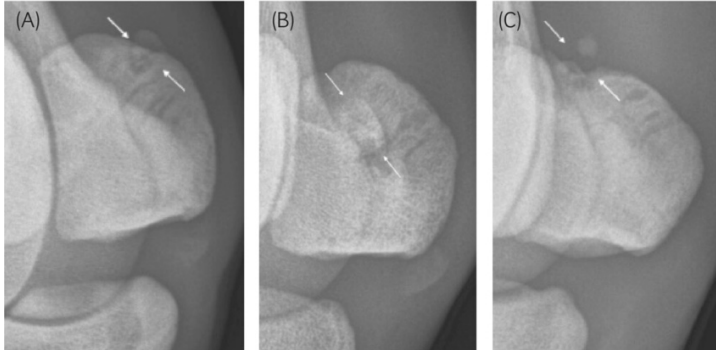


FIGURE 4.2 (A and B) Abaxial sesamoid fragmentation. (C) Apical sesamoid fragmentation, following partial arthroscopic removal before sale.

Measurements for vascular channel grading were made on the respective oblique projection that highlights a given sesamoid<sup>22</sup>. The image zoom function was used to enable accurate placement of the measurement tool on the edges of the channels. To be categorised as “abaxial margin concavity” or “abaxial new bone” only, a sesamoid was required to have no enlarged vascular channels. Any sesamoid with vascular channel enlargement as well as either a radiolucent concavity or new bone on the abaxial margin was categorised based on the number of enlarged vascular channels.

Fragments involving the apical and abaxial margins of the proximal sesamoid bones were recorded separately from the vascular channel and abaxial contour categories; i.e. sesamoid bones with fragmentation were categorised based on the appearance of the vascular channels as for all other sesamoid bones, then an additional record was made of the presence and size of any apical or abaxial fragment. Basilar fragments were not recorded for the purposes of this study, thus any sesamoids with fragmentation of the basilar margin were placed accordingly in one of the above six categories.

The majority of radiographs were read by two veterinarians with 18 and 10 years’ experience in repository radiology, respectively. Prior to evaluating the study radiographs, all four involved veterinarians underwent a period of observer training during which they discussed the grading system and applied group consensus to a sample of images. Each observer then independently evaluated the same set of four radiographs for each of forty sesamoids to test interobserver agreement. During evaluation of the study images proper by individual observers, questionable findings were discussed and graded via consensus,

which is consistent with the approach used in previous work and mirrors the collegial discussion that often takes place in the repository setting<sup>13</sup>.

Radiographs were viewed using DICOM viewing software OsiriX MD and Horos. Observers were blinded to sale and race results during radiograph evaluation.

#### *4.3.3 Racing performance*

Racing performance data collection spanned until the end of the horses' four-year-old racing season. Racing data were provided by Equibase Company LLC<sup>36</sup>. Racing performance was measured using eight outcome variables that have been shown to be most common in assessing racing performance<sup>37</sup>. Three variables relating to race starts included whether the horse started at least one race by the end of their four-year-old year<sup>12</sup>, age at first start and total number of starts. Two variables relating to prizemoney included total earnings and earnings per start (USD). International earnings were converted to USD using the exchange rate on the date of racing. Three variables related to calibre of racing performance achieved included best start, weighted Listed and Group race starts and Class Performance Index. Best start was a binary outcome for whether a horse started at least one race at Listed or Group level. Weighted Listed and Group race starts was a sum total of weighted starts, weighting each Listed race (1 point), Group 3 race (2 points), Group 2 race (3 points) and Group 1 race (4 points) respectively. Class Performance Index is a lifetime ratio based on an individual horse's average earnings per start divided by the average earnings per start of all other horses of the same sex and birth year.

#### *4.3.4 Non-starter follow-up information*

Any horse that did not start at least one race was considered a non-starter. Follow-up information for non-starters was sought via email and telephone communication with the connections of each horse, using a flow-chart framework that was designed to obtain increasing levels of detail while minimising bias (Appendix 3).

#### *4.3.5 Data Analysis*

Data analysis was performed via consultation with statisticians using the open-source statistical software package R, version 4.0.2 by RStudio with packages irr<sup>38</sup>.

Total US foal crop for the year and number of yearlings sold at auction were obtained from the Jockey Club Information Systems, Inc. <sup>39</sup>. The percentages of the annual foal crop that were included in the study as yearlings and as two-year-olds were calculated.

Agreement between all pairs of observers classifying sesamoid appearance into the six different categories was assessed using Cohen's unweighted kappa, reported as a group range and mean<sup>40,41</sup>. To differentiate between ordinal grades (number of enlarged vascular channels 0-3, where any with abaxial new bone only or abaxial resorption were classed as 0 for vascular channel appearance) and nominal grades (enlarged vascular channel absence/presence, abaxial new bone only or abaxial resorption only), separate Krippendorff's alpha analyses were run for the ordinal and nominal groupings respectively. This best prepared the observers for consistent grading of images. Interobserver agreement was interpreted in accordance with Landis & Koch (177), whereby a kappa statistic of 0.21-0.40 is considered "fair" agreement, 0.41-0.60 is "moderate", 0.61-0.80 is "substantial" and 0.81-1.00 is considered "almost perfect" agreement.

Radiological findings were summarised using descriptive statistics with frequency distributions at the sesamoid-level by age, by sex, by fore- and hindlimbs and by left and right side. Radiological findings were summarised at the horse-level by age and by fore- and hindlimbs, using maximum grades per horse. Changes between radiological findings in a given sesamoid at one and two years of age were examined using descriptive statistics, including percentages and measures of uncertainty representing 95% confidence intervals. The proportion of horses with sesamoids possessing more than one enlarged vascular channel at yearling age compared to two years of age was analysed using McNemar's Chi-squared test.

The relationships between radiological findings and each of the eight racing performance outcomes described in Section 4.3.3 were analysed using multivariate generalised regression approaches. Binary outcomes, started at least one race and best start were analysed using logistic regression; number of starts was considered with negative binomial regression; and the remaining continuous outcomes were assessed using standard linear regression. Analysis of the outcome "started at least one race" considered all horses, the remaining outcomes considered only horses who started a race. Model diagnostics were performed and

data transformations were used when necessary to satisfy model assumptions, e.g. normally distributed errors. Days to first start was log transformed to resemble an accelerated failure time model. Earnings related measures required log transformation, with all \$0 earnings, and \$0 earnings per start, set to \$1. Class Performance Index required log transformation with all CPI of zero set to 0.05, which is half the minimum performance value of 0.1. Weighted Listed and Group starts required square root transformation.

To test associations between radiological findings and racing performance, horses were assigned a maximum grade for each radiological variable, separately for forelimb and hindlimb sesamoids<sup>13</sup>. Thus eight horse-level radiological findings were examined in each racing performance analysis: 1) maximum forelimb sesamoid vascular channel grade, 2) forelimb sesamoid abaxial new bone (yes/no), 3) forelimb sesamoid abaxial concavity (yes/no), 4) forelimb sesamoid fragment (yes/no), 5) maximum hindlimb sesamoid vascular channel grade, 6) hindlimb sesamoid abaxial new bone, 7) hindlimb sesamoid abaxial concavity and 8) hindlimb sesamoid fragment.

To build the final regression model, each racing performance outcome was first analysed in a multivariable regression against each of the eight horse-level radiological sesamoid variables, always controlling for horse sex as a fixed effect. Each radiological sesamoid variable was then “screened” against that racing performance outcome using a likelihood ratio (logistic or negative binomial regression analyses) or an F-test (linear regression analyses), with inclusion thresholds of  $P < 0.1$  to reduce the rate of false positives across our analysis. No other adjustments were made to p-values to account for a large number of tests. All radiological sesamoid variables that passed screening were simultaneously included in a multivariate regression analysis for that racing performance outcome. Radiological variables were retained by backwards selection for the final model, using a threshold of  $P < 0.05$  for statistical significance.

Coefficient estimates and 95% confidence intervals produced by regression analyses were “back transformed” to provide estimates that pertained to the original scale of the racing performance data, describing the average value of the outcome for each level of radiological sesamoid finding.

## 4.4 Results

### 4.4.1 Study enrolment

Repository radiographs for 2508 yearlings were used (Appendix 4), which represented 10.9% of the annual foal crop and 36.0% of the North American yearlings sold at auction in 2016 (Jockey Club Information Systems Inc). The age of the study yearlings on the first day of the sale ranged from 458 to 614 days old, with a median of 535 days old. Repository radiographs for 436 two-year-olds were used (Appendix 5), representing 19.7% of the two-year-olds sold at auction in North America in 2017.

### 4.4.2 Interobserver agreement

Overall agreement between observers for radiological sesamoid appearance had a mean unweighted Cohen's kappa of  $Ck=0.65$  (group range 0.43-0.89). Krippendorff's alpha for multiple observer agreement in the ordinal categorisation of vascular channel appearance was  $K\alpha=0.79$ . Krippendorff's alpha for multiple observer agreement in categorisation of the nominally grouped data (enlarged vascular channel absence/presence and abaxial contour change) was  $K\alpha=0.74$ .

### 4.4.3 Radiological vascular channel and abaxial contour findings

Table 4.2 presents the prevalence of the various grades of vascular channel appearance and abaxial contour change in individual sesamoids, alongside summary counts of the most severe sesamoid grade per horse. Table 4.3 shows the distribution of findings by sesamoid location, i.e. medial and lateral sesamoids in forelimbs and hindlimbs. The frequency distribution of sesamoid findings was similar in males and females and in left and right-sided limbs (data not shown).

### 4.4.4 Paired yearling and 2-year-old sales radiographs

Radiographs were available at both the yearling sale and a two-year-old sale for 422 horses. Results are shown in Table 4.4. Significantly fewer horses had a Grade 2 or 3 sesamoid at a two-year-old sale

TABLE 4.2. Distribution of sesamoid findings in all sales yearlings and two-year-olds, by individual sesamoid bones and at the horse-level by maximum vascular channel grade per horse, where *n* represents individual proximal sesamoid bones and *N* represents individual horses. Percentages are shown with uncertainty representing 95% confidence intervals.

Sesamoid grade	Yearling sesamoids, n (%)	Two-year-old sesamoids, n (%)	Maximum vascular channel grade per yearling, N (%)	Maximum vascular channel grade per two-year-old, N (%)
Grade 0 vascular channels	15,777 (78.6±0.6%)	2,987 (85.6±1.2%)	948 (37.8±1.9%)	200 (45.9±4.7%)
Grade 1 vascular channels	2,887 (14.4±0.5%)	339 (9.7±1.0%)	1047 (41.7±1.9%)	179 (41.1±4.6%)
Grade 2 vascular channels	678 (3.4±0.3%)	54 (1.5±0.4%)	391 (15.6±1.4%)	45 (10.3±2.9%)
Grade 3 vascular channels	156 (0.8±0.1%)	14 (0.4±0.2%)	122 (4.9±0.8%)	12 (2.8±1.5%)
Abaxial new bone only	314 (1.6±0.2%)	72 (2.1±0.5%)	-	-
Abaxial concavity only	252 (1.3±0.2%)	22 (0.6±0.3%)	-	-
Total	20,064 (100%)	3,488 (100%)	2,508 (100%)	436 (100%)

TABLE 4.3. Distribution of sesamoid findings by medial and lateral location in sales yearling and two-year-old fore- and hindlimbs, where *N* represents individual sesamoids and the percentages shown are the medial vs lateral distribution for each grade, then the proportion of all findings in each grade. Percentages are shown with uncertainty representing 95% confidence intervals.

Sesamoid appearance	Yearling forelimb sesamoids, N (% medial vs lateral)			Two-year-old forelimb sesamoids, N (% medial vs lateral)		
	Medial	Lateral	Total	Medial	Lateral	Total
Grade 0	3630 (46.8±1.1%)	4129 (53.2±1.1%)	7759 (77.3±0.8%)	711 (47.1±2.5%)	798 (52.9±2.5%)	1509 (86.5±1.6%)
Grade 1	824 (57.4±2.6%)	612 (42.6±2.6%)	1436 (14.3±0.7%)	102 (70.3±7.4%)	43 (29.7±7.4%)	145 (8.3±1.3%)
Grade 2	202 (64.1±2.3%)	113 (35.9±2.3%)	315 (3.1±0.3%)	18 (69.2±17.7%)	8 (30.8±17.7%)	26 (1.5±0.6%)
Grade 3	44 (58.7±11.1%)	31 (41.3±11.1%)	75 (0.7±0.2%)	5 (55.6±32.5%)	4 (44.4±32.5%)	9 (0.5±0.3%)
Abaxial new bone only	101 (50.8±6.9%)	98 (49.3±6.9%)	199 (2.0±0.3%)	18 (52.9±16.8%)	16 (47.1±16.8%)	34 (1.9±0.6%)
Abaxial concavity only	215 (86.7±4.2%)	33 (13.3±4.2%)	248 (2.5±0.3%)	18 (85.7±15.0%)	3 (14.3±15.0%)	21 (1.2±0.5%)
Total	5016	5016	10,032	872	872	1744
	Yearling hindlimb sesamoids			Two-year-old hindlimb sesamoids		
	Medial	Lateral	Total	Medial	Lateral	Total
Grade 0	3851 (48.0±1.1%)	4167 (52.0±1.1%)	8018 (79.9±0.8%)	701 (47.4±2.5%)	777 (52.6±2.5%)	1478 (84.7±1.7%)
Grade 1	851 (58.7±2.5%)	600 (41.4±2.5%)	1451 (14.5±0.7%)	120 (61.9±6.8%)	74 (38.1±6.8%)	194 (11.1±1.5%)
Grade 2	208 (57.3±5.1%)	155 (42.7±5.1%)	363 (3.6±0.4%)	20 (71.4±16.7%)	8 (28.6±16.7%)	28 (1.6±0.6%)
Grade 3	48 (59.3±10.7%)	33 (40.7±10.7%)	81 (0.8±0.2%)	4 (80.0% CI 44.9- 100%)	1 (20.0% CI 0- 55.1%)	5 (0.3±0.3%)
Abaxial new bone only	56 (48.7±9.1%)	59 (51.3±9.1%)	115 (1.1±0.2%)	26 (68.4±14.8%)	12 (31.6±14.8%)	38 (2.2±0.7%)
Abaxial concavity only	2 (50.0±49.0%)	2 (50.0±49.0%)	4 (0.0±0.0%)	1 (100±0.0%)	0 (0.0±0.0%)	1 (0.1±0.1%)
Total	5016	5016	10,032	872	872	1744

TABLE 4.4. Distribution of sesamoid grades per individual sesamoid in two-year-old horses (columns), relative to each sesamoid's respective grade at yearling age (rows), for 422 horses with paired yearling and two-year-old sales radiographs. Shaded values represent grades that did not change between sales. †Enlarged vascular channel(s) have resolved in order for abaxial contour change to be the recorded finding.

Sesamoid grade at yearling sale	Sesamoid grade at two-year-old sale, N (%)						
	Grade 0	Grade 1	Grade 2	Grade 3	Abaxial new bone only	Abaxial concavity only	Total sesamoids n=3376
Grade 0	2398 (91.3±1.1%)	164 (6.2±0.9%)	15 (0.6±0.3%)	1 (0.0% CI 0.0-0.1%)	42 (1.6±0.5%)	7 (0.3±0.2%)	2627 (100%)
Grade 1	376 (72.3±3.8%)	113 (21.7±3.5%)	14 (2.7±1.4%)	3 (0.6% CI 0.0-1.2%)	12 † (2.3±1.3%)	2 † (0.4% CI 0.0-0.9%)	520 (100%)
Grade 2	51 (48.6±9.6%)	37 (35.2±9.1%)	11 (10.5±5.8%)	4 (3.8±3.7%)	2 † (1.9% CI 0.0-4.5%)	0 † (0.0±0.0%)	105 (100%)
Grade 3	6 (40.0±24.8%)	1 (6.7% CI 0.0-19.3%)	6 (40.0±24.8%)	2 (13.3% CI 0.0-30.5%)	0 (0.0±0.0%)	0 (0.0±0.0%)	15 (100%)
Abaxial new bone only	34 (58.6±12.7%)	5 (8.6±7.2%)	3 (5.2% CI 0.0-10.9%)	0 (0.0±0.0%)	15 (25.9±11.3%)	1 (1.7% CI 0.0-5.1%)	58 (100%)
Abaxial concavity only	33 (64.7±13.1%)	4 (7.8±7.3%)	1 (2.0% CI 0.0-5.8%)	1 (2.0% CI 0.0-5.8%)	0 (0.0±0.0%)	12 (23.5±11.6%)	51 (100%)
Total sesamoids	2898 (85.8±1.2%)	324 (9.6±1.0%)	50 (1.5±0.4%)	11 (0.3±0.2%)	71 (2.1±0.5%)	22 (0.7±0.3%)	3376 (100%)

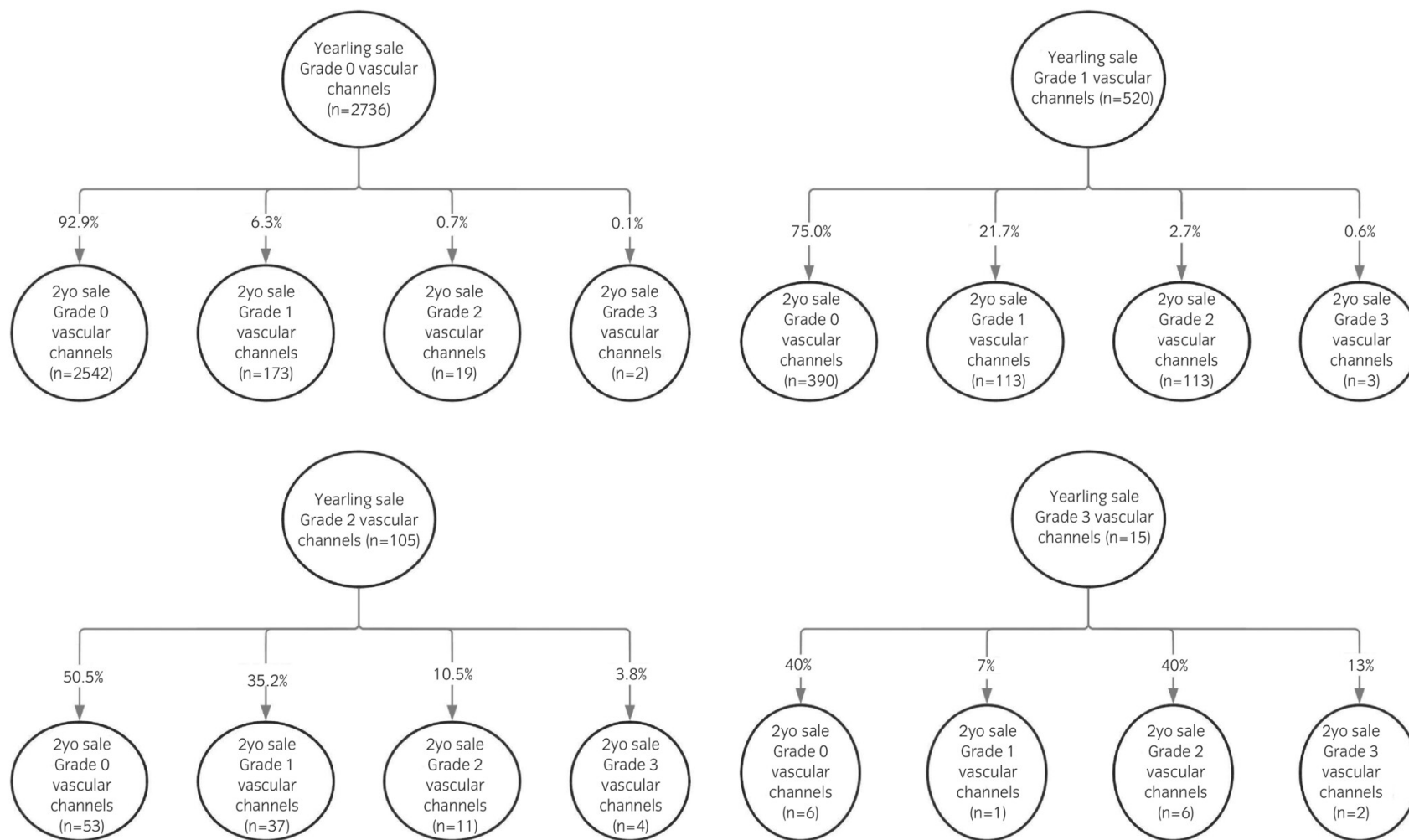


FIGURE 4.3. Evolution of the vascular channel appearance in individual sesamoids, beginning with the grade present at yearling sales. Grade 0 includes sesamoids with no enlarged vascular channels that had abaxial contour change. N refers to individual proximal sesamoid bones from 422 horses with both yearling and 2-year-old sales radiographs.

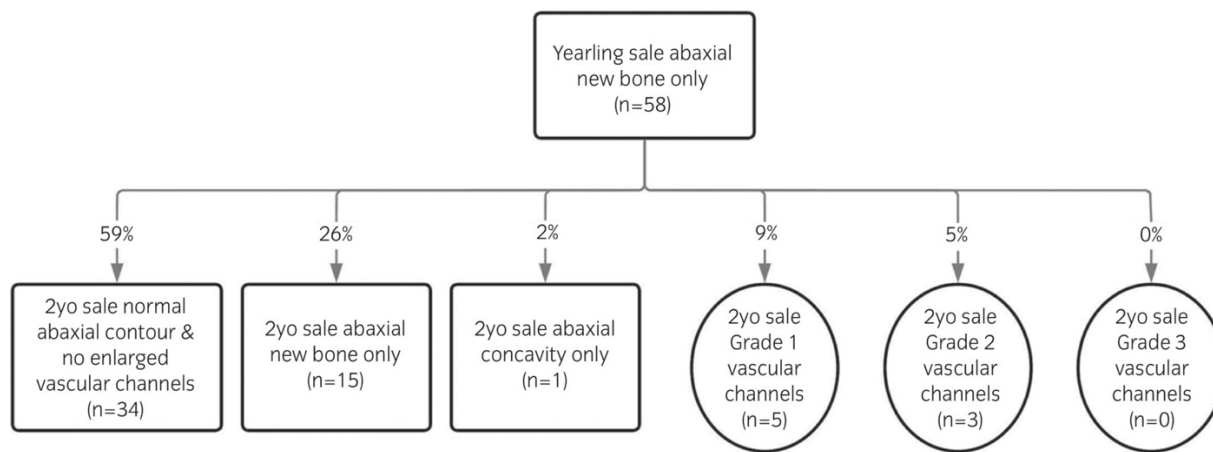


FIGURE 4.4. Evolution of the appearance of sesamoids that had new bone on the abaxial margin at yearling sales, in the absence of any enlarged vascular channels. N refers to individual proximal sesamoid bones from 422 horses with both yearling and 2-year-old sales radiographs, of which there were 58 yearling sesamoids with abaxial margin new bone as the only finding.

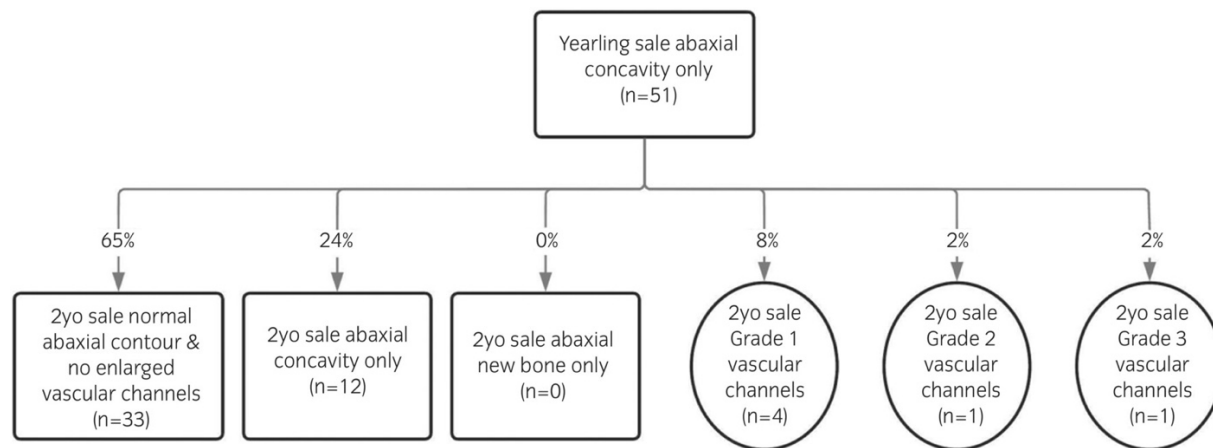


FIGURE 4.5. Evolution of the appearance of sesamoids that had a distinct concavity in the abaxial margin at yearling sales, in the absence of any enlarged vascular channels. N refers to individual proximal sesamoid bones from 422 horses with both yearling and 2-year-old sales radiographs, of which there were 51 yearling sesamoids with abaxial margin concavity as the only finding.

(12.3%) compared to the proportion from the same group of horses that had a Grade 2 or 3 sesamoid at the yearling sale (18.0%),  $P=0.008$ . Of the horses that had a Grade 2 or 3 sesamoid on yearling sale radiographs that presented for sale the following year, 69.7% had no sesamoids greater than Grade 1 on two-year-old sale radiographs. 55.8% of the horses that had a Grade 2 or 3 sesamoid as two-year-olds had no sesamoids greater than grade 1 as yearlings (Figures 4.3-4.5).

#### *4.4.5 Apical and abaxial sesamoid fragments*

Fragmentation of the apical and abaxial aspects of the sesamoids occurred in 80 of 2508 yearlings with a total of 97 fragments, affecting 0.5% of all yearling sesamoids and 3.2% of yearlings. 81.3% of affected yearlings had one sesamoid with a fragment (65/80), 17.5% had two sesamoids with fragments (14/80) and one horse had fragmentation of four sesamoids. At the two-year-old sales, 23 fragments were seen in 22 horses, involving 0.7% of two-year-old sesamoids and 5.0% of two-year-old horses.

Of the 97 yearling sesamoid fragments, 40.2% were in a forelimb and 59.8% were in a hindlimb (39/97 and 58/97, respectively). By specific location, 27.8% were medial forelimb sesamoid fragments (27/97), 12.4% were lateral forelimb (12/97), 29.9% were medial hindlimb (29/97) and 29.9% were lateral hindlimb sesamoid fragments (29/97). Of the 23 two-year-old fragments, 30.4% were in a forelimb and 69.6% were in a hindlimb (7/23 and 16/23, respectively); 26.1% were medial forelimb (6/23), 4.3% were lateral forelimb (1/23), 43.5% were medial hindlimb (10/23) and 26.1% were lateral hindlimb sesamoid fragments (6/23).

The vascular channel appearance and other changes in abaxial contour for yearling sesamoids with apical and abaxial fragments are shown in Table 4.5. Of the 80 yearlings with fragments, 40.0% had apical fragments only and 55.0% had abaxial fragments only ( $n=32$  and  $n=44$ , respectively), 2.5% had both apical and abaxial fragments ( $n=2$ ) and 2.5% had a record of arthroscopic sesamoid fragment removal from an unspecified location ( $n=2$ ). In the 25 yearlings with forelimb abaxial fragments, there were 31 individual abaxial fragments; 77.4% medial and 22.6% lateral ( $n=24$  and  $n=7$ , respectively) (Table 4.5).

Presale arthroscopic fragment removal was performed on 70.6% of yearlings with apical fragments (24/34) and 8.7% of yearlings with abaxial fragments (4/46). Apical fragments were removed in their

entirety in 83.3% of presale surgery cases (20/24). In 16.7% of affected yearlings a partial apical fragment or mineralisation remained visible on yearling sales radiographs (4/24). No abaxial fragments were removed in their entirety, with partial fragmentation or mineralisation visible radiographically in all 4 affected horses that had presale surgery.

TABLE 4.5. The radiological appearance of yearling sesamoids with fragments, in terms of the concomitant presence or absence of enlarged vascular channels or abaxial contour change in addition to the fragment finding. Numbers in the table refer to individual sesamoid bones and includes sesamoids for which the fragment was surgically removed prior to sale.

Radiological appearance of sesamoids with fragments	Sesamoids with an apical fragment	Sesamoids with an abaxial fragment
Grade 0 vascular channels	28 (73.7±14.0%)	26 (46.4±13.1%)
Grade 1 vascular channels	3 (7.9% CI 0.0-16.4%)	8 (14.3±9.2%)
Grade 2 vascular channels	2 (5.3% CI 0.0-12.4%)	9 (16.1±9.6%)
Grade 3 vascular channels	2 (5.3% CI 0.0-12.4%)	10 (17.9±10.0%)
Abaxial new bone only	3 (7.9% CI 0.0-16.4%)	2 (3.6% CI 0.0-8.4%)
Abaxial concavity only	0 (0.0±0.0%)	1 (1.8% CI 0.0-5.3%)
Total	38 (100%)	56 (100%)

#### 4.4.6 Associations with racing performance

##### *i. Started at least one race*

Of all sales yearlings studied, 2119 (84.5%, 95% CI 83.0-86.0%) started at least one race by the end of their four-year-old year. Three variables relating to sesamoid appearance were retained in the multivariate logistic model for starting at least one race. Likelihood ratio tests indicated that the grade of vascular channel appearance in forelimb sesamoids ( $P=0.005$ ), abaxial new bone in forelimb sesamoids ( $P=0.015$ ) and apical or abaxial fragments in forelimb sesamoids ( $P=0.002$ ) were each related to the odds of starting at least one race.

Table 4.6 describes the probability of racing based on levels of the radiological variables examined in the multivariate analysis. The multivariate logistic model that accounted for radiological characteristics

and horse sex estimated that horses with a Grade 3 forelimb sesamoid as a yearling had a 0.52 (95% CI 0.37-0.67) probability of starting a race (43/64 raced), which was significantly different compared to a 0.73 (95% CI 0.64-0.81) probability for horses with only Grade 0 forelimb sesamoid vascular channels (1909/2266 raced) ( $P<0.001$ ). Grade 1 ( $P=0.8$ ) or Grade 2 ( $P=0.3$ ) vascular channels in forelimb sesamoids

TABLE 4.6. The proportion of sales yearlings that raced for each level of radiological variable reported in the multivariate logistic regression model. Displayed are the overall number of yearlings with a given radiological finding, the percentage of those yearlings that raced, the back-transformed probability of racing based on a logistic model controlling only for horse sex, and the multivariate regression model controlling for the other selected radiological findings.

	Total yearlings	N (%) of yearlings that raced	Probability of racing (95% CI), from multivariate model	P-value
<i>Forelimb sesamoid maximum vascular channel grade</i>				
0	1417	1204 (85.0%)	0.73 (0.64-0.81)	Reference level
1	803	687 (85.6%)	0.74 (0.65-0.81)	0.8
2	224	185 (82.6%)	0.69 (0.57-0.79)	0.3
3	64	43 (67.2%)	0.52 (0.37-0.67)	<0.001
<i>Forelimb sesamoid abaxial new bone</i>				
None	2351	1997 (84.9%)	0.73 (0.65-0.80)	Reference level
Abaxial new bone only	157	122 (77.7%)	0.62 (0.49-0.73)	0.01
<i>Forelimb sesamoid fragments</i>				
None	2477	2100 (84.8%)	0.78 (0.74-0.82)	Reference level
Apical or abaxial fragment	31	19 (61.3%)	0.55 (0.37-0.72)	0.005

were not significantly associated with a different probability of racing compared to having only Grade 0 vascular channels. For abaxial new bone in the absence of enlarged vascular channels, yearlings with affected forelimb sesamoids had on average a 0.62 (95% CI 0.49-0.73) probability of racing (122/157 raced) compared to a 0.73 (95% CI 0.65-0.80) probability for yearlings without abaxial new bone alone on

forelimb sesamoids (1997/2351 raced) ( $P=0.01$ ). For apical or abaxial fragments in forelimb sesamoids, including fragments that were surgically removed prior to sale, affected yearlings had on average a 0.55 (95% CI 0.37-0.72) probability of racing (19/31 raced), compared to a 0.78 (95% CI 0.74-0.82) racing probability for yearlings without a forelimb sesamoid fragment (2100/2477 raced) ( $P=0.005$ ). Table 4.7 provides the breakdown of racing starters for apical and abaxial fragment locations, by forelimb and hindlimb. The table omits two horses with both apical and abaxial fragments and two horses with a surgical record of fragment removal where the fragment location was unspecified and there was no radiological indication of the fragment site.

TABLE 4.7. The proportion of sales yearlings with sesamoid fragments that started at least one race, by fragment location within the sesamoid and by forelimb and hindlimb location. Counts represent individual horses and include those in which fragments underwent arthroscopic removal prior to or after the yearling sale.

Proportion of sales yearlings with sesamoid fragments that raced, by fragment location

	Apical fragment	Abaxial fragment
Forelimb sesamoid fragmentation	75.0% (3/4)	60.0% (15/25)
Hindlimb sesamoid fragmentation	82.1% (23/28)	68.4% (13/19)
Combined total for forelimb and hindlimb sesamoids	81.3% (26/32)	63.6% (28/44)

*ii. Follow-up of non-starters*

Follow-up information was obtained for 42.7% of non-starters (166/389). For 50.6% of non-starters followed (84/166), confirmation was given that the horse never raced but the reason for this was unknown, or the horse was exported from the United States and the racing history was inaccessible. Follow-up information was complete for 49.4% of non-starters followed (82/166). Of these, 80.5% failed to race for reasons related to performance (66/82), 12.2% did not race due to fatal or near fatal accidents unrelated to performance (10/82), 6.1% did not race due to medical events requiring euthanasia (5/82), and 1.2% were purchased from the yearling sale for a non-racing discipline (1/82). Of the non-starters with a performance-related reason for not racing: 68.2% did not race because of lameness (45/66), 21.2% were deemed to have insufficient athletic talent (14/66), 4.5% had upper respiratory tract dysfunction (3/66), 1.5% did not start

for behavioural reasons, 1.5% had severe exertional rhabdomyolysis, 1.5% had cervical vertebral malformation and 1.5% had recurrent exercise-induced pulmonary haemorrhage (1/66 each).

In the horses where lameness was the reason for not racing, 17.8% had a clinical diagnosis involving a sesamoid or suspensory ligament branch (8/45). Two of these horses sustained sesamoid fractures in training; 1 fractured a forelimb sesamoid that had Grade 3 vascular channels as a yearling and the other fractured a hind sesamoid that had a Grade 0 appearance as a yearling and two-year-old. The yearling vascular channel appearance in the other 6 horses with lameness attributed to the sesamoid-suspensory branch unit was Grade 0 in 2 horses, Grade 1 in 2 horses, Grade 2 in 1 horse and Grade 3 in 1 horse. In the non-starters that did not race because they were deemed too slow, the majority had only Grade 0 sesamoids (57.1%, 8/14), 21.4% had no worse than a Grade 1 sesamoid (3/14), 14.3% had Grade 2 sesamoids (2/14) and 7.1% had a Grade 3 sesamoid (1/14).

*iii. Age at first race start*

For horses that raced, the median age at first race start was 969 days old (interquartile range (IQR) 882-1102). Only hindlimb vascular channel grade met selection for multivariate regression analysis. The presence of Grade 3 vascular channels in a hindlimb sesamoid was significantly associated with an older age at first race start, 1047 days (95% CI 1004-1091) compared to 993 days (95% CI 984-1002) for Grade 0 ( $P=0.01$ ). Thus, on average, yearlings with Grade 3 vascular channels in a hindlimb sesamoid raced 54 days later than unaffected horses (95% CI 20-89) (Table 4.8).

TABLE 4.8. Results from a multivariate log-linear regression model for age at first race start in days old by maximum hindlimb vascular channel grade as yearlings, for 2119 sales Thoroughbreds that started at least one race.

Hindlimb sesamoid maximum vascular channel grade	Number of yearlings, N	Age at first start in days old (95% CI), from multivariate model	P-value
0	1202	993 (984-1002)	Reference level
1	670	994 (982-1006)	0.9
2	192	975 (953-997)	0.1
3	55	1047 (1004-1091)	0.01

*iv. Number of race starts*

Horses that raced had a median of 11 starts by the end of the study period (IQR 6-17). Forelimb vascular channel grade met selection for multivariate regression analysis of horses' total number of race starts. Compared to horses with Grade 0 forelimb sesamoids (12.7 starts, 95% CI 12.2-13.2), horses with Grade 3 vascular channels in a forelimb sesamoid as yearlings started significantly fewer races in their career to age five (9.9 starts, 95% CI 8.0-12.2,  $P=0.03$ ) (Table 4.9).

TABLE 4.9. Results from a multivariate negative binomial regression model for mean number of race starts from 2-4 years of age by maximum forelimb sesamoid vascular channel grade as yearlings, for 2119 sales Thoroughbreds that started at least one race.

Forelimb sesamoid maximum vascular channel grade	Number of yearlings, N	Mean number of race starts (95% CI), from multivariate model	P-value
0	1204	12.7 (12.2-13.2)	Reference level
1	687	11.9 (11.3-12.5)	0.05
2	185	11.6 (10.5-12.8)	0.1
3	43	9.9 (8.0-12.2)	0.03

*v. Earnings*

Median total individual earnings for horses that raced were \$32,253 (IQR \$9,166-\$73,535). Median earnings per start for the same horses were \$2,674 (IQR \$1,112-\$5,901). The presence of abaxial new bone on forelimb sesamoids was significantly associated with reduced total earnings and reduced earnings per start. On average, total earnings for horses with forelimb abaxial new bone were \$7,236 (95% CI \$4,396-11,910) compared to \$15,734 (95% CI \$13,914-17,793) for horses without ( $P=0.003$ ). Earnings per start were \$992 (95% CI \$673-1,462) for horses with abaxial new bone on forelimb sesamoids compared to \$1,845 (95% CI \$1,677-2,031) for horses without ( $P=0.002$ ). Therefore, the presence of forelimb sesamoid abaxial new bone was associated with a 54% reduction in total earnings (95% CI 24-72) and a 46% reduction in earnings per start (95% CI 21-64), compared to the average earnings of horses without this finding.

#### *vi. Race caliber*

None of the eight radiological variables were significantly associated with any measures used to assess performance calibre. The average Class Performance Index for all horses that raced was 1.47 (median 0.70, IQR 0.33-1.47). Career Best Starts for all 2119 horses that raced included 1772 horses that raced but did not reach Listed or Group level (83.6%), 101 horses that achieved a Listed start as their highest class of race (4.8%), 89 horses that achieved a Group 3 start as their highest class of race (4.2%), 65 horses that achieved a Group 2 start as their highest class of race (3.1%) and 92 horses that reached a Group 1 race (4.3%). Weighted Listed and Group Starts data remained highly left skewed despite square root transformation.

### **4.5 Discussion**

This is the first time that the radiological appearance of sesamoids has been prospectively investigated in a large number of sales yearlings and followed over the period during which sales horses enter athletic training. The data has produced previously unavailable information on changes to radiological findings from yearling to two-year-old sales, as well as associations with subsequent racing performance. In addition, a refined sesamoid grading system has been established for use in sales horses. The results enable some findings to be regarded as an acceptable appearance in sales repositories, particularly grade 1 vascular channels and abaxial sesamoid concavity. The results have also identified the findings that are associated with an evidence-based risk of reduced performance, namely grade 3 vascular channels, abaxial new bone and fragments in forelimb sesamoids.

Central to this study was the development of a robust grading scale for the radiological evaluation of sesamoids. Interobserver agreement in the application of the new grading scale was substantial <sup>42</sup>, representing a considerable improvement on previous sesamoid grading agreement <sup>41</sup>. The grading system used in the current study was designed to simplify categorisation to enable uniform grading by veterinarians; to provide objectivity as to what constitutes an acceptably sized abaxial channel opening and to accommodate extremely enlarged vascular channels in grading by total width. Previous work has evolved

from simply counting the number of radiologically visible channels <sup>19</sup>, to measuring channel width <sup>43</sup>, considering non-parallel sides <sup>13</sup>, then allowing a conical-shaped opening at the abaxial margin of the channel <sup>22</sup>. Visible sesamoid vascular channels occur in 93-98% of the yearling sales population <sup>13,23</sup>. Therefore, categorising sesamoid appearance by the number of enlarged channels, rather than counting all visible channels, improves the utility of a grading system for determining pathological significance because it eliminates the inclusion of many normal sesamoids and refines the threshold for what is considered abnormal. For simplicity, the new grades were numbered to correspond with the number of enlarged channels. Importantly, sesamoids with only one enlarged channel (grade 1) were analysed separately from sesamoids with multiple enlarged channels for the first time.

Nearly 20% of yearling sesamoids in the current study were found to have one or more enlarged vascular channels (grades 1-3). This is similar to the prevalence found in a sample of 291 sales yearlings that subsequently entered a single Florida training centre, where 18% of sesamoids had one or more enlarged vascular channels using a modified Spike-Pierce (2003) grading scale <sup>23</sup>. Over 95% of yearlings with a maximum of grade 0 and grade 1 vascular channels, that subsequently presented at a two-year-old sale, either remained unchanged or improved in vascular channel appearance. This included resolution of 75% of yearling grade 1 vascular channels. The withdrawal of two-year-olds intended for resale if they developed worsening sesamoid change could not be accounted for. However, whilst this potential bias exists, the results still present evidence that vascular channel grade improvement is possible, even when faced with the onset of athletic training between yearling and two-year-old sales. Furthermore in the current study, yearlings with a maximum of one enlarged vascular channel (grade 1) on their worst sesamoid had no evidence of reduced racing performance by any outcome measure, nor did yearlings with grade 2 vascular channels. Only grade 3, the most severe grade of vascular channel appearance, was negatively associated with racing performance. Grade 3 vascular channels were present in 5% of yearlings and when this occurred in a forelimb sesamoid horses were significantly less likely to race. Affected horses that did race did so fewer times than their unaffected peers. When the grade 3 sesamoid was in a hindlimb, horses were no less likely to race but had, on average, a 54-day delayed start to their racing careers. In contrast to

previous research, in which 10 sales yearlings had a grade 3 equivalent sesamoid and were shown to start fewer races and earn less prizemoney, no significant evidence of reduced earnings was found in the current study for affected horses that did race<sup>22</sup>. An 11-fold increase in the number of affected horses studied this time may explain this difference.

Forelimb sesamoids without any enlarged vascular channels but with abaxial new bone production were associated with significantly reduced total earnings and earnings per start in this study. Productive bony changes on the abaxial surface of the sesamoid may represent enthesiophyte formation of importance to the suspensory ligament branch<sup>44</sup>. Most commonly in this study, abaxial new bone was either present on its own or concurrently with grade 3 vascular channels. The grading system was intentionally designed to address the question of whether abaxial new bone is significant when it occurs in the absence of enlarged vascular channels. Previously, Kane *et al* (2003) found reduced odds of racing for 14 sales yearlings with enthesiophyte formation on forelimb sesamoids and fewer placings and lower earnings for those with enthesiophyte formation on hindlimb sesamoids. However, this included both abaxial and basilar new bone, the latter associated with distal sesamoidean ligament attachments, and appears to have included healed fragments. In contrast, Spike-Pierce *et al* (2003) found that abaxial border bone proliferation, in the absence of vascular channel changes, did not affect number of race starts or prizemoney earned, thereby concluding that an insult causing productive bony change, but of a magnitude insufficient to create changes in the sesamoid vasculature, was not detrimental to future performance. The results of the present study refute this and suggest that abaxial new bone in isolation should not be dismissed when it is seen on forelimb sesamoids.

Altering the contour of the sesamoid border in an opposite manner, abaxial concavity was almost exclusively a forelimb sesamoid finding in this study. It was predominantly seen in yearlings and the vast majority of affected sesamoids were medial. By two years of age, the prevalence of this finding had greatly reduced and less than 1% of two-year-old sesamoids were affected. Paired sales radiographs demonstrated that over 65% of abaxial concavities resolved completely by two-year-old sales. When abaxial concavity remained visible in two-year-olds, invariably it was subjectively shallower. No abaxial concavity developed

in two-year-olds that did not have the finding as a yearling. There was no evidence of a significant difference in racing performance for yearlings that had sesamoid abaxial concavity, by any outcome measure, compared to unaffected horses. This corroborates the opinion of Spike-Pierce and Bramlage that abaxial concavity likely resolves and is innocuous<sup>22</sup>. Although at times dramatic in appearance, this finding usually improves radiologically with age, is not detrimental to performance and should not be considered “sesamoiditis”.

Results of this study show that grade 3 vascular channels, abaxial new bone, and apical or abaxial fragments reduced the likelihood of racing when present in forelimb sesamoids, whereas grade 3 vascular channels only delayed the start of racing when present in hindlimb sesamoids. Originally, O’Brien *et al* (1971) speculated that the increased musculoskeletal load on forelimbs may mean that forelimb sesamoid changes have a greater effect on performance than hindlimb changes. This new evidence supports that assumption. Until now, the importance of forelimb versus hindlimb location with respect to some sesamoid findings could not be determined or was not supported, in part due to sample size<sup>12,22</sup>. The importance of sesamoid location has only previously been established for fragment prognosis<sup>45-47</sup>. Forelimb sesamoid fragments were predominantly abaxial in this study, whereas the majority of hind sesamoid fragments were apical. The association of forelimb fragments with a reduced likelihood of racing was irrespective of surgical fragment removal pre- or post-sale, though it is worth noting that most of the removed fragments were apical and therefore were from hindlimb sesamoids. Enlarged vascular channels occurred alongside abaxial fragmentation more commonly than alongside apical fragmentation and, although the analysis did not test abaxial and apical fragments separately, forelimb fragments significantly reduced the likelihood of racing even when accounting for the increased presence of grade 3 vascular channels alongside abaxial fragments, as both were retained in the model. In the work of Kane *et al* (2003), a low percentage of yearlings with apical or abaxial forelimb fragments raced but, with only 5 affected horses, statistical significance was not obtained. This warranted further investigation if a larger number of horses could be studied<sup>12</sup>, which has now occurred. Previous studies involving sesamoids consisted of: 1127 yearlings in a 7% sample across four years of sales<sup>12,13</sup>, 487 yearlings from four years of sales<sup>22</sup> and a select cohort of

291 sales yearlings<sup>23</sup>. The inclusion of 2508 yearlings in the present study represented a 66% sample of all yearlings at one sale that had radiographs.

The population studied was specific to Thoroughbred yearlings and two-year-olds with radiographs submitted for sale at public auction and were horses deemed clinically fit for presentation at sales. Had the study design been able to include horses that were excluded prior to sale because of adverse radiological findings or clinical lesions, a greater severity of pathologic change would likely have been seen and perhaps higher numbers of grade 3 sesamoids included. Consequently, more horses may have failed to race or perform well, thus associations with some performance outcomes may have been stronger. For the minor radiological findings that do not influence whether or not a horse is entered for sale, the strength of the performance associations should approximate those of the non-sale population. Care should be taken when interpreting the evidence linked to paired yearling and two-year-old sale radiographs, as this process will underestimate the proportion of findings in the whole population that worsen enough to be excluded from resale. Due to the specific population studied, application of the results is most appropriate in the Thoroughbred sales repository environment. However, the grading scale was developed for universal use in the evaluation of equine proximal sesamoid bones. Radiological evaluation was limited to the standard projections stipulated in sales repository protocol and these views identify the most common findings in young Thoroughbreds but occasional lesions require non-standard views for clarification. The period of observer training proved valuable and would be a useful training tool in practice to improve consistency between veterinarians involved in sales radiology.

#### **4.6 Conclusions**

Thoroughbred sales yearlings were significantly less likely to race if they had grade 3 vascular channels in forelimb sesamoids, abaxial new bone on forelimb sesamoids, or apical or abaxial fragments off forelimb sesamoids. Among horses that did race, those with forelimb grade 3 vascular channels had fewer starts, those with hindlimb grade 3 vascular channels had a delayed start to their racing careers, and those with abaxial new bone on forelimb sesamoids had lower total earnings and earnings per start. There

was no evidence of reduced performance associated with grade 1 or grade 2 vascular channels, or abaxial sesamoid concavity. Given the ambiguity surrounding the term “sesamoiditis”, adopting the use of a vascular channel grade and an abaxial contour descriptor instead will provide a means of reducing the controversy that affects the interpretation of sesamoid findings in young Thoroughbreds.

CHAPTER 5 – ULTRASONOGRAPHY OF THE SUSPENSORY LIGAMENT BRANCHES IN  
YEARLING AND 2-YEAR-OLD THOROUGHBRED SALES HORSES: PREVALENCE,  
PROGRESSION OF FINDINGS AND ASSOCIATIONS WITH RACING PERFORMANCE<sup>d</sup>

### 5.1 Summary

Ultrasonography of the equine suspensory ligament branches is becoming increasingly commonplace presale, as consignors and prospective purchasers seek to gather more information on which to base their sale and management decisions. However, the ultrasonographic appearance of the suspensory ligament branches has never been studied at Thoroughbred sales. There is also no published reference data for suspensory branch size in young Thoroughbreds.

This was a prospective cohort study using an enrolled sample. The objectives were to 1) define the prevalence of ultrasonographical findings in the forelimb suspensory ligament branches of yearling and 2-year-old sales Thoroughbreds, 2) identify any associations between suspensory branch findings and future racing performance and 3) monitor changes in suspensory branch findings between yearling and 2-year-old sales in horses that present for sale at both ages.

Horses were enrolled from a 2016 yearling sale and five 2017 2-year-old sales with consignor permission. Ultrasonography was performed immediately prior to the sales. Ultrasonographic findings relating to suspensory ligament branch (branch) size, fibrillar pattern, the presence of hyperechoic foci, periligamentar tissue thickness and the adjacent proximal sesamoid bone surface were examined. Associations with racing performance from 2 to 4 years of age were investigated using multivariate regression analyses. Clinical follow-up was sought to ascertain why horses that did not race never started.

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<sup>d</sup> Peat, F. J., Kawcak, C. E., McIlwraith, C. W., Berk, J. T., Keenan, D. P., Selberg, K. T. & Ojeda, A. (2024). Ultrasonography of the suspensory ligament branches in yearling and 2-year-old Thoroughbred sales horses: Prevalence, progression of findings and associations with racing performance. *Equine Veterinary Journal*, (In Press). *Included with permission of journal Editor-In-Chief.*

A total of 593 sales yearlings and 367 2-year-olds had forelimb branch ultrasonography performed. Grade  $\geq 2$  fibrillar branch change was present in 8.9% of yearlings and 14.4% of 2-year-olds. A 0.25cm increase in branch width was associated with a 49-day delayed start to racing careers ( $P < 0.001$ , 95% Confidence Interval (CI): 21 – 77 days). The presence of Grade 2 hyperechoic foci was associated with significantly lower total earnings ( $P = 0.01$ , 95% CI: \$2,000-\$16,022) and lower earnings per start ( $P = 0.003$ , 95% CI: \$349-\$1,718) in United States Dollars. Grade 3 fibrillar branch change had clinically important reductions in the probability of racing, calibre of racing performance and earnings. Grade 1 fibrillar pattern was associated with significantly higher earnings per start ( $P = 0.004$ , 95% CI: \$2,641-\$5,759).

The findings are applicable to horses prepared for public auction and deemed fit to be entered for sale. The results may underestimate the proportion of severe lesions in horses not entered for sale.

Reference values specific to young Thoroughbreds have been established. Grade 1 fibrillar suspensory branch change should be regarded as an acceptable appearance in sales yearlings and 2-year-olds. Approximately one third of Grade 2 yearling branches progressed to a Grade 3 lesion. Evidence of enlarged branch width and Grade 2 hyperechoic foci at 2-year-old sales constitute a risk to racing performance.

## **5.2 Introduction**

Ultrasonographic changes in the branches of the equine suspensory ligament are a potential concern in young Thoroughbred horses sold at auction. The suspensory ligament bifurcates in the distal metacarpal and metatarsal regions, giving rise to medial and lateral branches that insert onto the abaxial aspect of the proximal sesamoid bones (sesamoids)<sup>16</sup>. Because of the anatomical relationship between the two structures, insertional suspensory branch desmopathy has been implicated in horses with radiological sesamoid changes<sup>48</sup>. Ultrasonography of the suspensory branches is becoming increasingly commonplace at sales, as consignors and prospective purchasers sensitised to the issue of “sesamoiditis” seek to gather more information on which to base their sale decisions.

To date, the ultrasonographic appearance of the suspensory ligament branches at yearling and 2-year-old sales has not been studied and the future impact of subclinical branch changes in sales Thoroughbreds is unknown. Clinical suspensory branch injury occurring prior to the start of the 2-year-old racing season and the effect it has on future performance has been studied<sup>33,34</sup>. However, low numbers of severe branch lesions precluded further analysis of this group and larger studies were called for. Mild or low-grade ultrasonographic branch findings do not necessarily correspond to clinical injury and have been shown to exist in a proportion of clinically normal racehorses, thus it has been cautioned that mild changes should not be overinterpreted until studied further<sup>28,29</sup>. Additionally, the absence of published reference data for suspensory branch size in young Thoroughbreds has been noted<sup>28</sup>.

The aim of this study was to objectively evaluate the ultrasonographic appearance of the suspensory branches in a large number of sales Thoroughbreds; to define the range and prevalence of suspensory branch findings at sales and to analyse associations with racing performance. In horses presented for sale at both yearling and 2-year-old age, the study aimed to follow ultrasonographic findings between sales to observe the frequency of improvement or worsening of findings. Lastly, the intention was to provide an evidence-based determination of which ultrasonographic findings in sales horses should be regarded as acceptable appearance and which findings constitute a risk to future performance.

## **5.3 Methods**

### *5.3.1 Study design*

A prospective cohort study commenced at the 2016 Keeneland September Thoroughbred Yearling Sale in Kentucky, USA. Permission to perform ultrasonography was requested in writing from all consignors presenting yearlings at the sale; accompanied by a letter providing information about the purpose of the radiology and ultrasonography studies being undertaken by a research group, with permission from the sales company<sup>48,49</sup>. The consent form gave consignors the option to allow ultrasonography of horses in their sales draft and a separate option to allow access to repository radiographs; consignors could choose one or both options and were under no obligation to choose either. The same was

asked of subsequent consignors of study horses that were catalogued for sale again at the five major North American 2017 2-year-old sales: Fasig-Tipton Gulfstream, Ocala Breeders' Sales (OBS) March, OBS Spring, Fasig-Tipton Maryland and OBS June. Research ultrasonography was performed by a group of equine veterinarians experienced in ultrasonography including the first author. Images were acquired within a 14-day window prior to the first day of sale. For yearlings, this took place on-farm prior to arrival at the sales grounds. For 2-year-olds, this occurred prior to the timed workout (breeze); either at the sales grounds for out-of-state horses, or at Florida training stables for local consignors. Images were stored for confidential post-sale evaluation and no findings were discussed during image acquisition.

### *5.3.2 Ultrasonographic technique*

A standardised protocol was followed to obtain images of the medial and lateral suspensory branches of the left and right forelimbs on each horse. Limb preparation involved cleaning with isopropyl alcohol and the application of coupling gel. Hair was not clipped and an acoustic standoff pad was not used. Linear transducer frequency, depth, grey map and focal zone settings were optimised to achieve the highest quality diagnostic image for each ultrasound machine. Horses were positioned with body weight evenly distributed in a square stance. Intravenous sedation was only used when necessary, with consignor permission. The entirety of each suspensory branch was scanned from proximal to distal and dorsal to palmar. Images saved for each branch included: a minimum three-second cine loop of the distal third of each branch in transverse orientation, side-by-side transverse and longitudinal still images of the insertion of each branch onto the proximal sesamoid bone, clearly showing the bone contour, with the ultrasound beam perpendicular to the sagittal plane of the limb, a second set of side-by-side transverse and longitudinal images were obtained for the palmar aspect of the branch insertion, with the transducer positioned on the palmarolateral or palmaromedial aspect of the limb and directed dorsad at an approximately forty-five degree angle to the sagittal plane. In addition, if a suspected abnormality or artefact was detected during scanning, a dynamic cine loop of the region of fibres in question and representative still images were saved. In accordance with previous work, only insertional images were graded<sup>34</sup>, as it has been shown that the

majority of abnormalities occur in the distal third of the branch and insertional desmopathy was the focus of this study<sup>28,32</sup>.

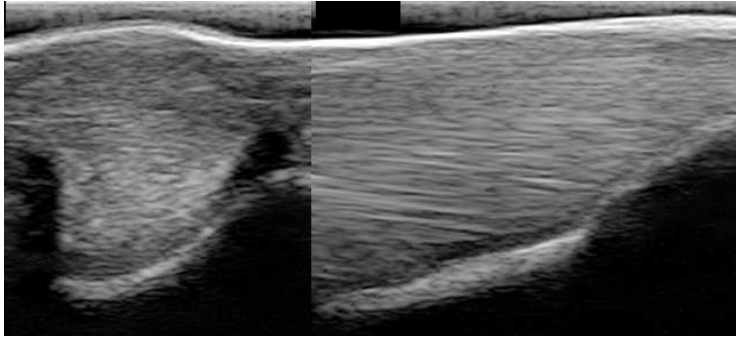
### 5.3.3 Image evaluation

All ultrasound images were evaluated by one observer with ten years' experience in sales radiology and board-certified in equine veterinary sports medicine (FP), after consensus grading of the first 100 branches alongside a board-certified veterinary radiologist (KS)<sup>31</sup>. Previously reported measures of interobserver agreement using similar grading systems were moderate to substantial<sup>28,34</sup>. Multiple operators were necessary to perform ultrasonography within the short presale window and the subsequent use of a single observer to grade and measure images was intentional, as recommended for serial ultrasound investigations<sup>50</sup>. The overall ultrasonographic appearance of each branch was evaluated to assess fibrillar pattern, echogenicity, branch size, presence of hyperechoic foci, periligamentar tissue thickening and proximal sesamoid bone contour at the branch insertion. Each branch was assigned grades for fibrillar pattern (graded 0-3), hyperechoic foci (0-2) and sesamoid bone surface (0-1) as per Table 5.1, adapted from previous studies<sup>28,29,31</sup>. Examples of the ultrasonographic appearance of each grade are shown in Figures 5.1-5.3. Findings per suspensory branch also consisted of three measured variables: insertional branch cross-sectional area (CSA, cm<sup>2</sup>), branch width (cm) and periligamentar tissue thickness (PLT, cm); and three calculated values: PLT as a percentage of branch width, percent difference in CSA between left and right lateral branches and percent difference in CSA between left and right medial branches (Avella 2009). Measured variables were acquired in transverse orientation at the insertional level at which the sesamoid bone first appears, as depicted in Ramzan *et al*'s Figure 2<sup>28</sup>, and the mean of three measurements was used. CSA measurements traced the visible perimeter of the branch and width was the linear thickness of the branch in the lateromedial dimension at the insertion. PLT was defined as the depth of subcutaneous echogenic material and was measured as the linear distance from the dermis to the palmar margin of the branch. The location of any fibrillar disruption within each branch was recorded using an appropriate combination of any of the following variables: dorsal/palmar/axial/central/abaxial. Images were viewed using open source DICOM software (Horos, version 3.3.6). Measurements made on DICOM images were

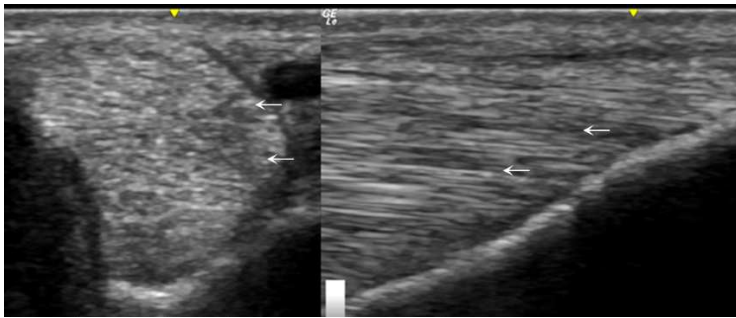
calibrated using the Horos calibration tool with the image's depth gauge prior to measurement. Measurements made on transverse still images from cine loops were calibrated using scientific image processing software (ImageJ/Fiji, version 2.1.0).

TABLE 5.1. Grading system for the ultrasonographic appearance of the suspensory ligament branches and the adjacent aspect of the proximal sesamoid bone in the forelimbs of yearling and 2-year-old Thoroughbred sales horses. Adapted from previously published work<sup>28,29,31</sup>.

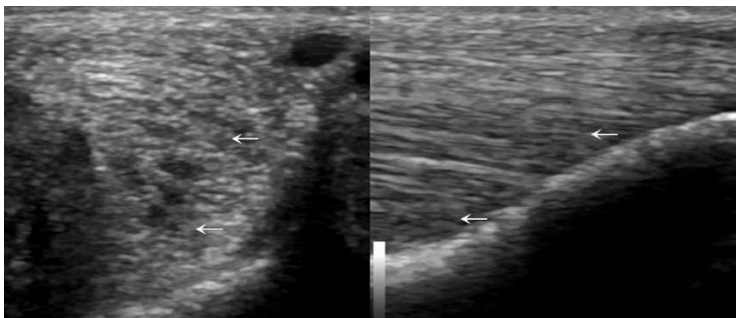
<b>GRADE</b>	<b>DEFINITION</b>
<b>Fibrillar Pattern</b>	
0	Homogenous echogenicity and uninterrupted fibrillar pattern
1	Regions of mild hypoechogenicity and/or small areas of subtly irregular fibrillar pattern
2	Regions of moderate hypoechogenicity and/or focal areas of mild-moderately disrupted fibrillar pattern
3	Regions of severe hypoechogenicity/anechoic areas and/or extensive fibrillar disruption, large core lesions
<b>Hyperechoic Foci</b>	
0	No hyperechoic foci visible within the ligament
1	Mild to moderately hyperechoic areas within the ligament that do not cause acoustic shadowing i.e., small, non-shadowing flecks or linear hyperechoic areas running parallel with the ligament fibres
2	Dystrophic mineralisation or fragment i.e., a defined hyperechoic focus within the ligament that inhibits acoustic passage, producing acoustic shadowing
<b>Sesamoid Bone Surface</b>	
0	Normal smooth surface of the proximal sesamoid bone at the level of the suspensory ligament branch insertion
1	Ultrasonographic evidence of bone irregularity or disruption/loss of the normal smooth surface of the proximal sesamoid bone at the suspensory insertion



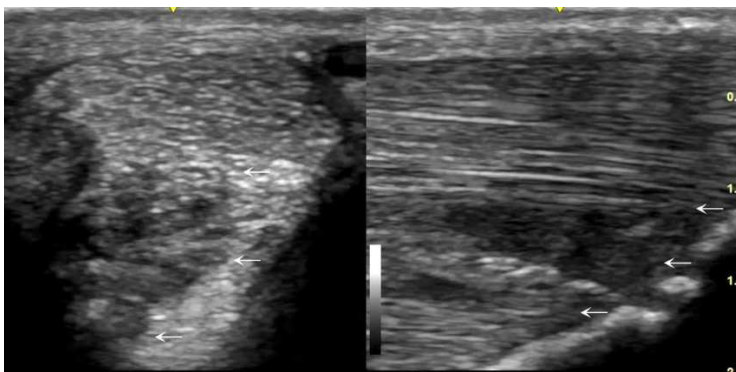
(A)



(B)



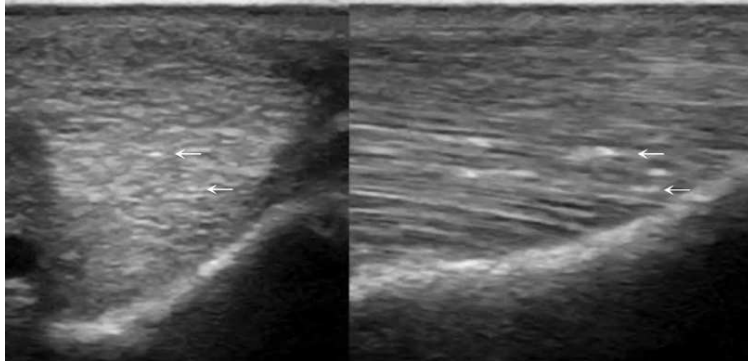
(C)



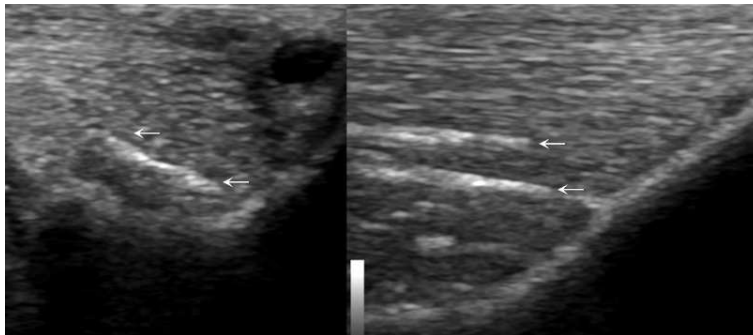
(D)

Figure 5.1. Examples of the grades used in this study for the ultrasonographic appearance of the fibrillar pattern in the insertional region of the equine suspensory ligament branch. (A) Grade 0 fibrillar pattern: homogenous echogenicity and uninterrupted fibrillar pattern. (B) Grade 1 fibrillar pattern: Regions of mild hypoechoogenicity and/or small areas of subtly irregular fibrillar pattern, seen here as an indistinct

hypoechoic region in the abaxial portion of the branch (arrows). (C) Grade 2 fibrillar pattern: Regions of moderate hypoechogenicity and/or focal areas of mild-moderately disrupted fibrillar pattern (arrows). (D) Grade 3 fibrillar pattern: Regions of severe hypoechogenicity/anechoic areas and/or extensive fibrillar disruption, large core lesions (arrows).

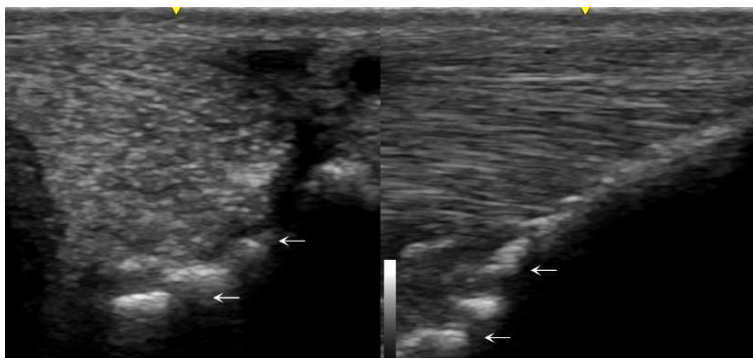


(A)

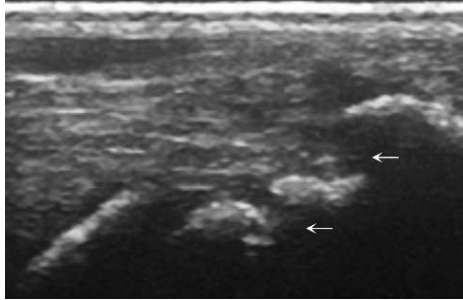


(B)

Figure 5.2. Examples of the grades used in this study for the ultrasonographic appearance of hyperechoic foci seen in the insertional region of the equine suspensory ligament branch. (A) Grade 1 hyperechoic foci: Mild to moderately hyperechoic areas within the ligament that do not cause acoustic shadowing, i.e., small, non-shadowing flecks or linear hyperechoic areas running parallel with the ligament fibres (arrows). (B) Grade 2 hyperechoic foci: Defined hyperechoic foci within the ligament with acoustic shadowing, seen here in the palmar aspect of the branch insertion (arrows).



(A)



(B)

Figure 5.3. (A) Grade 1 sesamoid bone surface: Ultrasonographic evidence of bone irregularity or disruption to the proximal sesamoid bone surface at the insertion of the equine suspensory ligament branch (arrows). (B) Severe bone disruption evident at the palmar aspect of the branch insertion (arrows).

#### *5.3.4 Racing performance*

Racing performance data from age two until the end of the four-year-old racing season, spanning 1 January 2017 to 31 December, 2019, was obtained from Equibase Company LLC as previously described for this body of Thoroughbred sales research<sup>48,49</sup>. Relevant information for horses that did not start a race was obtained through communication with the horse's connections, as previously described<sup>48</sup>. Eight racing performance outcome measures were used, beginning with whether the horse started at least one race by the end of their four-year-old year. For horses that raced, additional performance measures were: age in days old at first start, total number of starts, total earnings, earnings per start, class of best start, weighted Listed and Group starts and Class Performance Index. Earnings were recorded in United States Dollars (USD) and international earnings were converted to USD using the exchange rate on the date of racing.

#### *5.3.5 Non-starter follow-up information*

Any horse that did not start at least one race was considered a non-starter. Follow-up information for non-starters was sought via email and telephone communication with the connections of each horse, using a flow-chart conversation framework that was designed to obtain increasing levels of detail while minimising bias (Appendix 3).

#### *5.3.6 Data Analysis*

Data analyses were performed via consultation with statisticians using the statistical software package R, version 4.0.2 by RStudio with packages *irr*<sup>38</sup>.

At the individual suspensory branch level, ultrasonographic findings were summarised using descriptive statistics with frequency distributions by age, by sex, by medial and lateral, and by left and right side, including percentages and measures of uncertainty representing 95% confidence intervals. At the horse level, ultrasonographic findings were summarised using the highest score for each graded variable and the largest measurement and ratios from any of the four suspensory branches, i.e., the most severe findings per horse. For horses scanned as both yearlings and 2-year-olds, differences between ultrasonographic findings in a given suspensory branch at one and two years of age were examined using descriptive statistics.

Ultrasonographic findings examined for associations with racing performance were: maximum grade per horse for each of fibrillar pattern, hyperechoic foci and sesamoid bone contour; largest measurement per horse for each of branch CSA, branch width and PLT; largest value per horse for each of: PLT as a percentage of branch width, percent difference between left lateral branch CSA and right lateral branch CSA and percent difference between left medial branch CSA and right medial branch CSA. All ultrasonographic findings examined for associations with the eight racing performance measures listed in Section 2.1 were from 2-year-old ultrasound data. Multivariate generalised regression approaches were used. Analysis of the outcome “started at least one race” considered all 2-year-old horses. The remaining outcomes considered only 2-year-olds that raced. Binary outcomes, “started at least one race” and “best start” were analysed using logistic regression; “number of starts” was considered with negative binomial regression; and the remaining continuous outcomes were assessed using standard linear regression. Model diagnostics were performed and data transformations were used when necessary to satisfy model assumptions, e.g. normally distributed errors. “Days to first start” was log transformed to resemble an accelerated failure time model. Earnings related measures required log transformation, with all \$0 earnings, and \$0 earnings per start, set to \$1. “Class Performance Index” required log transformation with all CPI of zero set to 0.05, which is half the minimum performance value of 0.1. “Weighted Listed and Group” starts required square root transformation.

To build the final regression model, each racing outcome was first analysed in a multivariable regression against each horse-level ultrasonographic variable, always controlling for horse sex as a fixed

effect. Each ultrasonographic variable was then “screened” against that racing outcome using a likelihood ratio (logistic or negative binomial regression analyses) or F-test (linear regression analyses). Inclusion thresholds of  $P < 0.1$  were used to reduce the rate of false positives. No other adjustments were made to p-values to account for a large number of tests. All ultrasonographic variables that passed screening were simultaneously included in a multivariate regression analysis for that racing performance outcome. Ultrasonographic variables were retained by backwards selection for the final model, using a threshold level of 5% ( $P < 0.05$ ) to indicate statistical significance. Coefficient estimates and 95% confidence intervals produced by regression analyses were “back transformed” to provide estimates that pertained to the original scale of the racing performance data, describing the average value of the outcome for each level of ultrasonographic finding<sup>12,48</sup>.

## **5.4 Results**

### *5.4.1 Study enrolment*

Ultrasonography of the forelimb suspensory branches was performed with consignor consent on 593 sales yearlings immediately prior to the 2016 Keeneland September Yearling Sale in Lexington, Kentucky. Permission to ultrasound at the subsequent 2-year-old sales was granted for 415 horses and ultrasonography was logistically able to be performed on 367 sales 2-year-olds. Of these, 97 were horses that had ultrasonography prior to both the yearling and 2-year-old sales.

### *5.4.2 Graded ultrasonographical findings*

#### *i. Suspensory branch fibrillar pattern*

Overall, 88.1% of yearling forelimb suspensory branches had a grade 0 fibrillar pattern (2089 of 2372), 9.0% had a grade 1 fibrillar pattern (213 of 2372), 2.6% had a grade 2 fibrillar pattern (62 of 2372) and 0.3% had a grade 3 fibrillar pattern (8 of 2372). In two-year-olds, 81.5% of the forelimb suspensory branches had a grade 0 fibrillar pattern (1197 of 1468) and 13.4% had a grade 1 fibrillar pattern (197 of 1468). 4.0% of two-year-old forelimb suspensory branches had a grade 2 fibrillar pattern (59 of 1468) and 1.0% had a grade 3 fibrillar pattern (15 of 1468).

The distribution of fibrillar pattern grades between medial and lateral forelimb suspensory branches for each age group are shown in Table 5.2. The distribution of fibrillar pattern grades between left and right forelimbs for each age group are shown in Table 5.3. A comparison of fibrillar pattern grades between colts/geldings and fillies for each age group is shown in Table 5.4. When considering the highest fibrillar pattern grade per horse, 1.0% of yearlings and 4.1% of 2-year-olds had at least one grade 3 suspensory branch (6 of 593, 15 of 367 respectively). Horse-level maximum fibrillar pattern grades are shown in Table 5.5.

TABLE 5.2. Distribution of ultrasonographical fibrillar pattern grades by location, comparing medial and lateral forelimb suspensory branches, in Thoroughbred sales yearlings and 2-year-olds.

Ultrasonographic fibrillar pattern	Yearlings, N (%)		Two-year-olds, N (%)	
	Medial branches	Lateral branches	Medial branches	Lateral branches
Grade 0	981 (82.7%)	1108 (93.4%)	559 (76.2%)	638 (86.9%)
Grade 1	159 (13.4%)	54 (4.6%)	133 (18.1%)	64 (8.7%)
Grade 2	40 (3.4%)	22 (1.9%)	32 (4.4%)	27 (3.7%)
Grade 3	6 (0.5%)	2 (0.2%)	10 (1.4%)	5 (0.7%)
Total	1186 (100%)	1186 (100%)	734 (100%)	734 (100%)

TABLE 5.3. Distribution of ultrasonographic forelimb suspensory ligament branch fibrillar pattern grades by side, comparing left and right forelimbs, in Thoroughbred sales yearlings and 2-year-olds.

Ultrasonographic fibrillar pattern	Yearlings, N (%)		Two-year-olds, N (%)	
	Left forelimb	Right forelimb	Left forelimb	Right forelimb
Grade 0	1036 (87.4%)	1053 (88.8%)	597 (81.3%)	600 (81.7%)
Grade 1	111 (9.4%)	102 (8.6%)	101 (13.8%)	96 (13.1%)
Grade 2	33 (2.8%)	29 (2.4%)	28 (3.8%)	31 (4.2%)
Grade 3	6 (0.5%)	2 (0.2%)	8 (1.1%)	7 (1.0%)
Total	1186 (100%)	1186 (100%)	734 (100%)	734 (100%)

TABLE 5.4. Distribution of ultrasonographic forelimb suspensory ligament branch fibrillar pattern grades by horse sex, comparing colts/geldings and fillies, in Thoroughbred sales yearlings and 2-year-olds.

Ultrasonographic fibrillar pattern	Yearlings, N (%)		Two-year-olds, N (%)	
	Colts	Fillies	Colts/geldings	Fillies
Grade 0	1086 (87.3%)	1003 (88.9%)	634 (77.7%)	563 (86.3%)
Grade 1	128 (10.3%)	85 (7.5%)	129 (15.8%)	68 (10.4%)
Grade 2	27 (2.2%)	35 (3.1%)	43 (5.3%)	16 (2.5%)
Grade 3	3 (0.2%)	5 (0.4%)	10 (1.2%)	5 (0.8%)
Total	1244 (100%)	1128 (100%)	816 (100%)	652 (100%)

TABLE 5.5. Distribution of maximum forelimb suspensory ligament branch fibrillar pattern grades per horse in Thoroughbred sales yearlings and 2-year-olds.

Maximum fibrillar pattern grade per horse	Yearlings, N (%)	Two-year-olds, N (%)
0	410 (69.1%)	205 (55.9%)
1	130 (21.9%)	109 (29.7%)
2	47 (7.9%)	38 (10.4%)
3	6 (1.0%)	15 (4.1%)
Total horses	593 (100%)	367 (100%)

*ii. Fibrillar pattern changes between yearling and 2-year-old sale age*

Ninety-eight horses had ultrasonography prior to both the yearling and 2-year-old sales, resulting in 392 suspensory branches with paired ultrasounds. Of suspensory branches that had a grade 0 fibrillar pattern at the yearling sale, 2.1% had a grade 2 or 3 fibrillar pattern at 2-year-old sale ultrasonography (7 of 336). Of suspensory branches that had a grade 1 pattern at the yearling sale, 7.0% had a grade 2 or 3 pattern at a 2-year-old sale (3 of 43). Of suspensory branches that had a grade 2 pattern at the yearling sale, 38.5% had a grade 2 or 3 pattern at a 2-year-old sale (5 of 13). None of the horses with paired sales ultrasounds had a grade 3 suspensory branch fibrillar pattern as a yearling. The full results are shown in Table 5.6 and depicted in Figure 5.4.

TABLE 5.6. Distribution of ultrasonographic forelimb suspensory ligament branch fibrillar pattern grades in 2-year-old horses (columns), relative to each branch's grade at yearling age (rows), for 392 suspensory branches from 98 horses with paired yearling and 2-year-old sales ultrasonography. Shaded cells represent grades that remained static between sales. Percentages refer to row totals, i.e. the proportion of the respective yearling grade distributed across grades 0-3 at 2-year-old age.

Suspensory branch fibrillar pattern grade at yearling sale	Suspensory branch fibrillar pattern grade at 2-year-old sale				
	Two-year-old Grade 0	Two-year-old Grade 1	Two-year-old Grade 2	Two-year-old Grade 3	Total suspensory branches
Yearling Grade 0	288 (85.7%)	41 (12.2%)	5 (1.5%)	2 (0.6%)	336 (100%)
Yearling Grade 1	27 (62.8%)	13 (30.2%)	3 (7.0%)	0 (0.0%)	43 (100%)
Yearling Grade 2	5 (38.5%)	3 (23.1%)	1 (7.7%)	4 (30.8%)	13 (100%)
Yearling Grade 3	0	0	0	0	0
Total suspensory branches	320 (81.6%)	57 (14.5%)	9 (2.3%)	6 (1.5%)	392 (100%)

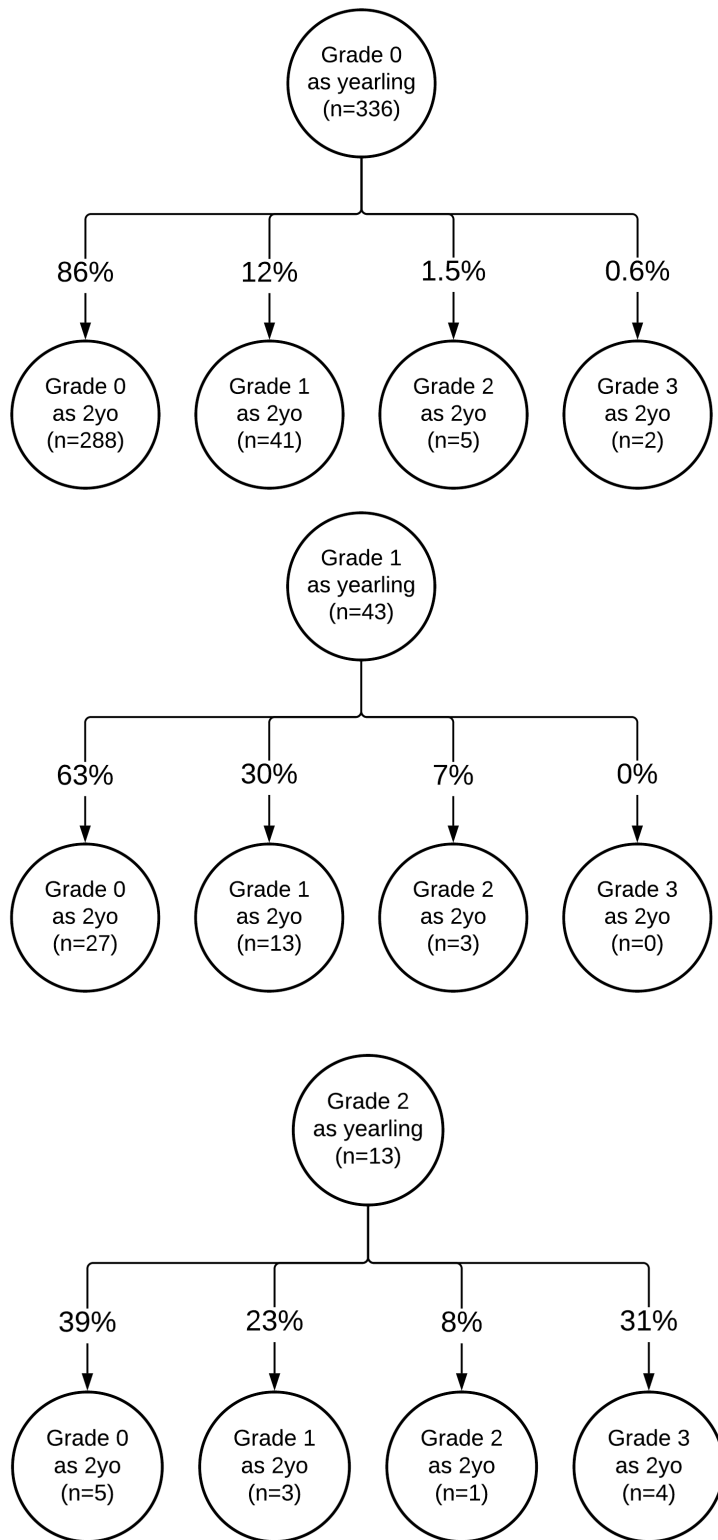


FIGURE 5.4. Proportions of forelimb suspensory ligament branches with grade 0, grade 1 and grade 2 fibrillar patterns as yearlings that progressed, regressed or remained unchanged in grade, for horses that underwent presale ultrasonography at both yearling and 2-year-old sales.

*iii. Suspensory branch hyperechoic foci*

The presence of hyperechoic foci in medial and lateral forelimb suspensory branches as detected at presale ultrasonography is detailed in Table 5.7. When considering the highest hyperechoic foci grade per horse, 2.2% of yearlings and 7.6% of two-year-olds had a forelimb suspensory branch containing grade 2 hyperechoic foci (13/593 and 28/367, respectively). The horse-level maximum hyperechoic foci grades are shown in Table 5.8.

TABLE 5.7. Distribution of ultrasonographic hyperechoic foci grades by location, comparing medial and lateral forelimb suspensory ligament branches, in Thoroughbred sales yearlings and 2-year-olds.

Hyperechoic foci	Yearling suspensory branches, N (%)		Two-year-old suspensory branches, N (%)	
	Medial forelimb branches	Lateral forelimb branches	Medial forelimb branches	Lateral forelimb branches
Grade 0	1076 (90.7%)	1132 (95.4%)	640 (87.2%)	652 (88.8%)
Grade 1	100 (8.4%)	50 (4.2%)	81 (11.0%)	62 (8.4%)
Grade 2	10 (0.8%)	4 (0.3%)	13 (1.8%)	20 (2.7%)
Total	1186 (100%)	1186 (100%)	734 (100%)	734 (100%)

TABLE 5.8. Distribution of maximum forelimb suspensory ligament branch hyperechoic foci grades per horse in Thoroughbred sales yearlings and 2-year-olds via ultrasonographic examination presale.

Maximum forelimb suspensory branch hyperechoic foci grade	Yearlings, N (%)	Two-year-olds, N (%)
0	472 (79.6%)	250 (68.1%)
1	108 (18.2%)	89 (24.3%)
2	13 (2.2%)	28 (7.6%)
Total horses	593 (100%)	367 (100%)

*iv. Proximal sesamoid bone abaxial surface contour*

9.2% of yearling forelimb suspensory branches (218/2372) and 13.4% of two-year-old forelimb suspensory branches (197/1468) had irregular sesamoid bone contour detected ultrasonographically at the branch insertion. Table 5.9 compares the presence of irregular sesamoid bone contour as detected ultrasonographically at the medial and lateral suspensory branch insertions in each age group. Comparing left and right forelimbs, the prevalence of irregular sesamoid bone contour was similar for each limb side.

In yearlings, the prevalence of irregular sesamoid bone contour seen ultrasonographically in the left and right forelimbs was 9.7% (115/1186) and 8.7% (103/1186), respectively. In 2-year-olds, the respective left and right forelimb prevalences were 13.8% (101/734) and 13.1% (96/734). When considering the most severe sesamoid bone contour grade from all four forelimb suspensory branches in each horse, 24.6% of yearlings and 37.3% of two-year-olds had at least one branch insertion with irregular sesamoid bone contour evident ultrasonographically (146/593 and 137/367, respectively).

TABLE 5.9. Distribution of the presence (Grade 1) and absence (Grade 0) of irregular proximal sesamoid bone contour as detected ultrasonographically in Thoroughbred sales yearlings and 2-year-olds, comparing medial and lateral forelimb suspensory ligament branches.

Ultrasonographic sesamoid bone contour	Yearling suspensory branches, N (%)		Two-year-old suspensory branches, N (%)	
	Medial	Lateral	Medial	Lateral
Grade 0 (regular)	1029 (86.8%)	1125 (94.9%)	616 (83.9%)	655 (89.2%)
Grade 1 (irregular)	157 (13.2%)	61 (5.1%)	118 (16.1%)	79 (10.8%)
Total	1186 (100%)	1186 (100%)	734 (100%)	734 (100%)

#### 5.4.3 Measured ultrasonographical findings

##### *i. Suspensory branch cross-sectional area*

In yearlings, the median cross-sectional area of medial forelimb suspensory branches was 1.20 cm<sup>2</sup> and of lateral forelimb suspensory branches was 1.12 cm<sup>2</sup> (medial IQR 1.14-1.28, lateral IQR 1.05-1.19). Likewise in two-year-olds, the median cross-sectional area of medial forelimb suspensory branches was 1.21 cm<sup>2</sup> and of lateral forelimb suspensory branches was 1.12 cm<sup>2</sup> (medial IQR 1.14-1.30, lateral IQR 1.05-1.20). The entire range of forelimb suspensory branch cross-sectional area measurements presale spanned from a minimum of 0.86 cm<sup>2</sup> to a maximum of 2.14 cm<sup>2</sup>.

Comparison of medial and lateral suspensory branch CSA respectively, between left and right forelimbs revealed a median difference of 5.0% in each branch in both yearling and two-year-old populations. No significant difference was detected in the average maximum branch cross-sectional area per horse between yearlings and two-year-olds ( $P = 0.3$ , 95% CI: -0.02, 0.06). In 384 suspensory branches from 96 horses with measurements at both ages, the mean difference between two-year-old and yearling

branch cross-sectional area was -0.01 (sd 0.15). The distributions of branch CSA measurements for each grade of fibrillar pattern in yearlings and in 2-year-olds are depicted in Figure 5.5a,b.

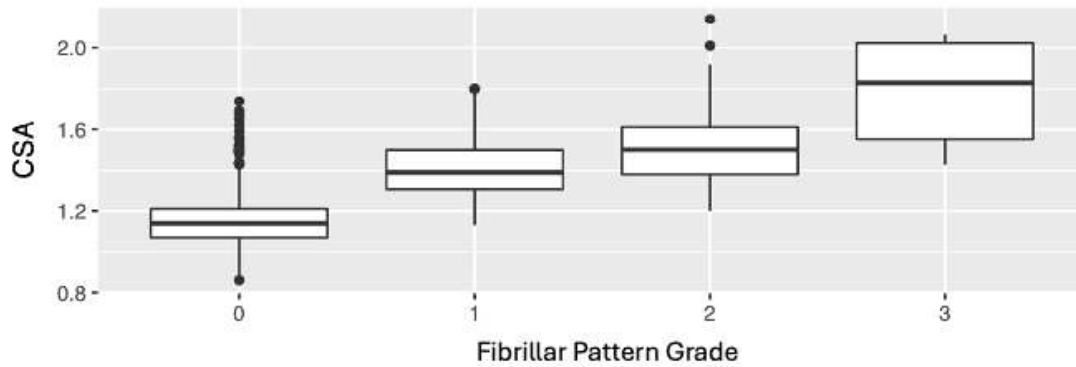


FIGURE 5.5a. Yearling distribution summary of suspensory ligament branch ultrasonographic measurements for cross-sectional area (CSA) in cm<sup>2</sup> by fibrillar pattern grade (0-3) for all forelimb branches

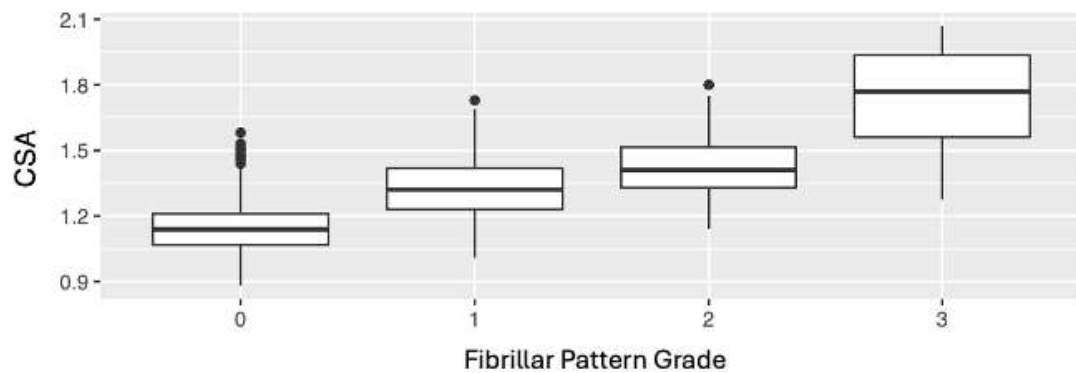


FIGURE 5.5b. Two-year-old distribution summary of suspensory ligament branch ultrasonographic measurements for cross-sectional area (CSA) in cm<sup>2</sup> by fibrillar pattern grade (0-3) for all forelimb branches

*ii. Suspensory branch width*

In yearlings, the median width of medial forelimb suspensory branches was 1.34 cm and of lateral forelimb suspensory branches was 1.23 cm (medial IQR 1.25-1.43, lateral IQR 1.14-1.30). In 2-year-olds, the median width of medial forelimb suspensory branches was 1.38 cm and of lateral forelimb suspensory branches was 1.26 cm (medial IQR 1.28-1.50, lateral IQR 1.17-1.34). No significant difference was detected in the maximum branch width per horse between yearlings and two-year-olds ( $P = 0.2$ , 95% CI: -0.063, 0.014). There was also no difference between the median branch width in left forelimbs compared

to right forelimbs in yearlings or in 2-year-olds. The distributions of branch width measurements for each grade of fibrillar pattern in yearlings and in 2-year-olds are depicted in Figure 5.6 a,b.

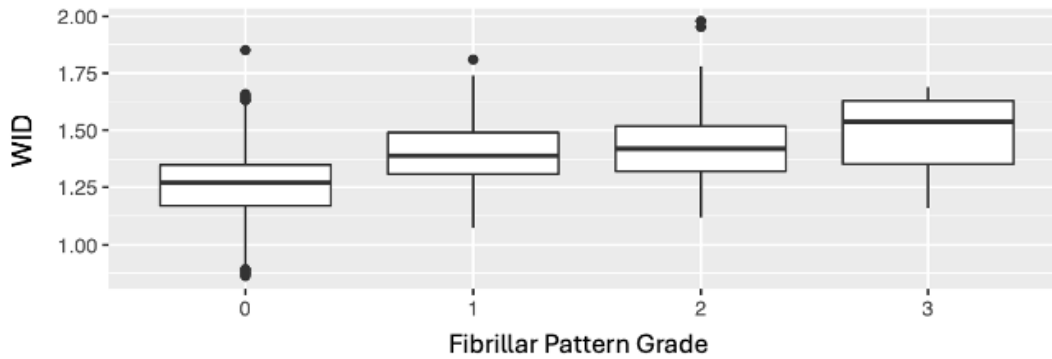


FIGURE 5.6a. Yearling distribution summary of ultrasonographic suspensory ligament branch width (WID) in cm by fibrillar pattern grade (0-3) for all forelimb branches

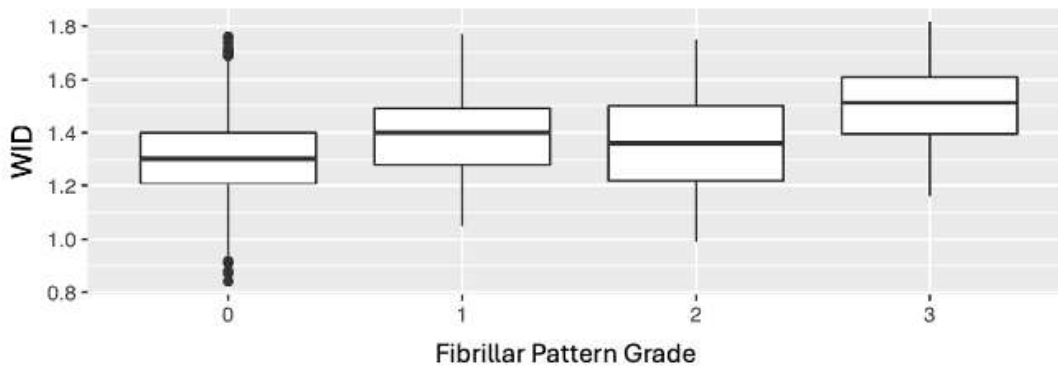


FIGURE 5.6b. Two-year-old distribution summary of ultrasonographic suspensory ligament branch width (WID) in cm by fibrillar pattern grade (0-3) for all forelimb branches

*iii. Periligamentar tissue thickness*

The median PLT for all forelimb suspensory branches was 0.10 cm in both yearlings and 2-year-olds (yearling IQR 0.08-0.11, 2-year-old IQR 0.09-0.12). The proportion of PLT to branch width had a median of 0.08, or 8.0%, in both yearlings and two-year-olds for all forelimb suspensory branches (yearling IQR 0.06-0.09, 2-year-old IQR 0.07-0.09). The distributions of PLT ratio for each grade of branch fibrillar pattern in yearlings and in 2-year-olds are depicted in Figure 5.7a,b.

Periligamentar tissue thickness did not increase with increasing fibrillar pattern grade in yearlings, except for two individual yearlings with severe grade 3 lesions. Increased relative periligamentar tissue thickness was only a feature in some 2-year-olds with grade 3 lesions.

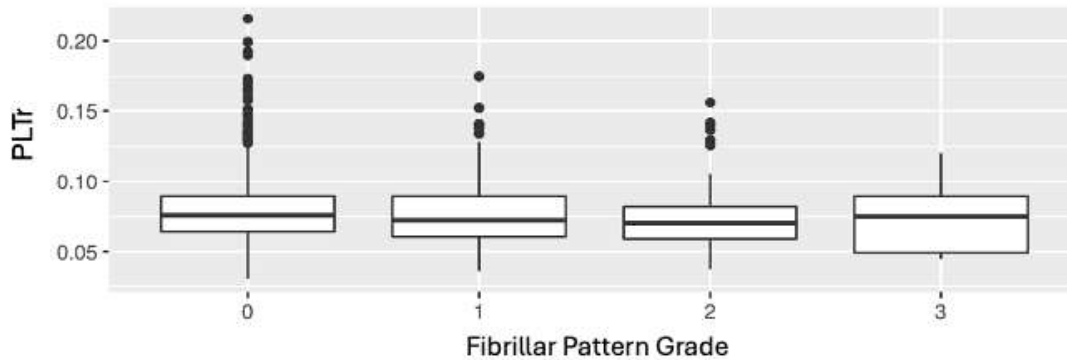


FIGURE 5.7a. Yearling distribution summary of the proportion of periligamentar tissue thickness to suspensory ligament branch width (PLTr), as measured ultrasonographically, by fibrillar pattern grade (0-3) for all forelimb branches

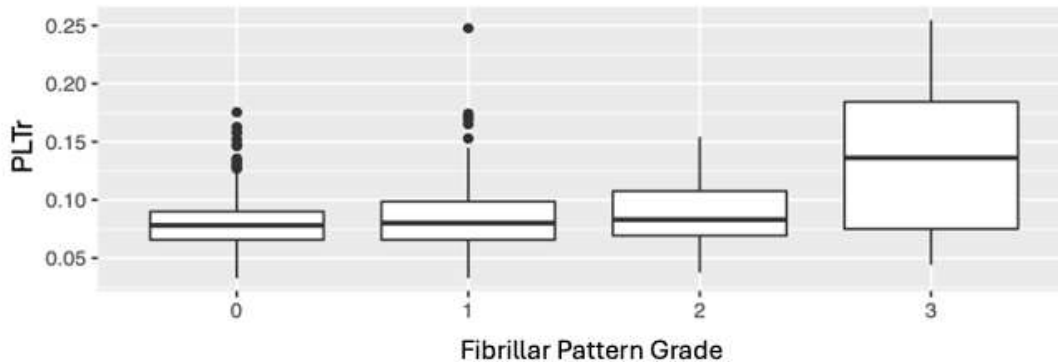


FIGURE 5.7b. Two-year-old distribution summary of the proportion of periligamentar tissue thickness to suspensory ligament branch width (PLTr), as measured ultrasonographically, by fibrillar pattern grade (0-3) for all forelimb branches

#### 5.4.4 Associations with racing performance

##### *i. Race starts*

Overall, 89.9% of horses with two-year-old sale ultrasounds started at least one race by the end of their four-year-old racing season (330/367). For horses that raced, the median total number of race starts from two to four years of age was 12 (IQR 7-18 starts). No statistically significant differences were found between any ultrasonographic variables and the probability of horses starting at least one race (Table 5.10).

Of the 15 horses with a Grade 3 fibrillar pattern, 80.0% started at least one race. For horses with a maximum fibrillar pattern of Grade 0-2, 89.8-92.1% started at least one race. This difference was not statistically significant (likelihood ratio  $P = 0.7$ ).

TABLE 5.10. The proportion of horses that raced for each level of maximum ultrasonographic finding in the forelimb suspensory ligament branches at two-year-old sales and the probability of racing from the logistic regression model controlling for horse sex. Note: Displayed are the overall number of 2-year-olds with a given maximum ultrasonographic finding for each ultrasonographic variable, the percentage of those horses that raced and the back-transformed probability of racing based on the logistical model controlling for horse sex.

	Grade	Total horses	N (%) of horses that raced	Probability of racing (95% CI)
Fibrillar pattern	0	205	184 (89.8%)	0.90 (CI: 0.85-0.93)
	1	109	99 (90.8%)	0.91 (CI: 0.83-0.95)
	2	38	35 (92.1%)	0.92 (CI: 0.77-0.97)
	3	15	12 (80.0%)	0.80 (CI: 0.52-0.93)
Hyperechoic foci	0	250	227 (90.8%)	0.91 (CI: 0.87-0.94)
	1	89	79 (88.8%)	0.89 (CI: 0.80-0.94)
	2	28	24 (85.7%)	0.85 (CI: 0.65-0.94)
Bone surface	0	230	209 (90.9%)	0.91 (CI: 0.86-0.94)
	1	137	121 (88.3%)	0.88 (CI: 0.81-0.93)

*ii. Follow-up of non-starters*

Follow-up information was obtained for 48.6% of non-starters (18/37). Eight non-starters had complete follow-up information, that is, a confirmed reason for never racing was obtained. Of these 8 horses: 4 did not race due to lameness, 2 did not race due to fatal or near fatal accidents unrelated to performance, 1 was deemed to have insufficient athletic ability and 1 did not race due to colic requiring euthanasia. The clinical diagnoses in the 4 non-starters with lameness comprised 3 horses with bowed flexor tendons and 1 horse with a fractured hind sesamoid. For 10 non-starters, the follow-up was partial; meaning that confirmation was given that the horse never raced but the reason for not racing was unknown, or the horse was exported from the United States and the racing history was inaccessible. Of the combined 29 non-starters with either partial or no follow-up, 15 horses had only Grade 0 suspensory branch fibrillar

patterns, 9 horses had at worst a Grade 1 branch, 2 horses had a Grade 2 branch and 3 horses had a Grade 3 branch.

*iii. Age at first race start*

For horses that did race, the median age at first race start was 973 days old, i.e. 2 years and 8 months old (IQR 891-1073 days). Maximum branch width was significantly associated with an older age at first race start. On average in two-year-olds, a 0.25 cm increase in branch width above average was associated with a 49-day delay in initiating a racing career ( $P<0.001$ , 95% CI: 21-77 days).

*iv. Earnings*

For horses that raced, median total earnings to the end of the 4-year-old racing season were \$38,622 (IQR \$14,633-\$86,422). The presence of Grade 2 hyperechoic foci in at least one forelimb branch was significantly associated with lower total earnings (\$5662,  $P=0.01$ ). The back-transformed estimated total earnings for each grade of ultrasonographic finding are presented in Table 5.11. Grade 3 fibrillar pattern passed the screening threshold for lower total earnings (F-test  $P=0.03$ ) but was not retained in the final multiple regression model ( $P=0.06$ ).

Per start, horses that raced earned a median of \$3375 in each race (IQR \$1470-\$6723). Two ultrasonographic variables were significantly associated with lower earnings per start: Grade 2 hyperechoic foci and maximum branch width ( $P=0.003$  and  $P<0.05$ , respectively). Horses with a maximum fibrillar pattern of Grade 1 were associated with significantly higher earnings per start ( $P=0.004$ ). The back-transformed estimated earnings per start for each grade of ultrasonographic finding are presented in Table 5.11.

TABLE 5.11. Estimated total prizemoney earnings and earnings per race start (USD) for each level of maximum ultrasonographic forelimb suspensory ligament branch finding in 2-year-old horses that subsequently raced, from the multivariate linear regression model controlling for horse sex. ^denotes a statistically significant difference ( $P<0.05$ ).

	Grade	Total earnings (95% CI)	Earnings per start (95% CI)
Fibrillar pattern	0	\$17,900 (CI: 12,366-25,912)	\$2,094 (CI: 1,576-2,783)
	1	\$40,983 (CI: 24,681-68,053)	\$3,900^ (CI: 2,641-5,759)
	2	\$13,932 (CI: 5,915-32,811)	\$1,658 (CI: 858-3,202)
	3	\$12,319 (CI: 2,875-52,787)	\$1,638 (CI: 535-5,013)
Hyperechoic foci	0	\$22,041 (CI: 15,795-30,757)	\$2,428 (CI: 1,881-3,134)
	1	\$32,348 (CI: 18,388-56,905)	\$3,423 (CI: 2,221-5,277)
	2	\$5,662^ (CI: 2,000-16,022)	\$775^ (CI: 349-1,718)
Bone surface	0	\$23,681 (CI: 16,680-33,620)	\$2,526 (CI: 1,929-3,307)
	1	\$19,377 (CI: 12,138-30,933)	\$2,293 (CI: 1,600-3,285)

#### *v. Race caliber*

Of all horses that raced, 17.9% achieved at least one Listed or Group level race start (59/330). Included among horses achieving a race of this calibre were: 16.8% of horses with a Grade 0 maximum fibrillar pattern (31/184), 22% of horses with a Grade 1 maximum fibrillar pattern (22/99), 14% of horses with a Grade 2 maximum fibrillar pattern (5/35) and 8% of horses with a Grade 3 maximum fibrillar pattern (1/12). No significant associations were found between ultrasonographic variables and race calibre using the measures Career Best Start or Weighted Listed and Group Starts. A significant association was found between Class Performance Index and difference in left and right medial branch CSA ( $P=0.04$ ).

## **5.5 Discussion**

This is the first time that the ultrasonographic appearance of the suspensory ligament branches has been investigated at yearling and 2-year-old sales. Reference values, specific to young Thoroughbreds, have been established for branch size measurements and for the prevalence of ultrasonographic findings at sales. Previously unavailable information has also been produced after prospectively following these horses to determine associations with racing performance. The results indicate that some findings should be regarded as an acceptable ultrasonographic appearance in sales Thoroughbreds, especially Grade 1 branch fibrillar

pattern. The results have also identified certain findings that do constitute an evidence-based risk of delayed or reduced performance, namely enlarged branch size as measured by width and Grade 2 hyperechoic foci.

The vast majority of individual suspensory branches in both age groups had a Grade 0 fibrillar pattern (85%). However, when examined at the horse level, 22% of yearlings and 30% of 2-year-olds had, at worst, one or more Grade 1 branches. This indicates that ultrasonographic changes, particularly Grade 1 fibrillar pattern, are seen in the suspensory branches of a considerable proportion of sales horses. Medial branches were more frequently affected than lateral branches at all grades above Grade 0, which is consistent with previous work finding an overrepresentation of medial branch changes<sup>28,29,33</sup>. No asymmetry was observed in the prevalence of left compared to right forelimb branches affected by each grade of fibrillar change. In fact, the distribution of grades by limb side was remarkably similar, with no more than a one percent difference per side for any grade, at either age. When comparing findings by horse sex in yearlings, more colts had Grade 1 branch fibrillar pattern than fillies. In 2-year-olds, colts/geldings had a higher prevalence of all grades of fibrillar pattern  $\geq 1$ , compared to fillies.

Among the horses that presented for sale at both ages, over 95% of yearling Grade 0 and Grade 1 maximum fibrillar pattern findings either improved or remained unchanged in appearance by 2-year-old sale. This included resolution of 63% of yearling Grade 1 branches. These figures do not account for any horses that developed worsening change of a magnitude necessitating withdrawal from 2-year-old sale, due to anticipated impact on sale value or developing clinical injury. The majority of pinhooked yearlings would not have been monitored ultrasonographically between sales, unless a clinical reason to investigate the suspensory branches arose, or a potential concern was identified at yearling sales in the suspensory branch or proximal sesamoid bones<sup>48</sup>. Most of the latter cases are unlikely to have been selected as 2-year-old sale prospects. Consequently, it is reasonable to expect that, in horses deemed suitable for pinhooking at yearling sales time, many instances of worsening of branch fibrillar pattern that remained subclinical would have gone undetected and therefore would be represented in these figures. This provides a degree of evidence that suggests Grade 1 branches need not be heavily critiqued at yearling sales.

Analysis of racing performance suggested, likewise, that the presence of Grade 1 branches at 2-year-old sales was not detrimental. In a U.K. study of clinically normal flat racing Thoroughbreds, the majority of which were 2 and 3-year-olds, the prevalence of Grade 1 fibrillar pattern in forelimb branches was 26-28%<sup>28</sup>. This is eminently comparable to the prevalence of Grade 1 change found in the present study at 2-year-old sales. An unchanged prevalence from the initiation of training and sale through productive racing years suggests a baseline level of appearance, which may not be pathological in this population of horses. In the authors' opinion, it may be that the mild reduction in echogenicity seen in Grade 1 branches is related to adaptive change within normal limits in young Thoroughbreds. Racing performance analysis in the present study found that horses with a maximum branch fibrillar pattern of Grade 1 as 2-year-olds had significantly higher earnings per start compared to horses with only Grade 0 branches (\$3900 versus \$2094, respectively). Similar relationships, though not statistically significant, were seen with other racing outcomes whereby Grade 1 tended to outperform Grade 0. In agreement with previous authors who did not consider Grade 1 to be representative of subclinical injury<sup>28,34</sup>, the current results show that Grade 1 branches were common and no evidence was found to suggest they were detrimental to future racing performance.

In contrast to the milder grades, 61% of yearling Grade 2 branches had improved by 2-year-old sale but 31% had progressed to a grade 3 lesion. Grade 2 yearling branches are more likely to have been monitored during 2-year-old sales preparation and so the proportion that developed more severe change is likely underestimated here. No associations were found between Grade 2 branch fibrillar pattern and poorer racing performance. However, the evidence that approximately one-third of yearling Grade 2 branches may worsen to Grade 3 by 2-year-old sale is important, particularly as the cumulative training load and intensity continues to increase post-sale with the onset of racing careers.

Grade 3 fibrillar pattern showed a notable difference in the odds of racing and class achieved, despite not reaching statistical significance. The 80% of starters for Grade 3 branches, compared to 90-92% for Grades 0-2, is clinically relevant. Also of note is that only 8% of horses with a Grade 3 branch at 2-year-old sales went on to race at Listed or Group level, compared to 14-22% of horses with Grade 1 and Grade

2 branches at the same age. In the analysis of career earnings, Grade 3 fibrillar pattern passed screening for inclusion but was not retained in the final multiple regression model ( $P = 0.06$ ). In a previous study, low numbers of Grade 3 branches precluded specific analysis of racing performance after clinical injury<sup>33</sup>. With a total of 15 horses with 2-year-old sale Grade 3 branches in the present study, greater numbers may likewise have been useful, though in practice this severe a lesion is an uncommon finding at sales. No subclinical Grade 3 branches were found in Ramzan *et al*'s study of active racehorses<sup>28</sup>, possibly suggesting progression to clinical injury for branches that were subclinically Grade 3 prior to racing. The incidence of clinical branch injury developing in the first year of race training was established as 9.50-9.97% by Plevin and McLellan<sup>23,33</sup>.

The suspensory branches are a known predilection site for dystrophic mineralisation in the horse, which can be incidental and has also been linked to poor prognostic outcomes<sup>32,51</sup>. In humans with calcific tendonitis of the rotator cuff, larger calcifications and positive power Doppler signal within the calcific deposit have been identified as ultrasonographic indicators of clinical importance<sup>52</sup>. This is the first time that suspensory branch mineralisation has been analysed against flat racing performance in young Thoroughbreds. The results show that the presence of Grade 2 hyperechoic foci is clinically important in this population, with significantly reduced total earnings and earnings per start. The ultrasonographic appearance in these branches was that of complete inhibition of the acoustic passage deep to the hyperechoic focus, causing a shadowing effect within the branch. This is presumed consistent with ligamentous calcification or dystrophic mineralisation and included some sesamoid avulsion fragments that were adjacent to corresponding sesamoid bone defects. Dystrophic mineralisation has been considered representative of a degree of chronic branch change<sup>31</sup> and what is known of the molecular nature of the mineralisation process has been discussed<sup>32</sup>. In this study, 2-year-olds were 3.5 times more likely to have a branch with Grade 2 foci than yearlings. Within 2-year-olds, Grade 2 foci were over 4 times more prevalent in colts than fillies. It is critical to note that the same association was not found for Grade 1 hyperechoic foci, seen as small hyperechoic flecks scattered diffusely or in a concentrated region of the branch, without acoustic shadowing. Grade 1 foci were a common finding, with 18% of yearlings and 24%

of 2-year-olds shown to have at least one branch with Grade 1 foci. No evidence was found to suggest hyperechoic foci without acoustic shadowing were detrimental to future performance. Care should be taken when scanning to avoid misinterpretation of artefactual hyperechoic foci and to avoid overinterpretation of the Grade 1 appearance.

Reference measurements specific to young Thoroughbreds have been established for branch CSA and width, from the quantitative assessment of size in a total of 960 horses in the present study. Medial branches ( $1.20 \text{ cm}^2$ ) were found to be larger than lateral branches ( $1.12 \text{ cm}^2$ ) across all grades of fibrillar pattern and hyperechoic foci. This is consistent with previous reports<sup>29,53</sup> and underscores the importance of comparing size with the contralateral limb, bearing in mind the corresponding contralateral branch may also be enlarged. No difference was detected in the average CSA between yearlings and 2-year-olds, even with the onset of training. Colts had slightly larger branches than fillies in both age groups and sex was controlled for in the regression analyses. As expected, CSA increased with increasing fibrillar pattern grade and also with increasing hyperechoic foci grade. The median CSA values in the present study are within the published range for the distal portion of the branch in racing Thoroughbreds of  $1.00\text{-}1.20 \text{ cm}^2$ <sup>53</sup>. However, they are smaller than the CSA reported for National Hunt racehorses (medial  $1.42 \text{ cm}^2$ , lateral  $1.28 \text{ cm}^2$ )<sup>29</sup> and elite Warmblood showjumpers (medial  $1.52 \text{ cm}^2$ , lateral  $1.41 \text{ cm}^2$ )<sup>30</sup>. Both of these groups had a high incidence of subclinical grade  $\geq 2$  changes, reinforcing the importance of using population-specific reference values.

Branch width (lateromedial thickness) was included as an additional measure of branch size. A significant association was found between increasing branch width and older age at first start and reduced earnings per start. A 20% increase above average width (0.25 cm), which is widely considered to be a clinically relevant percentage<sup>54</sup>, was associated with a 50-day delay to the start of racing careers. CSA is typically the primary size measurement used in branch ultrasonography. However, the perimeter tracing of branch cross-sections for CSA is inherently more variable than a linear measurement between identifiable anatomical landmarks for width. Width was initially to be measured on longitudinal images<sup>55</sup>, but this was changed to measuring on transverse images because of a perceived variability in width on longitudinal

images depending on the position of the transducer. This method of measuring branch width has proven a useful adjunct to CSA when evaluating branch size.

Two ultrasonographic variables were not found to be of predictive value in this study. Periligamentar fibrosis is a feature of chronic suspensory branch injury, especially in hindlimbs, and is recognised as an indicator of poor prognosis in sport horses<sup>30,31</sup>. However, increased PLT was a rare finding in the forelimbs of yearling and 2-year-old sales horses. The median PLT was consistent across both age groups (0.10 cm) and the median proportion of PLT to branch width was established as 8% in this population. In contrast, the same proportion measured in sport horses with clinical branch injury was 32% in forelimbs and 42% in hindlimbs<sup>31</sup>. The present study demonstrated that sales Thoroughbreds with acute branch enlargement have unchanged PLT and this results in a decreased ratio of PLT to branch width. Periligamentous oedema or fibrosis has been noted in clinical branch injury in racing Thoroughbreds, including within the first year of training<sup>33,56</sup>. However, periligamentar fibrosis was not a feature in any yearlings in the present study, irrespective of fibrillar pattern grade. It was only apparent in a few 2-year-olds with Grade 3 fibrillar lesions. Thus, whilst PLT remains a valid and important feature to identify chronic branch change and an associated poor prognosis, there are other ultrasonographic features of greater utility in most young sales Thoroughbreds.

Irregularity of the abaxial sesamoid bone surface at the branch insertion was the other ultrasonographic variable that did not show predictive value in this study. Medial branches were overrepresented in both age groups. At the horse-level, 25% of yearlings and 37% of 2-year-olds had at least one branch insertion with irregular sesamoid bone contour identified ultrasonographically. There was no evidence of an association with racing performance outcomes. It may be that the range of irregularity detected ultrasonographically was too broad to be of use. When present at a radiologically evident level, the production of abaxial new bone on forelimb sesamoids was found to have a significant negative association with racing performance in this population of sales horses<sup>48</sup>.

Measures were taken to ensure that granting permission for ultrasonography would not impact horses' sale prospects. The absence of clipping did not compromise image quality in sales-ready horses

with fine hair coats. It became clear early in the course of yearling ultrasonography that consignors were more agreeable to ultrasonography involving the forelimbs only, as this could usually be performed unседated. Requiring intravenous sedation to safely obtain hindlimb images would have drastically reduced enrolment. Thus, the method was modified to include forelimbs only, as in previous work<sup>28,29,34</sup>. Forelimb branch injuries were more common than hindlimb branch injuries in young Thoroughbreds in race training, with a reported distributions of 69% to 31%<sup>23</sup> and 62% to 38%<sup>33</sup>, respectively. Subsequently, Plevin and McLellan<sup>33</sup> found no difference in racing outcomes when comparing fore- and hindlimb branch desmitis. The results of the present study may be applicable to hindlimbs, though hindlimb-specific ultrasonography is needed to investigate this hypothesis. It may be that negative associations with performance are less detrimental, as was the case for hindlimb proximal sesamoid bone changes compared to forelimbs<sup>48</sup>. When extrapolating the ultrasonographic appearance to hindlimbs, there may be hindlimb-specific features of the suspensory branches that need to be considered, such as potential size differences compared to forelimbs and less of a medial branch predominance for abnormalities. An increased propensity for periligamentar fibrosis in hindlimb branch injuries has been noted in horses used for non-racing disciplines<sup>31</sup>.

Clinical applications of the current methodology that clinicians may find useful include the importance of saving cine loops when subsequent image evaluation is planned and using the same observer for serial measurements. It was important to recognise the normal ultrasonographic appearance of a fibrous septum separating the axial and abaxial lobes, which is prominent in some branches and should not be incorrectly interpreted as a lesion<sup>57</sup>. A predominance of palmar margin changes, not seen with the transducer positioned perpendicular to the medial or lateral aspect of the fetlock, underscored the importance of examining the palmar or plantar aspect of the suspensory branches in Thoroughbreds<sup>28</sup>. Moving the transducer from dorsal to palmar when evaluating the branch insertion in longitudinal section was also important to ensure changes were not missed<sup>31</sup>. Although not practical in the current study, the addition of non-weightbearing ultrasonographic examination and power Doppler can be useful adjuncts in the diagnosis of certain branch lesions<sup>57,58</sup>.

The sample population in this study was specifically young Thoroughbreds that had undergone sales preparation and were entered for a yearling or 2-year-old sale. Clinical exams were not performed by the research ultrasonographers, thus the definition of subclinical referred to horses deemed fit for presentation at sale by consignors and their veterinarians. This was intentional so as to produce usable information directly applicable to sales horses. The nature of the study design meant that any treatment interventions directed at suspensory branches were unknown, such as extracorporeal shockwave therapy or periligamentous injection<sup>59</sup>. Other management variables included differing training track surfaces including synthetic, dirt and turf; with three 2-year-old sales breezing on a wax-coated synthetic fibre and sand blend and two sales breezing on dirt tracks. Foot conformation and shoeing technique, which may affect suspensory branch loading<sup>51</sup>, were not assessed. However, the number of horses enrolled encompassed a large cross-section of consignors and trainers. Previous studies of subclinical ultrasonographic suspensory branch abnormalities in Thoroughbreds have been limited to 50, 60 and 62 horses<sup>28,29,34</sup>. The inclusion of 593 yearlings and 367 two-year-olds has provided a large dataset of information, with a wide sample of management and training regimes specific to sales-prepared horses. Although only Thoroughbreds sold in North America were studied, racing performance in overseas jurisdictions was included where available; including for horses exported to the United Kingdom, Ireland, France, Asia, South America and Australasia. The analysis of racing performance in this study intentionally did not separate performance outcomes on a per year basis. Previous work has found some evidence of reduced performance at 2 years of age for mild findings, which was no longer evident by the 3-year-old racing season<sup>33</sup>.

## **5.6 Conclusions**

Reference values have been established for the ultrasonographic appearance of the suspensory branches in sales Thoroughbreds. Mild changes in the suspensory branches should not be overinterpreted in presale examinations. One third of sales 2-year-olds had at least one suspensory branch with Grade 1 fibrillar pattern and one quarter of sales 2-year-olds had at least one branch with Grade 1 hyperechoic foci.

There was no evidence of reduced performance associated with Grade 1 or Grade 2 branch fibrillar pattern. However, approximately one third of yearling Grade 2 branches progressed to a Grade 3 lesion. Grade 3 branch lesions had a clinically important reduction in the likelihood of racing and the class of racing performance achieved. 2-year-olds with enlarged branch width had a delayed start to racing and reduced earnings per start. Hyperechoic foci that caused acoustic shadowing were associated with reduced total earnings and earnings per start.

CHAPTER 6 – CONCURRENT RADIOLOGICAL AND ULTRASONOGRAPHICAL FINDINGS IN THE FORELIMB PROXIMAL SESAMOID BONES AND ADJACENT SUSPENSORY LIGAMENT BRANCHES IN YEARLING AND 2-YEAR-OLD THOROUGHBRED SALES HORSES<sup>e</sup>

### 6.1 Summary

Changes in the proximal sesamoid bones (sesamoids) and the insertional region of the adjacent suspensory ligament branch (branch) are of particular importance in young Thoroughbreds sold at public auction. Little is known about the prevalence of concurrent ultrasonographic branch change relative to the various grades of radiological sesamoid appearance.

This was a prospective cohort study using an enrolled sample. The objectives were to 1) examine the existence of concurrent radiological and ultrasonographic findings in individual sesamoid-branch units in sales horses, 2) determine whether there are any radiological findings that are consistently accompanied by a particular degree of insertional branch change and 3) provide practical recommendations as to when suspensory branch ultrasonography may be warranted in a sales environment.

Horses were enrolled with consignor permission from a large Thoroughbred yearling sale and five 2-year-old Thoroughbred sales the following year. Data from the radiological evaluation of forelimb sesamoids and ultrasonographic examination of the adjacent forelimb suspensory branches was analysed using descriptive statistics.

A total of 2,204 yearling forelimb sesamoid-branch units and 1,336 2-year-old forelimb sesamoid-branch units were included from 551 sales yearlings and 334 sales 2-year-olds. The proportion of yearling sesamoids with Grade  $\leq 1$  vascular channels that had adjacent Grade  $\geq 2$  fibrillar branch change was 1.2%. The same proportion for 2-year-olds was 3.8%, with medial forelimb sesamoids with Grade 1 vascular

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<sup>e</sup> Peat, F. J., Kawcak, C. E., McIlwraith, C. W., Berk, J. T. & Keenan, D. P. (2024). Concurrent radiological and ultrasonographical findings in the forelimb proximal sesamoid bones and adjacent suspensory ligament branches in yearling and 2-year-old Thoroughbred sales horses. *Equine Veterinary Journal*, (In Press). *Included with permission of journal Editor-In-Chief.*

channels overrepresented in 2-year-olds. In yearlings, 31% of sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change and 59% of sesamoids with Grade 3 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change. In 2-year-olds, 47% of sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  fibrillar branch change and 67% of sesamoids with Grade 3 vascular channels had the same. Only 1 yearling and 1 2-year-old sesamoid with radiological abaxial concavity had Grade  $\geq 2$  fibrillar branch change.

The main limitation of this study is that hindlimbs were not included. Clinical examinations were not performed and the status of any past or present inflammatory process at the sesamoid-branch entheses could not be inferred from radiographs and ultrasonographic images alone.

The existence and prevalence of concurrent radiological and ultrasonographic findings in the proximal sesamoid bones and adjacent suspensory ligament branches has been established in yearling and 2-year-old Thoroughbred sales horses. General recommendations have been made for selective branch ultrasonography on the basis of sesamoid radiological appearance. The results support a separate aetiology for radiological sesamoid abaxial concavity that does not primarily involve the suspensory branch insertion.

## **6.2 Introduction**

Sesamoid bones are present in tendons and ligaments in areas that experience both tensile strain and compressive mechanical stresses<sup>60</sup>. The proximal sesamoid bones (sesamoids) in the forelimb are critical anatomical structures in the horse, where they are positioned within the suspensory apparatus as it changes direction coursing over the palmar aspect of the metacarpophalangeal joint (MCPJ). The medial and lateral branches of the suspensory ligament (branches) insert abaxially onto the respective medial and lateral sesamoids<sup>16,17</sup>. Vascular supply to the sesamoids originates from branches of the medial and lateral palmar digital arteries that enter the proximal half of the sesamoids through the palmar abaxial bone surface<sup>20</sup>. The sesamoids reduce friction on the suspensory ligament and place it further away from the axis of the joint, thereby increasing the mechanical advantage of the suspensory apparatus in stabilising the MCPJ

under hyperextension and high loads during locomotion<sup>15,17,61,62</sup>. Injuries to the suspensory apparatus may impair athletic ability and can have significant consequences in the horse<sup>14,15</sup>.

Given their anatomical relationship and clinical importance, a link between pathological change in the sesamoids and suspensory branches has been discussed for over 50 years<sup>18-21</sup>. This is an issue of particular importance in young Thoroughbreds sold at public auction, where presale radiographs are lodged in a repository system and abnormalities identified on radiographs can impact the sale and management of affected horses<sup>12,13,22</sup>. Foundational work on the significance of sesamoid changes and subclinical and clinical branch injury was performed by McLellan, Plevin et al, following yearlings in their first year of training<sup>23,34</sup>. The ultrasonographic appearance of the suspensory branches was recently investigated at Thoroughbred sales for the first time<sup>63</sup>. Together with a study of radiological sesamoid findings<sup>48</sup>, this new information has established the prevalence and significance of sesamoid and branch changes in a large cohort of sales yearlings and 2-year-olds. The grading system used to define sesamoid changes was revised with the benefit of previous work<sup>12,13,22,23,34</sup>. The most common radiological change in sesamoids, that of a single enlarged vascular channel (Grade 1), is now considered separately from sesamoids with greater numbers of enlarged vascular channels<sup>48</sup>. It was shown that Grade 3 vascular channels, abaxial new bone and apical or abaxial fragments in forelimb sesamoids as yearlings reduced the likelihood of racing and that Grade 3 sesamoids in hindlimbs as yearlings delayed the start of racing careers<sup>48</sup>. There was no evidence of Grade 1 or Grade 2 vascular channels being detrimental to future racing performance when present in yearlings, but the appearance of the adjacent suspensory branch was not reported in this previous publication. There is also a distinct radiological finding that appears as an abaxial concavity in yearling sesamoids in the absence of vascular channel enlargement. Abaxial concavity can dramatically alter sesamoid morphology on sales radiographs, but has been shown to usually resolve by 2 years of age and had no evidence of reduced performance<sup>48</sup>. It was suggested that this has an aetiology that does not primarily involve the sesamoid-branch enthesis but the appearance of adjacent branches has not been reported<sup>48</sup>.

A study of subclinical ultrasonographic findings in Thoroughbred racehorses found the majority of suspensory branch abnormalities were in the insertional third of the branch and, more specifically, the palmar or abaxial margin of the insertion. Given the distal location of branch abnormalities, the authors recommended further work correlating such findings with the radiological appearance of the sesamoid bones<sup>28</sup>. Branch ultrasonography at sales showed that enlarged forelimb branch width in 2-year-olds was associated with a delayed start to racing, Grade 3 branch lesions had a clinically important reduction in the likelihood of racing and the calibre of performance achieved, and the presence of Grade 2 hyperechoic foci within branches was associated with significantly lower prizemoney earned<sup>63</sup>. Mild regions of hypoechogenicity (Grade 1 fibrillar pattern) have been found in the branches of 22% of sales yearlings, 30% of sales 2-year-olds and 26-28% of clinically normal racehorses in training<sup>28,63</sup>. Subclinical Grade 2 fibrillar branch change has been found in 6.7% of racehorses<sup>28</sup>. Clinical branch injury occurred in 9.5-10.0% of Thoroughbreds in their first year of training and the coexistence of concurrent changes in sesamoid-branch units increased the potential for subsequent injury in a small number of horses<sup>23,33</sup>. It has now been established that low grade ultrasonographic branch change exists in a substantial portion of young Thoroughbreds and an understanding of the concurrent sesamoid appearance in these sesamoid-branch units is needed using a large sample.

Since radiographs are viewed first in the sales environment and ultrasonography usually only follows if there is concern over radiological findings, little is known about whether ultrasonographic branch change is present in the absence of radiological changes. However, justifiable conversation occurs within the industry over the increasing use of presale imaging. A veterinary recommendation to add further diagnostic imaging to presale examinations should be founded on evidence that suggests it will add valuable information to the sales decision-making process, ultimately to inform the suitability and management of the horse for its intended purpose.

The purpose of this study was to examine the existence of concurrent findings in individual sesamoid-branch units in sales horses. The study aimed to determine whether there were any radiological sesamoid findings that were consistently accompanied by a particular degree of insertional branch change.

It was hypothesised that only sesamoids with radiological Grade  $\geq 2$  vascular channels would have ultrasonographic Grade  $\geq 2$  fibrillar change in the adjacent suspensory branch. It was also hypothesised that radiologically visible abaxial concavity in sesamoids without enlarged vascular channels would not be accompanied by branch change. The objective was to provide practical recommendations as to when branch ultrasonography is warranted in the sales environment.

## **6.3 Methods**

### *6.3.1 Data collection*

A prospective cohort study was conducted using an enrolled sample of Thoroughbred horses. The cohort comprised yearling and 2-year-old Thoroughbreds presented for sale at public auction in the USA; beginning at the 2016 Keeneland September Yearling Sale in Lexington, Kentucky and continuing at five 2017 2-year-old sales, starting with the Fasig-Tipton Gulfstream Sale in Hallandale Beach, Florida. Enrolment, radiograph and ultrasound image acquisition and image evaluation occurred as detailed in previously published work pertaining to separate radiological and ultrasonographical studies using this cohort of horses<sup>48,49,63</sup>. Measures were taken to ensure that neither granting access to sales radiographs, nor giving permission for suspensory branch ultrasonography, would impact horses' sale prospects. Consignors had the option to approve for research only one imaging modality, or neither, if desired. The present study used only horses with both sales radiographs and research ultrasonography available. For each horse, the radiological findings for the four forelimb sesamoids and the concurrent ultrasonographical findings for the corresponding four forelimb suspensory branches were analysed. The observer (FP) was blinded to radiological findings during ultrasonographic branch image evaluation<sup>63</sup>.

The radiological appearance of each sesamoid was categorised into one of six grades for vascular channel and abaxial contour appearance, using a grading scale established for this research<sup>48</sup>. Sesamoid fragments, either apical or abaxial in origin, were noted separately. Basilar sesamoid fragments were not recorded for the purposes of this study. Ultrasonographical findings in the suspensory branches were graded

for each of fibrillar pattern (0-3), the presence of hyperechoic foci (0-2) and sesamoid bone surface irregularity at the branch insertion (0-1) Measurements of branch cross-sectional area (CSA), width and periligamentar tissue thickness (PLT) were also recorded <sup>63</sup>.

### *6.3.2 Data analysis*

Data analysis was performed using the statistical software R, version 4.3.2, with packages *car*, *dplyr*, *freqtables* and *ggpubr* <sup>a, 38</sup>. Data distribution was evaluated and the data was analysed using descriptive statistics of frequency <sup>28</sup>. Proportions had 95% confidence intervals (95 % CI) calculated. Median and interquartile range (IQR) were reported for non-normally distributed data. IQR was computed using continuous sample quantile type 8 <sup>64</sup>. Size measurements were analysed separately for medial and lateral branches because of the established medial-lateral size difference <sup>63</sup>. Periligamentar tissue thickness (PLT) was reported as a proportion of branch width, as previously described <sup>31</sup>.

## **6.4 Results**

A total of 551 sales yearlings and 334 sales 2-year-olds were included. This provided 2,204 yearling sesamoid-branch units and 1,336 2-year-old sesamoid-branch units for analysis. Among this sample were 90 horses for which radiographs and ultrasonographic images were available at both yearling and 2-year-old sale age (paired radiographs and paired ultrasonographic images).

### *6.4.1 Suspensory branch fibrillar pattern*

The distribution of suspensory branch fibrillar pattern grades (ultrasonographic finding) seen concurrently in sesamoid-branch units with each level of sesamoid grade (radiological finding) is presented in Table 6.1, separated by horse age.

### *6.4.2 Suspensory branch size and periligamentar tissue thickness*

The ultrasonographical measurements for branch CSA, width and PLT as a proportion of branch width are presented in Table 6.2 for each grade of sesamoid radiological finding, separated by age and by medial and lateral branches.

#### *6.4.3 Suspensory branch hyperechoic foci*

The distribution of suspensory branch hyperechoic foci grades seen concurrently with each grade of sesamoid radiological finding in all sesamoid-branch units is presented in Table 6.3, separated by age.

TABLE 6.1. Concurrent radiological and ultrasonographical findings for individual forelimb proximal sesamoid bone-suspensory ligament branch units in Thoroughbred sales yearlings and 2-year-olds; showing suspensory branch fibrillar pattern grades seen ultrasonographically, with corresponding radiological sesamoid grades present at the same time on sales repository radiographs. Row percentages refer to the proportion of each level of ultrasonographic finding for a given sesamoid grade. Percentages are shown with 95% confidence intervals (95% CI).

	Yearlings, N (%)					Two-year-olds, N (%)				
	Grade 0 branch	Grade 1 branch	Grade 2 branch	Grade 3 branch	Total yearling units	Grade 0 branch	Grade 1 branch	Grade 2 branch	Grade 3 branch	Total 2-year- old units
Grade 0 sesamoid	1578 92.6% (91.3- 93.8%)	111 6.5% (5.4- 7.8%)	14 0.8% (0.5- 1.4%)	1 0.1% (0.0- 0.4%)	1704 100%	988 85.1% (82.9- 87.0%)	139 12.0% (10.2- 14.0%)	30 2.6% (1.8- 3.7%)	4 0.3% (0.1- 0.9%)	1161 100%
Grade 1 sesamoid	256 83.7% (79.1- 87.4%)	40 13.1% (9.7- 17.3%)	10 3.3% (1.8- 6.0%)	0 0.0%	306 100%	71 69.6% (60.0- 77.8%)	17 16.7% (10.6- 25.2%)	12 11.8% (6.8- 19.6%)	2 2.0% (0.5- 7.6%)	102 100%
Grade 2 sesamoid	26 37.1% (26.6- 49.1%)	22 31.4% (21.6- 43.3%)	18 25.7% (16.8- 37.3%)	4 5.7% (2.1- 14.4%)	70 100%	4 21.1% (7.9- 45.3%)	6 31.6% (14.6- 55.5%)	6 31.6% (14.6- 55.5%)	3 15.8% (5.0- 40.0%)	19 100%
Grade 3 sesamoid	6 35.3% (16.4- 60.3%)	1 5.9% (0.8- 33.4%)	7 41.2% (20.5- 65.5%)	3 17.6% (5.6- 43.7%)	17 100%	0 0.0%	2 33.3% (7.2- 76.3%)	1 16.7% (1.9- 67.8%)	3 50.0% (14.7- 85.3%)	6 100%
Abaxial new bone only	25 75.8% (58.2- 87.5%)	5 15.2% (6.4- 32.0%)	3 9.1% (2.9- 25.0%)	0 0.0%	33 100%	23 74.2% (55.9- 86.7%)	4 12.9% (4.8- 30.1%)	3 9.7% (3.1- 26.5%)	1 3.2% (0.4- 20.2%)	31 100%
Abaxial concavity only	59 79.7% (69.0- 87.4%)	14 18.9% (11.5- 29.5%)	1 1.4% (0.2- 9.1%)	0 0.0%	74 100%	12 70.6% (45.0- 87.6%)	4 23.5% (8.8- 49.4%)	1 5.9% (0.8- 33.4%)	0 0.0%	17 100%
Column total sesamoid- branch units	1950 88.5%	193 8.7%	53 2.4%	8 0.4%	2204 100%	1098 82.2%	172 12.9%	53 4.0%	13 1.0%	1336 100%

TABLE 6.2. Concurrent radiological and ultrasonographical findings for individual forelimb proximal sesamoid bone-suspensory ligament branch units in Thoroughbred sales yearlings and two-year-olds; showing the median suspensory branch cross-sectional area (CSA), width and periligamentar tissue thickness ratio (PLTr), as measured ultrasonographically, for each radiological sesamoid grade present at the same time on sales repository radiographs. Median values are shown with interquartile range (IQR).

	Yearlings							
	Medial Suspensory Branches				Lateral Suspensory Branches			
	Median branch CSA (cm <sup>2</sup> )	Median branch width (cm)	Median PLTr (cm)	Total sesamoid-branch units from N = 1102	Median branch CSA (cm <sup>2</sup> )	Median branch width (cm)	Median PLTr (cm)	Total sesamoid-branch units from N = 1102
Grade 0 sesamoid	1.19 (1.13-1.26)	1.34 (1.25-1.42)	7.71 (6.46-9.15)	785	1.11 (1.04-1.18)	1.22 (1.13-1.30)	7.35 (6.28-8.51)	919
Grade 1 sesamoid	1.23 (1.15-1.34)	1.35 (1.27-1.44)	7.66 (6.51-9.67)	179	1.15 (1.10-1.25)	1.24 (1.17-1.32)	7.35 (6.20-8.94)	127
Grade 2 sesamoid	1.45 (1.24-1.69)	1.39 (1.28-1.50)	7.95 (5.90-10.40)	46	1.32 (1.19-1.49)	1.35 (1.20-1.38)	6.94 (4.78-9.32)	24
Grade 3 sesamoid	1.46 (1.36-1.84)	1.44 (1.35-1.61)	6.27 (4.55-7.96)	11	1.25 (1.09-1.32)	1.24 (1.19-1.27)	7.00 (6.35-8.31)	6
Abaxial new bone only	1.21 (1.14-1.32)	1.34 (1.23-1.43)	8.68 (6.55-11.30)	19	1.13 (1.07-1.20)	1.25 (1.22-1.31)	6.48 (6.10-7.22)	14
Abaxial concavity only	1.21 (1.18-1.30)	1.37 (1.28-1.46)	8.96 (7.50-10.70)	62	1.12 (1.00-1.19)	1.27 (1.10-1.36)	6.67 (5.26-7.45)	12

	Two-year-olds							
	Medial Suspensory Branches				Lateral Suspensory Branches			
	Median branch CSA (cm <sup>2</sup> )	Median branch width (cm)	Median PLTr (cm)	Total sesamoid- branch units from N = 668	Median branch CSA (cm <sup>2</sup> )	Median branch width (cm)	Median PLTr (cm)	Total sesamoid- branch units from N = 668
Grade 0 sesamoid	1.21 (1.13-1.28)	1.38 (1.27-1.49)	8.29 (7.00-9.80)	548	1.12 (1.05-1.20)	1.27 (1.18-1.35)	7.48 (6.20-8.57)	613
Grade 1 sesamoid	1.24 (1.18-1.36)	1.38 (1.24-1.51)	8.34 (6.94-9.93)	72	1.10 (1.05-1.20)	1.22 (1.14-1.29)	7.64 (6.90-9.92)	30
Grade 2 sesamoid	1.40 (1.34-1.53)	1.44 (1.20-1.60)	9.38 (8.12-15.4)	14	1.45 (1.26-1.67)	1.38 (1.18-1.55)	7.61 (6.82-9.27)	5
Grade 3 sesamoid	1.39 (1.38-1.94)	1.46 (1.44-1.76)	7.36 (4.19-8.05)	3	1.49 (1.46-1.72)	1.34 (1.19-1.40)	7.99 (6.72-17.3)	3
Abaxial new bone only	1.27 (1.20-1.39)	1.49 (1.34-1.59)	8.18 (6.83-10.10)	16	1.12 (1.05-1.19)	1.23 (1.15-1.36)	7.19 (5.74-7.70)	15
Abaxial concavity only	1.25 (1.15-1.33)	1.42 (1.32-1.49)	9.34 (7.64-10.60)	15	0.96 (0.95-1.04)	1.06 (1.03-1.09)	7.38 (6.15-9.52)	2

TABLE 6.3. Concurrent radiological and ultrasonographical findings for individual forelimb proximal sesamoid bone-suspensory ligament branch units in Thoroughbred sales yearlings and two-year-olds; showing suspensory branch hyperechoic foci grades seen ultrasonographically, with corresponding radiological sesamoid grades present at the same time on sales repository radiographs. Row percentages refer to the proportion of each level of ultrasonographic finding for a given sesamoid grade. Percentages are shown with 95% confidence intervals (95% CI).

	Yearlings, N (%)				Two-year-olds, N (%)			
	Grade 0 hyperechoic foci	Grade 1 hyperechoic foci	Grade 2 hyperechoic foci	Total yearling sesamoid- branch units	Grade 0 hyperechoic foci	Grade 1 hyperechoic foci	Grade 2 hyperechoic foci	Total two- year-old sesamoid- branch units
Grade 0 sesamoid	1630 95.7% (94.6-96.5%)	69 4.0% (3.2-5.1%)	5 0.3% (0.1-0.7%)	1704 100%	1036 89.2% (87.3-90.9%)	100 8.6% (7.1-10.4%)	25 2.2% (1.5-3.2%)	1161 100%
Grade 1 sesamoid	279 91.2% (87.4-93.9%)	26 8.5% (5.8-12.2%)	1 0.3% (0.0-2.3%)	306 100%	85 83.3% (74.8-89.4%)	16 15.7% (9.8-24.1%)	1 1.0% (0.1-6.7%)	102 100%
Grade 2 sesamoid	47 67.1% (55.3-77.2%)	18 25.7% (16.8-37.3%)	5 7.1% (3.0-16.1%)	70 100%	9 47.4% (26.3-69.4%)	8 42.1% (22.2-65.0%)	2 10.5% (2.5-34.7%)	19 100%
Grade 3 sesamoid	8 47.1% (25.0-70.4%)	8 47.1% (25.0-70.4%)	1 5.9% (0.8-33.4%)	17 100%	2 33.3% (7.2-76.3%)	4 66.7% (23.7-92.8%)	0 0.0%	6 100%
Abaxial new bone only	28 84.8% (68.0-93.6%)	5 15.2% (6.4-32.0%)	0 0.0%	33 100%	27 87.1% (69.9-95.2%)	3 9.7% (3.1-26.5%)	1 3.2% (0.4-20.2%)	31 100%
Abaxial concavity only	67 90.5% (81.4-95.4%)	6 8.1% (3.7-17.0%)	1 1.4% (0.2-9.1%)	74 100%	14 82.4% (56.3-94.4%)	3 17.6% (5.6-43.7%)	0 0.0%	17 100%
Column total sesamoid- branch units	2059 93.4%	132 6.0%	13 0.6%	2204 100%	1173 87.8%	134 10.0%	29 2.2%	1336 (100.0 %)

TABLE 6.4. Concurrent radiological and ultrasonographical findings for individual forelimb proximal sesamoid bone-suspensory ligament branch units in Thoroughbred sales yearlings and two-year-olds; showing sesamoid bone contour grades seen ultrasonographically at the suspensory branch insertion, with corresponding radiological sesamoid grades present at the same time on sales repository radiographs. Row percentages refer to the proportion of each level of ultrasonographic finding for a given sesamoid grade. Percentages are shown with 95% confidence intervals (95% CI).

	Yearlings, N (%)			Two-year-olds, N (%)		
	Grade 0 normal bone contour	Grade 1 irregular bone contour	Total yearling sesamoid-branch units	Grade 0 normal bone contour	Grade 1 irregular bone contour	Total two-year-old sesamoid-branch units
Grade 0 sesamoid	1607 94.3% (93.1-95.3%)	97 5.7% (4.7-6.9%)	1704 100%	1026 88.4% (86.4-90.1%)	135 11.6% (9.9-13.6%)	1161 100%
Grade 1 sesamoid	267 87.3% (83.0-90.6%)	39 12.7% (9.4-17.0%)	306 100%	78 76.5% (67.2-83.7%)	24 23.5% (16.3-32.8%)	102 100%
Grade 2 sesamoid	36 51.4% (39.8-62.9%)	34 48.6% (37.1-60.2%)	70 100%	10 52.6% (30.6-73.7%)	9 47.4% (26.3-69.4%)	19 100%
Grade 3 sesamoid	6 35.3% (16.4-60.3%)	11 64.7% (39.7-83.6%)	17 100%	1 16.7% (1.9-67.8%)	5 83.3% (32.2-98.1%)	6 100%
Abaxial new bone only	25 75.8% (58.2-87.5%)	8 24.2% (12.5-41.8%)	33 100%	25 80.6% (62.7-91.2%)	6 19.4% (8.8-37.3%)	31 100%
Abaxial concavity only	68 91.9% (83.0-96.3%)	6 8.1% (3.7-17.0%)	74 100%	14 82.4% (56.3-94.4%)	3 17.6% (5.6-43.7%)	17 100%
Column total sesamoid-branch units	2009 91.2%	195 8.8%	2204 100%	1154 86.4%	182 13.6%	1336 (100.0 %)

#### *6.4.4 Sesamoid bone contour*

The distribution of ultrasonographically visible sesamoid bone contour irregularity at the suspensory branch insertion is presented in Table 6.4, for each grade of sesamoid radiological finding in all sesamoid-branch units, separated by age.

#### *6.4.5 Sesamoid fragments*

There were 5 yearling and 6 two-year-old sesamoid-branch units that had an abaxial sesamoid fragment. Nine of the 11 fragments were from medial sesamoids. No consistent pattern of change was seen in the adjacent suspensory branch, with ultrasonographic appearances ranging from within normal limits to substantial branch enlargement and Grade  $\geq 2$  fibrillar lesions. The majority of sesamoids with fragments (8 of 11) did not have bone surface irregularity reported on ultrasound. 3 of 11 branches had Grade 1 hyperechoic foci visible on ultrasound and 1 had a visible Grade 2 hyperechoic focus consistent with an avulsion fragment. Two had very low PLT relative to branch width (4.4-4.5%), indicating branch enlargement without increased periligamentar tissue thickness. Two had high PLT relative to branch width (12.8-14.5%) indicative of periligamentar fibrosis.

### **6.5 Discussion**

The existence of radiological sesamoid change and concurrent ultrasonographical change in the adjacent suspensory branch has been examined at Thoroughbred sales for the first time. The results indicate that sesamoids with substantial radiological change have a high prevalence of adjacent branch change. It was uncommon to find moderate or severe branch change accompanying sesamoids that have radiological findings that are now considered acceptable<sup>48</sup>. One important exception to this appears to be Grade 1 vascular channels in 2-year-old medial forelimb sesamoids, where a clinically relevant proportion had adjacent Grade  $\geq 2$  fibrillar branch change. Radiologically visible abaxial concavity in sesamoids had less evidence of branch change than sesamoids with any number of enlarged vascular channels, thus a different aetiology is supported. The results have identified a radiological threshold for sesamoid appearance that

could be used to inform when ultrasonographic examination of the adjacent branch is likely to be useful at sales.

Overall, the prevalence of ultrasonographic branch lesions in the absence of substantial radiological sesamoid change was very low. Of all yearling sesamoids that had Grade  $\leq 1$  vascular channels, 1 sesamoid out of 2,010 had an adjacent Grade 3 fibrillar branch lesion (0.05%). The occurrence of Grade 2 branch change adjacent to yearling Grade  $\leq 1$  vascular channels was also low; with 0.8% of Grade 0 vascular channels having an adjacent Grade 2 branch and 3% of Grade 1 vascular channels having an adjacent Grade 2 branch. No yearling Grade 1 vascular channels had an adjacent Grade 3 branch lesion. The analysis of racing performance associated with yearling sales radiological findings concluded that Grade  $\leq 1$  vascular channels are an acceptable finding at sales and the results of the present study support this conclusion for yearlings<sup>48</sup>. For 2-year-old sesamoids that had Grade  $\leq 1$  vascular channels, 6 sesamoids out of 1,263 had an adjacent Grade 3 branch (0.5%). The occurrence of Grade  $\geq 2$  branch change adjacent to 2-year-old Grade 0 vascular channels was low at 3%, including 0.3% Grade 3 branches. It has been established that many Grade 1 vascular channels seen in yearlings have resolved by 2 years of age<sup>48</sup> but when a single enlarged vascular channel persisted or developed in 2-year-olds, the occurrence of adjacent Grade  $\geq 2$  branch change was higher than in yearlings, at 13.8%. It is important to note that by location this prevalence was 17% for medial Grade 1 sesamoids in 2-year-olds and 6.7% for lateral Grade 1 sesamoids. Seventeen percent equates to 1 in 6 horses so the authors consider this a clinically important prevalence medially.

The presence of substantial concurrent branch change rose considerably for forelimb sesamoids with 2 or more enlarged vascular channels (Grade  $\geq 2$ ). One third (31%) of yearling sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  branch change. Almost two thirds (59%) of yearling sesamoids with Grade 3 vascular channels had adjacent Grade  $\geq 2$  branches, including 18% that were Grade 3 lesions. In 2-year-olds, the proportions were even greater; almost half (47%) of Grade 2 vascular channels had adjacent Grade  $\geq 2$  branch change and two thirds (67%) of Grade 3 vascular channels had the same.

A study examining sesamoid findings and the development of clinical branch injury prior to first race found that 45% of case sesamoids (13/29) had one or more enlarged vascular channels and concluded that the presence of any number of enlarged vascular channels afforded 4.6 times the odds of developing injury in the adjacent branch <sup>23</sup>. Further investigation of non-sales yearlings in early training found that the presence of 1-3 enlarged vascular channels had 5.1 times the odds of there being grade  $\geq 2$  fibrillar change in the adjacent branch <sup>34</sup>. However, not all such sesamoid-branch units developed clinical injury and it remained difficult to predict which individuals with subclinical changes would progress to clinical disease. The authors noted that “it is probable that a subset of horses exist with mild sesamoid change and mild branch change that never demonstrate signs of clinical injury” and recommended further studies using larger populations. The results of the present study help to refine the implication that any number of enlarged vascular channels are problematic and direct the focus of ultrasonography towards sesamoids with Grade  $\geq 2$  vascular channels. Twenty years ago, the foundational sales radiology work of Spike-Pierce *et al* <sup>22</sup> concluded that their “results seemed to suggest that the number of enlarged vascular channels indicates a progressive increase in pathological change and, while tolerable in some instances, it is not acceptable once a certain degree of pathological change is reached”. Following the development of a refined sesamoid grading scale and recent work on associations with racing performance, the results of the present study suggest that this threshold lies between Grade  $\leq 1$  and Grade  $\geq 2$  vascular channels in yearlings <sup>48,63</sup>. The same threshold could be applied to 2-year-olds with the exception of Grade 1 vascular channels in medial forelimb sesamoids.

The presence of hyperechoic foci that cause acoustic shadowing within a branch (Grade 2 hyperechoic foci) has been associated with reduced future racing performance <sup>63</sup>. The proportion of sesamoids with Grade  $\leq 1$  vascular channels that had adjacent Grade 2 hyperechoic foci was less than 1% for yearlings and 2% 2-year-olds. Similar to the results for fibrillar branch change, this proportion rose considerably when two or more enlarged vascular channels were present; to 7% for yearling Grade  $\geq 2$  vascular channels and 8% for 2-year-old Grade  $\geq 2$  vascular channels. Although the overall prevalence of

hyperechoic foci was much lower than the prevalence of fibrillar branch change, rates of concurrence of this finding also appear to lie either side of a threshold between Grade  $\leq 1$  and Grade  $\geq 2$  vascular channels.

Ultrasonographic evidence of sesamoid bone surface irregularity at the branch insertion had a stepwise progression between each grade of vascular channel appearance. This was consistent across both ages. The proportions of sesamoids with an ultrasonographically irregular insertional bone surface for vascular channel grades 0,1,2 and 3 in yearlings were 6%, 13%, 49% and 65%, respectively. For 2-year-olds, the proportions were 12%, 24%, 47% and 83%, respectively. It is worth noting that the sesamoid grading scale prioritises enlarged vascular channels over abaxial contour change, so that any sesamoid with enlarged channel(s) is graded from 1-3 irrespective of radiologically evident abaxial new bone <sup>48</sup>. Even Grade 0 sesamoids (no enlarged vascular channels and no abaxial contour change on radiographs) had irregular bone surface identified on ultrasound for 6% of yearling sesamoids and 12% of 2-year-old sesamoids. This is likely because ultrasonography is more sensitive than radiography at detecting subtle bone surface changes. Interestingly, only 24% of sesamoids with abaxial new bone identified radiologically had an irregular bone surface seen at the branch insertion on ultrasound. For 2-year-olds, this figure was similar at 19%. It is important not to overinterpret ultrasonographically irregular bone at sales because only radiologically evident abaxial new bone has been shown to have a significant negative impact on racing performance <sup>48</sup>.

Fewer sesamoids with abaxial concavity identified on radiographs had concurrent change in the insertional bone surface on ultrasound than did sesamoids with any number of enlarged vascular channels. The rates of concurrent fibrillar change were also low; only 1 yearling sesamoid with abaxial concavity had Grade 2 fibrillar change and none had Grade 3. Likewise, only 1 2-year-old sesamoid with abaxial concavity had a Grade 2 branch and none were Grade 3. This supports the suggestion by Spike-Pierce and Bramlage <sup>22</sup> and Peat, Kawcak *et al* <sup>48</sup> that radiological abaxial concavity in sesamoids, in the absence of enlarged vascular channels, has a different aetiology and should not be regarded as an indicator of insertional branch desmopathy. The anatomical mid-abaxial location of the radiolucency that creates the concave abaxial contour on radiographs matches the site of radiological sesamoid change that is seen in cases of injury to

the palmar annular ligament (PAL) insertion<sup>65</sup>. The PAL, together with the intersesamoidean ligament, balances the high dorsal traction induced by the suspensory branches on the abaxial surface of the sesamoids during MCPJ extension, thereby counteracting the tendency of the sesamoids to move in a dorsal and abaxial direction under fetlock extension during weightbearing, particularly at high speed<sup>17,66</sup>. The PAL insertion was not examined ultrasonographically in this study<sup>44</sup>. However, no evidence of reduced performance associated with abaxial concavity was found in sales yearlings and the majority were no longer evident by 2 years of age<sup>48</sup>, suggesting that the radiological appearance may reflect previous PAL strain as a foal or weanling that has since resolved. Ultrasonography or magnetic resonance imaging (MRI), and ideally histological examination, of affected horses prior to yearling age would be necessary to confirm or refute this theory.

It has been proposed that radiological sesamoid change is a marker for previous injury and represents inflammatory change due to damage to the attached suspensory branch incurred as a weanling<sup>67</sup>. The findings of the present study and its two related studies<sup>48,63</sup> propose an alternative theory for Grade 1 change in sesamoids and suspensory branches. Both a singular enlarged vascular channel in yearling sesamoids (Grade 1 vascular channels) and a mild indistinct region of hypoechogenicity in 2-year-old branches (Grade 1 fibrillar pattern) were associated with increased racing performance outcomes, higher than those of their Grade 0 counterparts<sup>48,63</sup>. Furthermore, the present study has shown that 97% of yearling Grade 1 sesamoids have concurrent Grade  $\leq 1$  fibrillar change. It may be that low grade change at this age reflects adaptation to biomechanical loads that appears to confer a performance advantage. Strictly speaking, the term adaptive change infers a structural advantage and without histological evaluation this cannot be proven<sup>68</sup>.

It is a limitation of this study that clinical examinations were not performed in conjunction with imaging. Horses were considered to be free of clinical signs by nature of being deemed suitable for presentation at sale and participating in a timed gallop (breeze) for 2-year-olds. However, clinical signs associated with suspensory branch injury vary in intensity and lameness can be a transient and unreliable feature<sup>31,34</sup>. No inference can be made from the imaging findings alone about the status of any inflammatory

process that may have occurred or be occurring at the sesamoid-branch entheses. Radiological sesamoid changes are known to persist after the resolution of clinical disease<sup>69</sup> and ultrasonographic branch abnormalities can likewise persist beyond the return to clinical normality<sup>31</sup>. Further study using fused PET/CT images<sup>70,71</sup> would assist with investigating the physiologically active component at sales age and MRI may enhance understanding of concurrent sesamoid-branch findings<sup>72</sup>. Histological investigation remains the gold standard for identifying inflammatory and structural tissue change<sup>73</sup>. An additional limitation of this study is the exclusion of hindlimbs as it became impractical to scan hindlimbs in large numbers during presale ultrasonography<sup>63</sup>.

The results of this study are clinically relevant as practical recommendations can be given for ultrasonography at Thoroughbred sales, when combined with recently published analyses of racing performance that provide perspective as to the significance of the findings<sup>48,63</sup>. The results are presented objectively so that veterinarians can provide tailored advice to clients based on individual risk tolerance and intended use. The information will aid veterinarians' and clients' understanding of the relative merits of adding ultrasonographic examination to prepurchase decision making for each grade of repository radiological findings. In some instances, a horse's expected sale value or an individual buyer's level of risk tolerance may warrant the collection of further imaging information irrespective of radiological findings. However, from the results of this study, a suggested approach that would generally be acceptable for all but the most risk averse would be to ultrasound the adjacent branch for any forelimb sesamoids with Grade 2 or Grade 3 vascular channels or abaxial new bone only in yearlings. In 2-year-olds, the same is recommended, plus include any medial forelimb sesamoids with Grade 1 vascular channels. This equates to ultrasonography of 6% of all yearling forelimb suspensory branches at sales and 9.8% of all 2-year-old forelimb suspensory branches, based on published prevalence<sup>63</sup>. The incidence of clinical branch injury in the first year of race training has been reported at 9.5-10%<sup>23,33</sup>. Therefore, this suggested approach may enable detection of the horses that are most at risk of developing branch injury post-sale. Importantly, 94% of all yearling forelimb sesamoids at sales and 90% of all 2-year-old forelimb sesamoids should not require adjacent suspensory branch ultrasonography on the basis of these results.

## **6.6 Conclusions**

The existence of concurrent findings in the proximal sesamoid bones and adjacent suspensory ligament branches in yearling and 2-year-old Thoroughbred sales horses has been established. The minority of sesamoids with substantial radiological change have a high prevalence of adjacent branch change. Recommendations have been made for selective branch ultrasonography on the basis of sesamoid radiological appearance. Based on a generally accepted level of risk tolerance, 94% of all yearling and 90% of all 2-year-old forelimb sesamoids should not require branch ultrasonography, as it would be unlikely to yield information that changes either the sale value, the racing prognosis, or the clinical management of the horse. Branch ultrasonography is recommended when a specific 10% of 2-year-old forelimb sesamoids are identified at sale. Individual circumstances in certain cases may warrant ultrasonography outside of these recommendations.

## CHAPTER 7 – CONCLUSIONS

The research contained in the preceding chapters addresses current issues in the presale diagnostic imaging of Thoroughbred horses worldwide. The new scientific evidence presented here was produced by the largest imaging study of sales horses to-date and enables an understanding of radiological and ultrasonographical changes in the medial femoral condyles (MFCs), proximal sesamoid bones (sesamoids) and suspensory ligament branches (suspensory branches) of young Thoroughbreds that was previously not possible. Evidence-based determinations of the importance, or otherwise, of the various grades of findings seen in the MFCs, sesamoids and suspensory branches of yearlings and 2-year-olds have been provided. Many findings can be regarded as an acceptable appearance at a certain age and, for those that are associated with reduced performance, sales decisions can be made based on quantitative evaluations of risk that are in the best interests of the horse.

In enrolling 2,508 sales yearlings, the research aims of objectively evaluating radiological and ultrasonographical findings in the MFCs, sesamoids and suspensory branches of a large number of yearling and 2-year-old sales Thoroughbreds and collecting performance data to the end of the 4-year-old racing season were met. The range and prevalence of MFC, sesamoid and suspensory branch findings, the nature of their changing appearance between sales, and the significance of certain findings to future racing performance are now known.

Grade 1 subchondral lucencies (SCLs) were the most common MFC finding in yearling and 2-year-old sales horses. The majority of yearling Grade 1 MFC SCLs resolved or remained unchanged by 2-year-old sales. Approximately 20% of yearling Grade 1 MFC SCLs progressed to a Grade 2 deeper, dome-shaped lucency. It was possible for Grade 2 and 3 MFC SCLs to improve one grade in appearance between sales. Fewer sales yearlings with a Grade 3 MFC SCL raced, though in affected yearlings that did race there was no evidence of reduced performance. The small sample of horses that presented at a 2-year-old sale with a Grade 3 MFC SCL all raced successfully. Axial MFC lucencies did not affect racing performance.

A new grading scale for the evaluation of sesamoid radiological findings in the horse was developed and its application in this research has established its utility for future use by equine veterinarians. The new system assigns a numerical grade for vascular channel appearance that matches the number of enlarged vascular channels evident in a given sesamoid. Abaxial contour changes, when present in sesamoids that have no enlarged vascular channels (Grade 0), are noted separately as either abaxial new bone or abaxial concavity. Fragments are also noted and interpreted separately. It is recommended that use of the term “sesamoiditis” be discontinued in the presale radiology of clinically normal horses and replaced with a vascular channel grade for each sesamoid and an abaxial contour descriptor if needed.

Nearly 20% of yearling sesamoids were found to have one or more enlarged vascular channels (Grades 1-3). No association with reduced performance was found for Grade 1 or Grade 2 vascular channels. Yearlings were significantly less likely to progress to racing if they had Grade 3 vascular channels in forelimb sesamoids, abaxial new bone on forelimb sesamoids, or apical or abaxial fragments off forelimb sesamoids. Among horses that did race, those with forelimb Grade 3 vascular channels had fewer starts, those with hindlimb Grade 3 vascular channels had a 54-day delayed start to their racing careers, and those with abaxial new bone on forelimb sesamoids had lower total earnings and earnings per start.

Abaxial concavity was predominantly a finding of yearling medial forelimb sesamoids, usually resolving by 2-year-old sales and with no evidence of reduced performance. Ultrasonography findings support a separate aetiology for sesamoid abaxial concavity that does not primarily involve the suspensory branch insertion.

Reference values specific to young Thoroughbreds have been established for suspensory branch ultrasonography. Mild changes in the suspensory branches are common and should not be overinterpreted in presale examinations. Grade 1 fibrillar branch change should be regarded as an acceptable appearance in sales yearlings and 2-year-olds. No associations with reduced performance were found for Grade 1 or Grade 2 branch fibrillar pattern. However, approximately one third of yearling Grade 2 branches progressed to a Grade 3 branch lesion. Grade 3 branch lesions had a clinically important reduction in the likelihood of

racing and the class of racing performance achieved. Evidence of enlarged branch width and Grade 2 hyperechoic foci at 2-year-old sales constituted a risk to racing performance.

The prevalence of concurrent radiological and ultrasonographical findings in the sesamoid and the insertional region of the adjacent suspensory branch has been established. The minority of sesamoids with substantial radiological change have a high prevalence of adjacent branch change. One third of yearling sesamoids with Grade 2 vascular channels had adjacent Grade  $\geq 2$  branch change and almost two thirds of yearling sesamoids with Grade 3 vascular channels had adjacent Grade  $\geq 2$  branches. More so in 2-year-olds, almost half of Grade 2 vascular channels had adjacent Grade  $\geq 2$  branch change and two thirds of Grade 3 vascular channels had the same.

Recommendations have been made for selective branch ultrasonography on the basis of sesamoid radiological appearance. Based on a generally accepted level of risk tolerance, 94% of all yearling forelimb sesamoids and 90% of all 2-year-old forelimb sesamoids should not require branch ultrasonography, as it would be unlikely to yield information that changes either the sale value, the racing prognosis, or the clinical management of the horse. Branch ultrasonography is recommended for any forelimb sesamoids with Grade 2 or Grade 3 vascular channels or abaxial new bone only. Fewer than 3% of yearling sesamoids with Grade  $\leq 1$  vascular channels had an adjacent Grade  $\geq 2$  suspensory branch, therefore routine branch ultrasonography adjacent to Grade 1 vascular channels is not warranted in yearlings. Many Grade 1 vascular channels seen in yearlings have resolved by 2-year-old sales but when a single enlarged vascular channel persisted or developed in 2-year-olds, the occurrence of adjacent Grade  $\geq 2$  branch change was higher, especially medially. Therefore, in 2-year-olds, branch ultrasonography is also recommended for any medial forelimb sesamoids with Grade 1 vascular channels. This suggested approach may enable detection of the 10% of horses that are most at risk of developing branch injury post-sale. Individual circumstances surrounding certain sales horses may warrant ultrasonography outside of these recommendations.

The study samples were large and were representative of the population of interest at Thoroughbred sales. The main limitation of this research is that it may underestimate the prevalence of severe lesions in

non-sale horses. The study design could not address exclusions prior to sale, particularly with regard to withdrawals between yearling and 2-year-old sales due to the development of clinical disease. The results are directly applicable to presale imaging in Thoroughbreds prepared for public auction and deemed fit to be entered for sale.

The success of this work rests heavily on the widespread uptake by Thoroughbred consignors, breeders and trainers; in their willingness to enrol their horses in the study and their support of the work through word-of-mouth conversation and worldwide industry discussion. Public support and funding from the sales companies and significant donations from private individuals highlighted the need within the Thoroughbred industry to address the research questions. Now, the subsequent dissemination of the results to veterinarians and the wider Thoroughbred industry internationally is the next step in achieving an improved collective understanding of these presale imaging findings. It is critical to differentiate findings that have been shown to carry an increased likelihood of detrimental associations with racing performance or clinical injury from the range of findings that can now be considered insignificant within the normal population of sales horses. This will allow presale diagnostic imaging to function as intended, to increase confidence in decision making regarding bloodstock purchases and, above all, to benefit the animal through decisions appropriately tailored to an individual's findings.

Ultimately, it is hoped that the results of this research will prove clinically useful and will assist the veterinarian's role in the management and care of the athletic horse.

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## APPENDICES

APPENDIX 1 – SALES COMPANY LETTER TO YEARLING CONSIGNORS WITH STUDY  
PARTICIPATION CONSENT FORM



**Keeneland Association, Inc.**  
4201 Versailles Road  
Lexington, KY 40510  
P.O. Box 1690  
Lexington, KY 40588-1690  
859 254-3412 Tel.  
800 456-3412  
859 288-4347 Fax  
www.keeneland.com

September 24, 2016

Dear

As many of you have heard at the most recent CBA meeting, a group of equine veterinary researchers led by Drs. Wayne McIlwraith, Chris Kawcak, and Frances Peat, together with Dr. Jeff Berk, through Colorado State University Orthopaedic Research Center, have developed a protocol to do a radiographic and ultrasonographic study focused on the questions of sesamoiditis and its relationship to suspensory branch problems as well as a radiographic study evaluating lucencies in the medial femoral condyle of the stifle. The study is planned to begin at the Keeneland September Yearling Sale, and, subject to the consent of the owners of the horses at the time, these horses will have follow-up at the Two Year-Olds in Training Sales in 2017 and then again at the end of their three year-old racing season. The protocol for this study has been reviewed by other people experienced in such studies, namely Drs. Larry Bramlage, Debbie Spike-Pierce, Jonathan McLellan and Sarah Plevin.

After much discussion, the originally proposed protocol was modified so that all ultrasound examinations will be performed on farms prior to shipping to the sale, subject to the consignor's permission. Radiographs will be read both during and following the sale. All radiographic and ultrasonographic examinations will be done in a manner to preserve confidentiality, and the results will not be shared with consignors or buyers.

This study will be the largest of its kind, and we believe it will yield information that will be extremely useful to those involved in the sales process. The data from the study will aid veterinarians in providing their buying and selling clients with a more accurate assessment of the significance of these findings. With the cooperation of the sales companies, consignors, owners and veterinarians, this study stands to significantly benefit all involved.

We at Keeneland support this study by Colorado State University Orthopaedic Research Center. If you agree for the horses in your consignment to be included in the study, please indicate your consent on the attached form, and return the signed document where indicated.

Sincerely,

Bill Thomason  
CEO

Geoffrey Russell  
Director of Sales

Wayne McIlwraith  
BVSc, PhD, DSc, FRCVS, Diplomate ACVS, Diplomate ACVSMR

Chris Kawcak  
DVM, PhD, Diplomate ACVS, Diplomate ACVSMR

Frances Peat  
BVSc, PGCertSc

Jeffrey Berk  
VMD

U  
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The undersigned, on behalf of itself and the owner(s) of horses in its consignment, agrees for the horses in the undersigned's consignment to be a part of the Colorado State University Orthopaedic Research Center Study and hereby grants permission to view the radiographs of all yearlings in the undersigned's Keeneland 2016 September Sale consignment.

- - The undersigned, on behalf of itself and the owner(s) of horses in its consignment, agrees for the horses in the undersigned's consignment to be a part of the Colorado State University Orthopaedic Research Center Study, and hereby grants permission to Colorado State University, or its agents, to ultrasound scan all yearlings in the undersigned's consignment at the farm where the yearlings are located, prior to shipment to Keeneland for the September Sale.

Consignor: \_\_\_\_\_  
Address: \_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_

Good  
SAFE!!  
g

Please return the signed document to:

Jesse Ullery  
Keeneland Sales  
P.O. Box 1690, Lexington, KY 40588-1690  
FAX: (859) 233-2257  
Or scan and email: [jullery@keeneland.com](mailto:jullery@keeneland.com)

APPENDIX 2 – SALES COMPANY LETTER TO 2-YEAR-OLD CONSIGNORS WITH STUDY  
PARTICIPATION CONSENT FORM

Dear Two Year Old Consignors,

The Colorado State University Orthopaedic Research Center is conducting a study focused on the questions of sesamoiditis and its relationship to suspensory branch problems as well as a radiographic study evaluating lucencies in the medial femoral condyle of the stifle. Led by Drs. Wayne McIlwraith, Chris Kawcak, and Frances Peat, together with Dr. Jeff Berk, the study began with yearlings at the 2016 Keeneland September Yearling Sale subject to the consent of the owners of the horses at the time. These horses with the consent of consignors/owners, will have follow-up at the Two Year-Olds in Training Sales in 2017 and then again at the end of their three year-old racing season. The protocol for this study has been reviewed by other people experienced in such studies, namely Drs. Larry Bramlage, Debbie Spike-Pierce, Jonathan McLellan and Sarah Plevin.

The protocol for horses in the study is such that all ultrasound examinations will be performed on training centers prior to shipping to the sale, subject to the consignor's permission. Radiographs will be read following the sale. All radiographic and ultrasonographic examinations will be done in a manner to preserve confidentiality, and the results will not be shared with consignors or buyers.

This study will be the largest of its kind, and we believe it will yield information that will be extremely useful to those involved in the sales process. The data from the study will aid veterinarians in providing their buying and selling clients with a more accurate assessment of the significance of these findings. With the cooperation of the sales companies, consignors, owners and veterinarians, this study stands to significantly benefit all involved.

Fasig Tipton supports this study by Colorado State University Orthopaedic Research Center. If you agree for the selected horses in your consignment to be included in the study, please indicate your consent on the attached form, and return the signed document where indicated. A schedule is attached that lists the horses in the study that are in your consignment.

Sincerely,

Boyd Browning, Jr.  
President

Bayne Welker, Jr.  
Vice President of Sales

Wayne McIlwraith  
BVSc, PhD, DSc, FRCVS, Diplomate ACVS, Diplomate ACVSMR

Chris Kawcak  
DVM, PhD, Diplomate ACVS, Diplomate ACVSMR

Frances Peat  
BVSc, PGCertSc

Jeffrey Berk  
VMD

— — The undersigned, on behalf of itself and the owner(s) of horses in its consignment, agrees for the horses in the undersigned's consignment to be a part of the Colorado State University Orthopaedic Research Center Study and hereby grants permission to view the radiographs of the selected two year olds in the undersigned's 2017 Fasig Tipton Selected Two Old Sale at Gulfstream Park.

— — The undersigned, on behalf of itself and the owner(s) of horses in its consignment, agrees for the horses in the undersigned's consignment to be a part of the Colorado State University Orthopaedic Research Center Study, and hereby grants permission to Colorado State University, or its agents, to ultrasound scan two year olds in the undersigned's consignment at the training center where the selected two year olds are located, prior to shipment to Gulfstream Park for the Fasig Tipton two year old sale.

Consignor: — —  
Address: — —  
— —  
— —

Please return the signed document to:

Terri Newcombe  
Fasig Tipton Co.  
2400 Newtown Pike, Lexington, KY 40511  
FAX: (859) 254-0794  
Or scan and email: [newcombe@fasigtipton.com](mailto:newcombe@fasigtipton.com)

APPENDIX 3 - FRAMEWORK FOR THE COLLECTION OF FOLLOW-UP INFORMATION WHEN  
SPEAKING WITH CONNECTIONS OF HORSES THAT DID NOT START AT LEAST ONE RACE  
BY THE END OF THEIR 4-YEAR-OLD RACING SEASON

**1. Confirm that the horse never started a race in USA or overseas**

No race start

**2. Reason for not racing:**

Performance-related

Unrelated to performance

Different intended use  
e.g. breeding only or non-  
racing discipline

Unexpected event  
e.g. colic, paddock accident,  
systemic illness

**3. If performance-related:**

Lameness

Non-musculoskeletal  
e.g. respiratory/cardiac

Non-specific  
e.g. "just slow"/behavioural

Unknown

**4. If lameness issue:**

Fetlock

Stifle

Other

Unknown

**5. If fetlock or stifle:**

Sesamoid Bone

Suspensory Ligament

Femoral Cyst or Defect

Other

Unknown

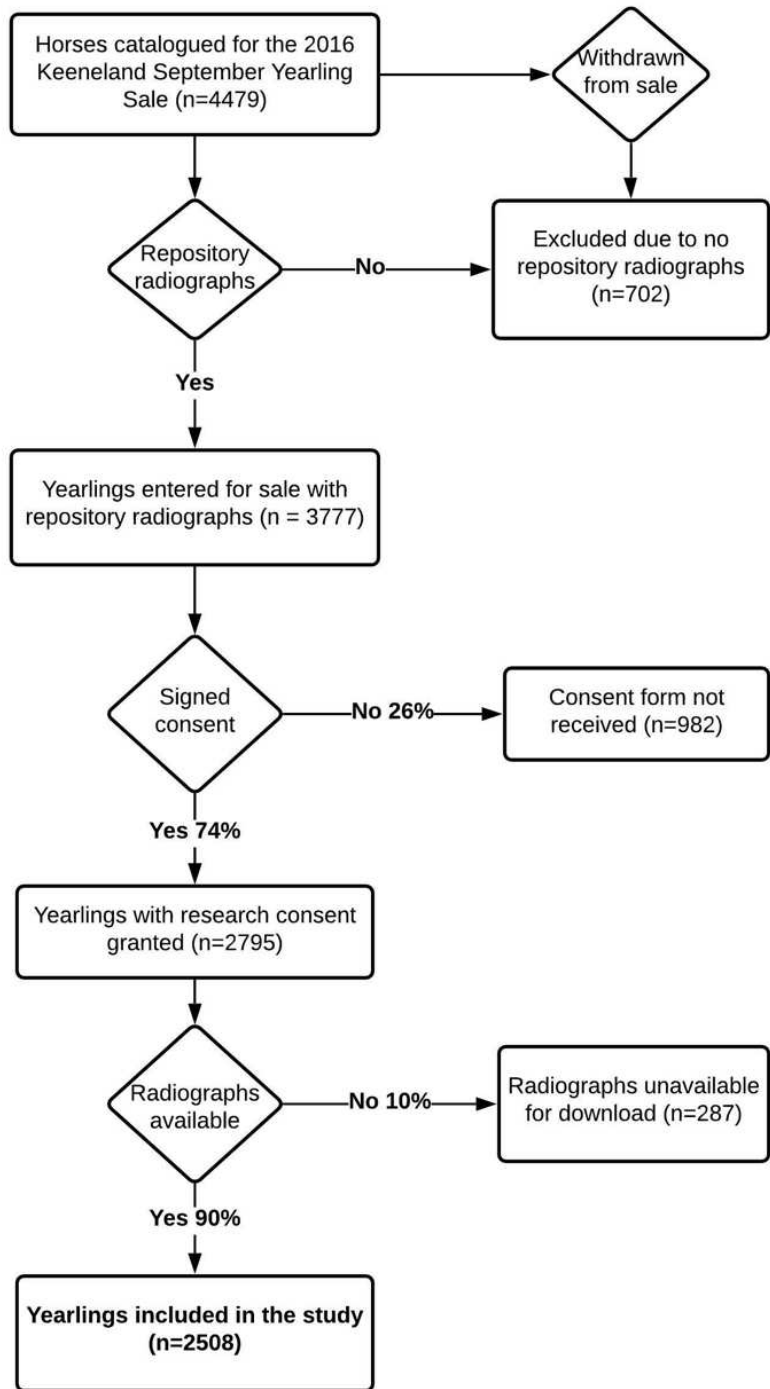
**6. Level of diagnostic accuracy:**

Veterinary diagnosis including diagnostic work-up  
and/or radiographs/ultrasound at exam

Veterinary diagnosis  
via clinical signs alone

Suspicion only/  
unconfirmed

APPENDIX 4 – FLOW CHART SUMMARISING YEARLING RECRUITMENT FOR STUDY  
 INCLUSION FROM THE 2016 KEENELAND SEPTEMBER YEARLING SALE IN LEXINGTON,  
 KENTUCKY, USA



APPENDIX 5 – FLOW CHART SUMMARISING 2-YEAR-OLD RECRUITMENT FOR STUDY  
 INCLUSION FROM FIVE MAJOR 2017 2-YEAR-OLD SALES IN FLORIDA AND MARYLAND,  
 USA

