Doctoral Thesis

Ultrasonographic approaches to identify contributing factors to changes in femoral articular cartilage morphology in collegiate rugby players

September 2024

Doctoral Program in Sport and Health Science

Graduate School of Sport and Health Science

Ritsumeikan University

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Doctoral Thesis Reviewed
by Ritsumeikan University

Ultrasonographic approaches to identify contributing factors to changes in femoral articular cartilage morphology in collegiate rugby players
（超音波検査法を用いた大学ラグビー選手における大腿骨軟骨の形態変化の要因の検討）

September 2024

2024 年 9 月

Doctoral Program in Sport and Health Science
Graduate School of Sport and Health Science
Ritsumeikan University
立命館大学大学院スポーツ健康科学研究科
スポーツ健康科学専攻博士課程後期課程

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ABSTRACT

Miyuki Hori: Ultrasonographic approaches to identify contributing factors to changes in femoral articular cartilage morphology in collegiate rugby players
(Under the direction of Tadao Isaka)

The main purpose of this dissertation was to investigate the influence of sport-specific activities and movements on the morphology of the femoral articular cartilage, serving as foundational research for establishing strategies to maintain joint health in collegiate rugby players. This study employed ultrasound imaging to examine morphological changes in the femoral articular cartilage among college rugby players.

The dissertation pursued its research objectives by addressing four specific aims. First, we examined the morphological characteristics of femoral articular cartilage among collegiate rugby players and track and field (T&F) athletes. Rugby players had greater intercondylar femoral articular cartilage thickness and larger cross-sectional area (CSA) in the lateral and middle compartments compared to T&F athletes. However, when femoral articular cartilage thickness was adjusted for body mass, no significant difference was appeared. Since body mass is associated with a high prevalence of future knee osteoarthritis (OA) and rugby players typically have a heavier body mass compared to T&F, therefore, these results suggested that body mass might affect cartilage morphology.

Participating in sports may increase the prevalence of sustained injury. Individuals with a history of joint injury have a higher incidence of OA compared to those without a history of joint injury. This present study aimed to examine the short-term longitudinal changes in ultrasonography femoral cartilage variables in an athletic population with and without a previous history of traumatic knee joint injury. The main findings were that femoral articular cartilage thickness was associated with a previous history of intracapsular knee joint injury in an

athletic population. Specifically, collegiate rugby players with a history of intracapsular knee joint injury demonstrated greater lateral condylar and intercondylar thickness, as well as CSA, compared to control players. Participation in over two consecutive seasons led to a decrease in femoral articular cartilage thickness, regardless of injury history. Additionally, there was no difference in cartilage changes based on injury history, suggesting that participation in rugby may influence morphological changes in cartilage.

Rugby involves multidirectional movements including running and cutting movements, it is still unclear which specific movements have more significant impact on cartilage thickness changes. Therefore, we compared running with 90-degree cutting movements. As a result, femoral articular cartilage thickness exhibited acute deformation after both running and cutting conditions, but no significant differences were observed between the conditions. However, 60 minutes after the cessation of running, femoral articular cartilage thickness recovered to its pre-exercise status, while the thickness did not fully recover after cuttings. This suggests that the cutting condition has a more substantial impact on femoral articular cartilage due to the slower recovery time compared to running condition.

ACKNOWLEDGMENTS

Throughout this dissertation, I have gained a wealth of knowledge and experience, an achievement that would not have been possible without the invaluable guidance and support of numerous individuals. Foremost, I extend my deepest appreciation to my supervisor, Dr. Tadao Isaka. My initial encounter with Dr. Isaka dates back to my college years, nearly two decades ago. Since then, his unwavering encouragement has been a constant source of motivation. Even when I sought advice regarding further studies, Dr. Isaka provided invaluable support, spurring me towards my goals. I have been graciously welcomed into the Isaka Laboratory and have continued to receive wholehearted encouragement to this day.

I am deeply grateful to Dr. Masafumi Terada and Dr. Tadashi Suga for their tireless dedication in assisting me throughout my study. Their meticulous comments and suggestions were instrumental in shaping the design and completion of this research.

My heartfelt appreciation also goes to Dr. Takahiro Tanaka and Dr. Yuki Kusagawa for their unwavering support and guidance. I appreciate Kento Shimoho for his cooperation in creating the cartilage analysis program. Additionally, I am indebted to Etsuko Okumura for her warm encouragement, without which my progress would have been hindered.

I extend sincere thanks to the coaches, athletic trainers, and athletes of the rugby and track and field teams for their invaluable support throughout this study. Their cooperation was indispensable in advancing this dissertation.

Lastly, I wish to express my gratitude to the members of the Isaka Laboratory for their invaluable advice and support. Without their assistance, this study would not have been possible.

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CHAPTER Ⅰ

1-1. Introduction

Participation in regular exercise is known to contribute to extending a healthy lifespan, but it also plays an important role in maintaining and improving joint health. Femoral articular cartilage thickness reflects the load placed on cartilage (1); moreover, previous research has established that the cartilage of individuals who engage in regular physical activity tends to be thicker than that of those who lead a sedentary lifestyle (2). While engaging in regular exercise is recommended for improving joint health, repetitive excessive loading and significant mechanical stress on the joints due to sports activities and joint injuries (3) (4) (5) increase the risk of developing osteoarthritis (OA). OA is not only prevalent in Japan but also globally recognized as one of the most common joint diseases. In Japan, it is estimated that there are approximately 10 million patients with symptomatic knee OA (6), while around 25 million individuals are estimated to have potential OA based on imaging diagnoses such as X-rays (7). The healthcare costs and economic burden associated with OA are substantial (8). Exercise-related pain and chronic pain caused by OA can decrease quality of life (QOL) and impair daily activities. According to Neogi (9), this pain can promote physical inactivity and overweight, leading to the development of comorbidities such as cardiovascular disease and metabolic abnormalities (9) (10) (11). Previous studies have reported participation in certain sports may increase the risk of OA (3) (4) (5). In particular, sports with high intensity of joint impact and torsional loading, such as rugby, have a high prevalence of knee OA (12). Preventing OA in sports with a high risk of OA development is crucial. However, the reasons why such sports elevate the risk of OA remain unclear. Therefore, it is imperative to elucidate the mechanisms underlying OA development in these sports.

Previous studies examining the relationship between sports and OA have predominantly focused on assessing the incidence of OA in elderly athletes (3) (4) (13) (14) (15) with fewer studies investigating changes in joint cartilage among active athletes (16) (17).There are very few studies that have examined the changes in joint cartilage resulting from participation in sports (18). Given the suggestion that participation in competitive sports may lead to the onset of OA in the future, examining the changes in joint cartilage in athletes is crucial for elucidating the mechanisms of OA development and establishing a foundational understanding in this field.

Participation in specific sports, particularly rugby, is considered a contributing factor to the future onset of OA (19). However, research aimed at understanding the actual situation of OA in athletes has not yet provided a clear picture due to several issues. These include focusing primarily on specific high-risk sports like rugby and the lack of comparative studies between these sports and others. For instance, there is a lack of research comparing morphological changes in cartilage between sports with high and low incidence rates of OA. These issues have hindered the clarification of the true situation regarding OA in athletes.

Rugby, a collision sport with a high intensity of physical contact and high-speed running, is associated with a high prevalence of knee OA (20). Rugby combines several physical demands and different types of loading conditions on the joints, such as acceleration and deceleration (21) (22), twisting (4) (19), and turning (4) motions. These types of loading conditions have a significant effect on changes in cartilage structures. Previous studies have observed the acute response of femoral articular cartilage structure changes following physical activities, including walking, running, and drop-landing (23) (24) (25) (26). Researchers reported that running caused greater acute femoral articular cartilage thickness change than drop-landing when compared to the baseline (23). Moreover, a previous study demonstrated running increased femoral articular cartilage thickness compared a walking (24) and drop-landing tasks caused greater femoral articular cartilage deformation than walking in the medial, intercondylar, and lateral compartments (26).

Previous investigations had quantified femoral articular cartilage thickness deformation only in unidirectional movements, such as walking and running. However, participating sprinting and long-distance running, which involves unidirectional repetitive movements (27), exert compressive forces on femoral articular cartilage and is associated with a lower prevalence of knee OA (28). The effects of unidirectional repetitive movements, as well as cutting, acceleration, and deceleration movements, on cartilage morphology remain unclear.

Sports-related injuries, especially joint injuries, are considered significant risk factors for developing OA. Secondary onset of OA due to anatomical and functional changes resulting from joint injuries is referred to as post-traumatic osteoarthritis (PTOA), which tend to occur at a younger age compared to typical OA (29). Injuries such as anterior cruciate ligament (ACL) tears and meniscal tears significantly impact the structure of femoral articular cartilage and are strongly associated with an increased risk of developing PTOA in the knee (30).

The progression of PTOA is influenced by various factors, including structural changes in joint tissue, molecular alterations, and neuromuscular and biomechanical changes resulting from injuries (31). Alterations in the biomechanics of the lower extremity following knee joint injuries can affect how cartilage responds to mechanical loads during both daily activities and sports (32) (33) (34) (35) (36) (37) (38). Athletes engaging in sports that involve sudden cutting movements, deceleration, and acceleration are found to have a heightened prevalence of knee OA. Therefore, assessing cartilage morphology in athletes with a history of knee joint injury holds significant importance.

1-2. Statement Problems

It can be speculated that athletes who engage in sports involving unidirectional motion, such as running, have a lower risk of developing knee OA compared to athletes participating in sports with cutting, accelerating, and decelerating movements, such as soccer and rugby. However, there is currently a lack of laboratory-based foundational research investigating which types of physical movements subject the femoral articular cartilage to high stress and which specific movements lead to structural changes in femoral cartilage.

1-3. Statement of Purpose

Our long-term goal is to promote healthy and regular physical activity throughout the lifespan by reducing the negative consequences of knee joint injury and OA and minimizing the number of disability-affected life years. The overall objective of this study was to investigate effects of rugby participation on the morphology of the femoral articular cartilage. This foundational research aims to establish innovative strategies for maintaining joint health in rugby players. Through both field- and laboratory-based studies, this dissertation systematically addresses these research objectives.

1-4. Specific Aims and Hypotheses

The four specific aims we used to achieve our objective are:

Specific Aim 1: To examine the morphological features of the femoral articular cartilage in collegiate rugby players and track and field (T&F) athletes (Chapter 3).

Hypothesis: Collegiate rugby players are anticipated to exhibit thicker femoral articular cartilage compared to T&F athletes.

Specific Aim 2: To determine disparities in femoral articular cartilage thickness among collegiate rugby players with and without a history of intracapsular knee joint injuries (Chapter 4).

Hypothesis: Collegiate rugby players with a prior history of intracapsular knee joint injury are expected to demonstrate thicker femoral articular cartilage and a larger cross-sectional area (CSA) compared to the control group

Specific Aim 3: To evaluate femoral articular cartilage thickness in collegiate rugby players with prior intracapsular knee joint injuries, who underwent supervised rehabilitation, over two consecutive seasons, compared to players without such injuries (Chapter 5).

Hypothesis: Collegiate rugby players with previous intracapsular knee joint injuries are expected to demonstrate significant changes in femoral articular cartilage thickness and CSA after two competitive rugby seasons compared to control players. Regardless of knee joint injury status, femoral articular cartilage thickness is anticipated to return to baseline levels at the beginning of the second season compared to the end of the first season. However, a decrease in femoral articular cartilage thickness is expected over the two-year competitive period."

Specific Aim 4: To compare femoral articular cartilage deformation in response to cutting and running conditions in healthy participants (Chapter 6).

Hypothesis: Femoral articular cartilage thickness experiences greater changes following the cutting condition compared to running. Additionally, the recovery time for femoral articular cartilage deformation under the cutting condition is expected to be longer than that under the running condition.

1-5. Terminology

Cartilage health - Cartilage health refers to maintaining and improving the function of articular cartilage, which distributes the load on the joints and provides smooth movement.

Extracapsular knee joint injury - Extracapsular knee joint injury refers to damage to the structures outside the knee joint capsule, such as sprains of the medial and lateral collateral ligaments.

Intracapsular knee joint injury - Intracapsular knee joint injury refers to damage to the structures within the knee joint capsule, such as sprains of the anterior and posterior cruciate ligaments, injuries to the lateral and medial meniscus, and femoral cartilage damage, resulting in swelling, pain, and temporal loss of function.

1-6. Abbreviations

ACL - Anterior cruciate ligament

ACR - American College of Rheumatology

AF - Arthritis Foundation

CI - Confidence interval

COD - Change of direction

COMP - Cartilage oligomeric matrix protein

CSA - Cross-sectional area

ECM - Extracellular matrix

GAG - Glycosaminoglycans

ICC - Intraclass correlation coefficient

KLG - Kellgren and Lawrence grades

KOOS - Knee Osteoarthritis Outcome Scores

MATLAB - Matrix Laboratory

MCL - Medial collateral ligament

MDC - Minimal detectable change

MRI - Magnetic resonance image

NSAID - Non-steroidal anti-inflammatory drug

OA - Osteoarthritis

PTOA - Post-traumatic osteoarthritis

QOL - Quality of life

ROM - Range of motion

SEM - Standard error of the measurement

SD - Standard deviation

T&F - Track and Field

US - Ultrasonography

CHAPTER II

2-1. Epidemiology of Knee Osteoarthritis

2-1-1. Prevalence of Knee Osteoarthritis

Osteoarthritis (OA) is the most prevalent joint disease worldwide, affecting 527.81 million people, with the knee being particularly susceptible (39). OA primarily affects the hips, knees, and hands; with the knee is particularly susceptible (39). In Japan, approximately 62% of individuals aged over 60 have been diagnosed with radiographic knee OA (6), and an estimated 25 million people in the Japanese population suffer from knee OA (7). The increasing number of OA patients significantly impacts society and the economy, making it crucial to effectively prevent the onset and progression of OA. However, preventive strategies to the progression of OA have not yet been established.

2-1-2. Prevalence of Knee Osteoarthritis in Athletes

OA is also a prevalent condition among athletes (4) (40). Previous studies have reported that the incidence of knee OA in athletes is higher than in non-athletes, with a particularly high prevalence in soccer players, distance runners, and weight lifters (4). A prior study indicated professional soccer players had a higher prevalence of clinical symptoms of OA (14.6%) and radiographic OA (53.7%) compared to the general population, which had rates of 12.9% and 31.9%, respectively (41).

In rugby players over the age of 50, the incidence of knee OA is as high as 60%, significantly higher than the 15% rate in the general population (42). Rugby players with a history of knee joint injury have a higher prevalence of diagnosed knee OA (20). Previous studies have shown that elite-level rugby players are more likely to develop OA than amateur-level or non-contact athletes (43) (44), with a high cumulative injury burden having continued effects even after retirement (43). Participation in certain sports has been reported to increase the chance of developing OA in the future (4), but the specific factors behind the development of OA have not been elucidated.

2-2. Financial Burden

The impact on society extends beyond direct costs to encompass indirect costs as well. Direct costs include medical expenses typically associated with hospital visits (11) (45), medication (11) (45), and rehabilitation (46). Additionally, they encompass total joint replacement surgery (11) (45) and long-term care in hospitals (46). In Japan, knee OA results in more frequent hospital visits than hip OA, with total healthcare costs being twice as high as those for hip OA (8). Medication costs significantly contribute to overall medical expenses, with OA-related surgical costs representing the highest medical expenses in Japan (8). Indirect costs encompass productivity losses from absenteeism, such as time away from work, and presenteeism, which involves disease-related decreases in productivity (11) (45). A previous study reported that approximately 71% of Japanese OA participants experienced presenteeism, while about 11% were absent from work due to health-related issues (47). Furthermore, individuals in the presenteeism group were found to more frequently use prescription medications and exhibit lower rates of health-related quality of life metrics (47).

In the early stages of OA, cartilage degeneration can progress silently, without causing pain. Consequently, by the time individuals seek medical attention, the degeneration may have advanced significantly, often necessitating medication or surgery. As morphological changes in joint cartilage are regarded as one of the indicators of impending OA, understanding the onset of OA and these changes in cartilage can provide valuable insights into preventing future OA development and alleviating the economic burden on both individuals and society.

2-3. Impact of Osteoarthritis on Physical Activity, Quality of Life, Well-being, and Comorbidity Risk

The impact of OA extends beyond direct and indirect financial costs to include intangible costs to individuals. Specifically, pain, activity limitations, and decreased quality of life are prominent factors (45). The primary clinical symptoms of OA are pain and loss of function (48). Due to pain experienced during physical activity, OA patients often become physically inactive (49) and experience reduced social participation (45). The inability to engage in physical exercise can also lead to the development of secondary conditions such as hypertension, heart disease, and diabetes (50) (51). In Japan, knee OA is closely associated with cardiovascular disease, gastrointestinal disorders, and low back pain (8). One study reported that 62% of individuals with knee OA had at least one comorbidity (50). Furthermore, older age and the severity of walking impairment and pain are associated with the onset of multiple comorbid conditions (50).

The pain associated with knee OA affects not only physical aspects but also psychological well-being. Knee OA patients are more prone to experiencing psychological symptoms such as depression (52) (53) (54) (55) and anxiety (52) compared to individuals without OA. Additionally, knee OA patients often report fatigue and sleep problems, including difficulties falling asleep, disrupted sleep, early morning awakening, and insomnia, compared to healthy individuals (53) (54). Several studies have highlighted the significant interaction between these psychological symptoms, fatigue, sleep disorders, and knee pain (52) (53) (54) (55) . Consequently, the individual burden of OA significantly impacts both physical and psychological QOL, potentially reducing healthy life expectancy. Chronic pain resulting from OA can restrict daily activities and diminish overall QOL.

2-4. Pathomechanics of Osteoarthritis

2-4-1. Mechanical Components of Cartilage Health

The Pathomechanics of OA are elucidated through the analysis of studies describing the biological, structural, and functional adaptations of articular cartilage (56). Biological adaptations of cartilage are evaluated using biomarkers (56), which are molecules released into the blood or urine during the breakdown or synthesis of cartilage and bone (57). By assessing changes in the extracellular matrix (ECM) content in synovial fluid, degeneration in joint tissues can be detected (58). Among biomarkers, cartilage oligomeric matrix protein (COMP) is frequently utilized not only in acute studies but also in longitudinal studies to assess biological changes. A previous study investigating the relationship between knee articular cartilage thickness measured by magnetic resonance image (MRI) and serum COMP concentration before and after 30 minutes of running and drop-landing reported an increase in serum COMP concentration after both exercises and a decrease in lateral tibial cartilage thickness after drop-landing (23). In a two-year longitudinal study involving Osteoarthritis Initiative patients, urinary and serum biomarker concentrations were examined alongside morphological and qualitative changes in knee joint cartilage assessed by MRI (57). This study revealed a significant correlation between serum biomarkers and lateral femoral cartilage and tibial cartilage degeneration assessed by MRI T2 mapping; however, it was noted that there was no significant correlation with morphological changes (57). While measuring changes in OA biomarker concentrations can validate the onset and progression of OA (57), it's important to note that biomarkers are released from various tissues throughout the body; hence, the values of biomarkers may not solely reflect changes in the knee joint (59). Therefore, evaluating cartilage degeneration alongside morphological changes using imaging and other methods is necessary.

Structural adaptations of cartilage involve evaluating morphological changes in cartilage using imaging techniques such as MRI and ultrasonography (US). Morphological changes in articular cartilage are crucial indicators for assessing the onset and progression of knee OA (60) (61) (62). MRI provides excellent visualization of soft tissue (63). Studies utilizing MRI have reported that patients with advanced joint narrowing in late-stage knee OA often exhibit thinning of the medial femoral cartilage (62). US is also an accurate imaging modality for assessing morphology, particularly of femoral articular cartilage (24) (63). However, US imaging is limited in its field of view for imaging femoral articular cartilage from the body surface. Nevertheless, there is a high correlation between measurements of cartilage thickness obtained from US images and those obtained from cadaver studies (63), as well as MRI (64). Articular cartilage experiences morphological changes in response to mechanical loading, as it absorbs and distributes the load across the cartilage. Thus, assessing the morphological changes in cartilage enables us to evaluate the impact of mechanical stimuli from activities on the articular cartilage.

Functional adaptations of cartilage involve assessing the load on the articular cartilage by analyzing gait patterns (56). Examining the reactive forces on the articular cartilage surface is a crucial variable for understanding the load applied to the joint (27). Clarifying the impact of diverse exercise-induced mechanical stimuli on the morphological and qualitative alterations of knee joint cartilage could promote joint cartilage health. During walking, greater peak internal knee extensor moments are applied to the knee joint, stimulating the thickening of the medial femoral cartilage (64). Additionally, during running, compressive forces are generated on the knee joint. Researchers reported that these compressive forces resulted in larger maximum medial tibiofemoral contact forces compared to sidestepping (65). Dynamic hopping activity has been shown to impose significant strain on tibial cartilage in healthy subjects (66). Furthermore, gait characteristics of chronic knee OA patients include a shorter stride length and delayed onset of mid-stance phase and mid-swing phases due to reduced range of motion (ROM) at three joints of the lower limbs (67).

2-4-2. Mechanism of General Osteoarthritis

OA is a multifactorial disease that affects articular cartilage, subchondral bone, and synovium, ultimately leading to synovial joint failure (48). Articular cartilage is composed of chondrocyte and ECM (48) (68) (69). The ECM consists of type II, VI collagen, proteoglycans, water, and other noncollagenous proteins and glycoproteins (48) (68) (69). These components help retain water within the ECM and maintain the mechanical properties of the cartilage (69). The regenerative capacity of articular cartilage is limited due to its avascular nature (68) (69).

The onset of knee OA occurs due to changes in joint loading on the articular surface caused by abnormal anatomy or by subjecting the joint to excess loads (70). These mechanical stimulation causes degeneration and wear of the cartilage and inflammation of the synovial membrane surrounding the joints (71), making the joint more susceptible to pain. The symptom of OA is spontaneous pain, which lead to difficulty in movement and restricted ROM (45). During OA progression, the entire synovial joint, including cartilage, subchondral bone, and synovium, are involved in the inflammation process (71), increasing susceptibility to pain.

2.4.3. Onset and Development of Post-traumatic Osteoarthritis

Previous joint injury is one of risk factors for developing OA (11) (48) (49) (72) (73) (74). Joint injuries alter joint structures and change the areas where joint loading occurs. As a result, the change in articular cartilage macrostructure and cartilage composition contribute to the onset of OA called post-traumatic osteoarthritis (PTOA) (75). Unlike general OA, PTOA is highly prevalent among young population (29), especially athletes (4). Joint injuries, particularly intracapsular knee joint injury such as ACL injury and meniscal tears, influence femoral cartilage structures and are associated with a high prevalence of developing knee PTOA

(30).

The onset and development of PTOA vary depending on the type and magnitude of mechanical stimuli received by the joint (31). Direct impact on knee joint structures due to trauma can alter the structure of articular cartilage and subchondral bone (19). Secondary to knee joint injury, mechanical joint instability occurs, especially due to ligament injury, which increases the load on the articular cartilage and promotes articular cartilage degeneration (19). Various treatment methods have been recommended to delay the onset and slow the development of knee OA after knee joint injury. Non-operative treatments are recommended to restore ROM (19) (30), improve lower limb muscle strength (19) (30), and enhance neuromuscular control (19) (30). Operative treatments involves reconstructing or removing damaged tissue and joint morphology (19) (30). It is also important to carry out rehabilitation and training tailored to the postoperative course to ensure success (19). A previous study indicated that patients who experienced low-intensity impacts had a minimal risk of rapid onset of PTOA, whereas those who experienced high-intensity impacts developed PTOA rapidly (76). Considering the exercise performed after knee joint trauma is also an important indicator for suppressing the onset of PTOA.

People who suffer from knee joint injury have four to six times more likely to develop PTOA than those with no history of knee joint injury (4) (75) (77) (78). According to Buckwalter (12), athletes who suffer from OA may have no clear history of the injury, or damage to the articular cartilage or subchondral bone may be overlooked even when they are examined at a hospital. It is important to understand that undiagnosed joint damage like this is also a risk for developing PTOA.

2-4-4. Risk Factor of Knee Osteoarthritis

Knee OA is a multifactorial progression disease (49) (71) (73) (75); therefore, the risk of developing knee OA varies. The main risk factors for knee OA include aging (11) (39) (48) (49) (71) (72) (73), obesity (49) (71) (72) (74), gender (48) (49) (71) (72), physical inactivity (11) (72) and abnormal joint biomechanics, such as leg length discrepancy (48) and joint laxity (72). Additionally, joint injury becomes a risk factor for developing knee OA (11) (48) (49) (72) (73) (74).

Some types of physical activities impose diverse forces on articular cartilage, and even basic exercises movements have the potential to increase the risk of future development of OA (4) (72). For example, cutting movements generate large shear forces on the knee joint (19) (79), and acceleration and deceleration produce significant shear and tensile forces on the knee joint (21). Furthermore, participation in sports involving these movements has also been suggested to be a risk factor for the onset of OA (4) (12).

2-5. Evaluation, Assessment, and Diagnosis of Osteoarthritis

2-5-1. Measurement of morphological parameters of the femoral articular cartilage

The primary complaints of knee OA are pain and loss of function of the knee joint. Traditionally, diagnosis of radiographic OA is using Kellgren and Lawrence grades (KLG) (80). The KLG is commonly used as an epidemiological study for researchers and clinical guidelines for healthcare providers (81). Evaluation of knee OA using the KLG is based on the joint space width, osteophyte formation, appearance of subchondral sclerosis, and cysts (48) (72). The formation of osteophytes and sclerosis indicates an advanced stage of cartilage wear and bone deformation. Patients are required to seek medical attention after experiencing signs and symptoms appear, people who have joint symptoms are diagnosed with radiographic OA (72). Since cartilage lacks nerve distribution, by the time pain occurs, indicating the progression of cartilage degeneration, it becomes challenging to prevent the onset of OA after the occurrence of pain is difficult.

Describing both the development and progression of OA, evaluating cartilage thickness becomes an important indicator (60) (61) (62). Recently, methods that can visualize cartilage degeneration, such as MRI and US, have more advantages over radiography to assess the progression of OA. MRI images are non-invasiveness, multiplanar capability, and excellent visualization of soft tissue (63), making MRI the gold standard for diagnosing knee OA. Buck et al. (62) conducted a one-year observational MRI study and reported examining the medial femoral cartilage thickness for each KLG. This study has shown that knees at KLG 3 showed more thinning in the medial femoral cartilage compared to thickening. Additionally, even at KLG 2, the study found that thinning and thickening of the cartilage were similar (62). Participants with a higher KLG showed associations with changes in the femoral articular cartilage thickness. Examining morphological changes in the femoral articular cartilage is useful for investigating the onset and/or progression of knee OA. MRI can also assess qualitative changes in cartilage. T2 mapping is a method used to measure the water content in cartilage, leading to the assessment of collagen content and orientation (82) and T1rho mapping can measure proteoglycan content (34). MRI allows early observation of qualitative changes in cartilage structures that may indicate early signs of OA (83) (84). However, MRI imaging is expensive and time-consuming; moreover, there are limited facilities capable of performing it (63).

US is an accurate, inexpensive, and non-invasive technique for image processing units (63). US images of the articular cartilage from the body surface, unlike MRI images, the area in which femoral articular cartilage can be evaluated using US is limited. However, it has been reported that there is good agreement between cartilage thickness measurements using US images and MRI images (64) (85). US is a varied tool for assessing femoral articular cartilage thickness (24) (63). One study measured femoral cartilage thickness using T1 weighted MRI and US image (64). This study was conducted using US images obtained through two different imaging methods, longitudinal and transverse approaches. There was a significant positive correlation with middle and posterior thickness of the femoral articular cartilage using both longitudinal and transverse approaches US images and MRI (64). Another study showed a high level of agreement between MRI and US measurements of mean femoral articular cartilage thickness in children with juvenile idiopathic arthritis (85). Also, this study reported intercondylar notch of the femoral articular cartilage was the best point for measurements of the knee. Several studies have shown that femoral cartilage thicknesses measured by US were smaller compared to MRI (64) (85). The utility of ultrasound as a clinical tool for assessing cartilage thickness.

2-5-2. Ultrasonographic Assessment of the Femoral Articular Cartilage

For the US measurement of the femoral cartilage using 2D and 3D imaging systems. There are two imaging methods for measuring femoral articular cartilage thickness with US, the longitudinal approach and the transverse approach. For the longitudinal approach, the transducer is placed sagittal plane in the middle of the medial or lateral border of the patella and the medial or lateral condyles (64). Using this method can capture the cartilage surface of the anterior, middle, and superior regions of the medial or lateral cartilage thickness can be evaluated (64). For the transverse approach, which has been introduced as an assessment of the distal femoral cartilage thickness (86), the transducer is placed transversely over the knee joint. Previous studies have described two methods: one involves aligning the transducer with the medial femoral condyle to measure the cartilage thickness at the mid-portion of the medial femoral condyle (64), and the other involves placing the knee in maximum flexion and positioning the transducer suprapatellar to broadly measure the anterior part of the femoral cartilage (87).

Previous studies have shown that the morphology of the anterior aspect of the femoral articular cartilage reflects changes in the thickness of the knee joint cartilage. Harkey et al. (25) reported that changes in the thickness of the medial femoral articular cartilage before and after walking, as measured using US, were similar to the changes in the anteromedial portion of the medial femoral condyle observed in studies using MRI. From the result of this study, overall compartmental strains at the femoral condyle were found to be significantly different in the medial and lateral compartments compared to the baseline (25). The maximum local compressive strain was observed at the anteromedial portion of the medial femoral condyle (88). It has been noted that the cartilage in the anterior aspect of the femoral articular cartilage is most subjected to load during dynamic movements such as walking. Unlike MRI images, US images capture the joint cartilage from the body surface, which limits the range that can be evaluated and makes it difficult to image the tibial cartilage, a primary load-bearing area of the knee joint. However, the measurement sites used in this study can assess the load on the knee joint due to exercise. Therefore, using US to examine femoral articular cartilage morphology can aid in understanding changes in knee joint cartilage morphology.

There are several methods for measuring cartilage thickness using US images. Most previous studies have manually measured femoral cartilage morphology. Recently, semi-automated assessments (89) (90) and automated assessments (91) are being conducted. Automated programs for the femoral articular cartilage segmentation had good reliability between manual assessment (91); however, this study reported that automated segmentation assessment was overestimated to the manual (91). Manual articular cartilage thickness measurements were measured as the linear distance (mm) drawn from the hyperechoic cartilage-bone interface to the synovial space-cartilage interface (24) (92). Previous studies have reported that manually segmented femoral articular cartilage thickness measurements exhibited good to excellence interrater reliability (25) (26) (93), and interrater reliability (26) (93). Semi-automated assessment using Matrix Laboratory (MATLAB) involves manually segmenting the total femoral articular cartilage CSA from US images, exporting these images to MATLAB for analysis, and calculating cartilage thickness and CSA (89) (90). This method demonstrated excellent intrarater- and interrater reliability with the expert raters for all femoral articular cartilage average thickness measurements (90) (94).

Validity and Reliability, Usefulness of Ultrasonographic Measurement

US has been utilized as a valid and reliable imaging tool to assess femoral articular cartilage morphology (24) (63) (89) and the number of cartilage deformations after physical activity (24) (63) (89). Researchers investigations had demonstrated agreement between a US method and cross-sectional cadaver measurements (63). This study measured cartilage thickness using US images and then anatomically measured the femoral articular cartilage through an incision and researchers reported that US measurements and actual measurements showed high agreement in the medial cartilage thickness and good agreement in the lateral thickness (63). In this study, the knee joint was flexed at 140°, and a US probe was placed transversely on the superior margin of the patella, centered on the intercondylar fossa, to image the distal femoral cartilage and measure the cartilage thickness.

Researchers have reported that the anatomic sample obtained using this US method is the trochlear groove, located between the femoral patella and femoral tibial cartilage. This area corresponds to the loading position of the femoral articular cartilage (63), where it is possible to verify changes in cartilage thickness due to load. Images obtained using this method allow for the visualization of a large area of femoral articular cartilage (63), specifically the middle area of the medial femoral articular cartilage, where changes in cartilage morphology associated with early signs of knee OA can be observed (95). Additionally, this US imaging method has been reported to have high interexaminer reliability and high intraexaminer reliability (63). Also previous researchers conducted Naredo’s method had strong intrasession reliability (24) (35), intersession reliability (24) for assessing femoral articular cartilage thickness. Therefore, this dissertation using transverse approach to verify thickness and CSA in the femoral articular cartilage.

2-6. Management of Osteoarthritis

Treatment of OA is focused on management pain and maintain or restores functional capacity becomes important role of improving the patient's QOL (8) (96). The guideline management of OA from the American College of Rheumatology (ACR) and the Arthritis Foundation (AF) 2019 recommendation that appropriate application of physical, physiological, and pharmacologic therapies by appropriate providers (97). From the guidelines from the ACR and AF, exercises are strongly recommended for knee OA, especially aerobic exercises and strengthening exercises help to reduce and improvements in OA-related pain (97). Also strongly recommendation for the pharmacologic management of knee OA is topical and/or oral nonsteroidal anti-inflammatory drugs (NSAIDs) (97). In Japan, 92% of patients of OA had prescription of NSAIDs, such as acetaminophen, is the first-line treatment for pain (96). The inter-capsular injection is the next choice for treatment pain among knee OA patients from the ACR and AF (97). In Japan, prescribed pain drugs are hyaluronic acid (35.6%), acetaminophen (21.4%), and steroid (20.0%) (96).

2-7. CHAPTER II Summary

OA is the most common type of joint disease and knee is the most prevalent joint to develop OA. The main signs and symptoms of OA are joint pain and restricted movements.

Those pain and impairments associated with OA significantly impact not only people's lives and QOL but also society.

Primary risk factors for knee OA include aging and obesity. However, OA can affect not only elderly individuals but also active individuals in younger age groups. The stimuli from regular physical activities play a crucial role in maintaining cartilage health; however, participation in physical exercises increases the onset of joint injury. Stimulation from certain exercises has been shown to significantly impact morphological changes in the femoral cartilage. Additionally, it is known that the load on the cartilage varies depending on the type of sports participation. However, it remains uncertain what amount and type of exercises are appropriate to sustain cartilage health or increase the risk of OA.

Diagnosis of radiographic OA is using KLG, which makes a diagnosis using plain radiography. Recently, the use of methods such as MRI and US to assess cartilage deformation and degeneration has enabled the evaluation of early-stage OA. US has been utilized as a valid and reliable imaging tool to assess femoral articular cartilage thickness and CSA. Participation in competitive sports poses a risk of developing OA, evaluating the morphological change of femoral articular cartilage in the athletic population can also contribute to delaying the onset of future OA.

Chapter III

The Morphological Difference in the Femoral Articular Cartilage Between Collegiate Rugby Players and Track and Field Athletes

3-1. Introduction

Regular exercise is recommended for maintaining and improving physical function. Particularly, daily exercise plays an important role in maintaining knee joint health (59). Moderate physical activity has a positive effect on cartilage thickness and volume to improve the joint health and prevent knee OA (13) (98). The femoral articular cartilage can change its shape and volume during physical activity and can recover after physical activity (24) (26), making physical exercise ideal to increase blood flow and nutritional supply to the connective tissues of the knee joint and prevent cartilage deterioration. Previous researchers reported athletes had thicker femoral articular cartilage who compared to non-athletes (2) and male physically active participants (aged from 18 to 70 years old) had greater femoral articular cartilage thickness in short-term and long-term physical activities (59). However, the evidence for effectiveness of physical activity in articular cartilage health is conflicting. Previous investigations have demonstrated that high-intense physical activity had a negative impact on the articular cartilage (21). Higher levels of physical activity accelerated of development of knee cartilage abnormalities (21) and knee joint replacement due to OA. Thus, it is crucial to engage in an appropriate level of physical exercise to preserve joint health.

Engaging in physical activities may change the morphology of cartilage to maintain the cartilage health. Cartilage morphology depends on the mechanical loading (99). A previous investigation observed that 30-minute of running and drop-landing task-induced acute morphological changes in articular cartilage at the knee joint; however, running had greater deformation of articular cartilage at the knee joint compared to drop-landing (23). Harkey et al.

(24) showed acute deformation of distal femoral articular cartilage immediately after 30-minute of walking and running conditions but observed no significant differences in the magnitude of cartilage deformation between walking and running conditions in healthy participants. Physical exercise creates different mechanical loading on the knee joint and influences to morphological change of femoral articular cartilage. However, it is unclear which types of exercise change the morphology of cartilage.

Participating sports influence to change the articular cartilage thickness. Triathletes had large areas of the femoral articular cartilage (100) and weightlifters had thicker femoral articular cartilage (101) compared with physically inactive participants. Previous researchers reported adolescent femoral cartilage was thinner than mature participants among volleyball players (102). In a previous study involving collegiate athletes from 19 different sports and a non-athlete group, the medial femoral cartilage of athletes from eight sports, including basketball, track and field, rugby, and American football, was significantly thicker compared to the non-athlete group (103). The study also reported that athletes in sports with many higher body mass players, such as basketball, American football, and rugby, have thicker medial femoral articular cartilage compared to swimmers, a sport that does not load the lower limbs during competition, although the difference was not statistically significant (103). These findings suggest that intense and frequent exercise, as well as increased knee joint load due to higher body mass, may thicken the femoral articular cartilage compared to non-exercise groups and lighter athletes. While research has been conducted on cartilage morphology between athletes and non-athletes, as well as across different age groups, there is a lack of studies examining the morphological characteristics of cartilage between different sports.

Recent literature has reported that the risk of future OA development depends on sports in which individuals participate (3). Specifically, rugby is one of the collision sports with a high prevalence of knee OA (20). Rugby requires a full contact with heavy body mass. Being overweight is a risk factor for development of OA, increase the joint loading. T&F, especially sprinters has a lower prevalence of knee OA (28). Risk and prevalence of knee OA is different of sports. Therefore, this is important to observe the morphology of femoral articular cartilage between different risks of sports.

As distal femoral articular cartilage thickness evaluated with US is one of the predictors for detecting the early onset and development of knee OA, examining the articular cartilage thickness in active athletic population provides insights into help to provide basic information for cartilage health. Therefore, the purpose of this study was to compare the anterior femoral articular cartilage between rugby that is considered as the high-risk sport of knee OA and T&F which represents as low-risk sport for knee OA. We hypothesized that the femoral articular cartilage will be thicker in collegiate rugby players when compared to T&F athletes.

3-2. Methods

Study Design

A cross-sectional study aimed to compare femoral articular cartilage morphology in college athletes and healthy individuals. All participants join US assessment during the off-season when the participants have no team practice, in late January through early February. All methodological protocols were approved by the Ethics Review Committee for Medical and Health Research Involving Human Subjects at Ritsumeikan University. A written informed consent was obtained from all participants, along with parental or guardian consent if necessary.

Participants

Fifteen collegiate T&F athletes, 11 short- and 4 middle-distance runners, from a Division I collegiate competitions and 12 rugby players, 9 forwards and 3 backs players, from a

Division I collegiate league participated in this study Our inclusion criteria for all participants was no previous history of knee joint injury within one year prior to participation. Anthropometric characteristics were taken upon arrival at the laboratory. Height and body mass were measured using scales, and body fat was measured using the impedance method (Inbody230, Inbody).

Ultrasonographic Assessment of the Femoral Articular Cartilage

A single investigator (M.H.) was responsible for taking all US assessments and analyzing the femoral cartilage. While arrive the laboratory, participants were barefoot and took a position on the treatment table in a supine position with both knees fully extended for 30 minutes to relieve pressure from the cartilage (26). Following a rest period, the examiner captured the images of the femoral articular cartilage as following directions. Participants top the head were settled at the edge of the table and with both knees positioned in the 140° flexion, using a handheld goniometer (24). A tape measure was secured to the treatment table and recorded the distance between the end of the table and the participants heels (24). A portable US unit (LOGIQe V2; General Electric Co., Fairfield, CT, USA) with a 12-MHz linear probe was used to obtain US images of the femoral articular cartilage for this study. The linear probe was placed anteriorly over the distal femoral articular cartilage of the medial and lateral femoral articular condyle in the axial position and above the superior margin of the patella. The liner probe had oriented to obtain the maximum reflection of the femoral trochlea and the overlying hyaline cartilage as a previous study reported (24) (26) (63). The location at the intercondylar notch was centered on the screen captured the image, a transparency grid was placed over the US monitor for image reproducibility (24) (25) (89) (Figure 3-1). Three images were recorded at the session and the liner probe being removes and repositioned on the knee between images. All system settings were consistent for all participants.

Figure 3-1. Participants set up and US probe positioning.
Participations were supine positioned with both knees in 140° flexion. The US probe was positioned anteriorly over the distal femoral articular cartilage of the medial and lateral femoral articular condyle in the axial position and above the superior margin of the patella.

The US examination of the distal femoral cartilage variables has been demonstrated to be valid (63) (64) and reliable (intraclass correlation coefficient (ICC) = 0.83-0.99) (24). Further, before data collection for the current study, we conducted a pilot study to establish a priori intratester reliability that included 6 participants and used the US image acquisition and processing as described in this study. We performed three US examination sessions separated by at least 1 week, and a US examiner (M.H.) established good to excellent intratester reliability ($ICC_{2,3}$ = 0.82-0.96).

Ultrasonographic Image Processing

The single investigator (M.H.) manually segmented the distal femoral cartilage of each US image (26) using OsiriX software (Pixmeo SARL, CH-1233 Bernex, Switzerland) (Figure 3-2). These segmented US images were then exported to a custom MATLAB program (version 9.10, The Math Works, Natick, MA, USA) designed to automatically calculate the mean cartilage thickness and CSA in a standardized cartilage region, following the steps outlined in a previous study (26). Initially, the program identified the central point of the US image. It then loaded the manually segmented cartilage region images from the US image and positioned the central point at 50% of the total width of the cartilage region within the overall US image. The program proceeded to divide the entire cartilage cross-sectional area into distinct regions: medial, intercondylar, and lateral. The intercondylar region was defined as the middle 25% of the cartilage region, constituting 12.5% of the length from the center to the central point. The medial and lateral regions encompassed the areas on either side of the intercondylar region, with each region's cross-sectional area being calculated (Figure 3-3). The mean femoral cartilage thickness was determined by dividing the cross-sectional area by the length of the cartilage-bone interface for each respective area (Figure 3-3) (Appendix 1). The previous study reported that US examination of the femoral articular cartilage variables has been demonstrated to be valid (63) (64) and reliable (ICC = 0.83-0.99) (24). Further, prior to data collection for the current study, the US examiner established good to excellent intratester reliability ($ICC_{1,3}$ = 0.86-0.99, standard error of measurement (SEM) = 0.06-0.11, minimal detectable change (MDC) = 0.12-0.21).

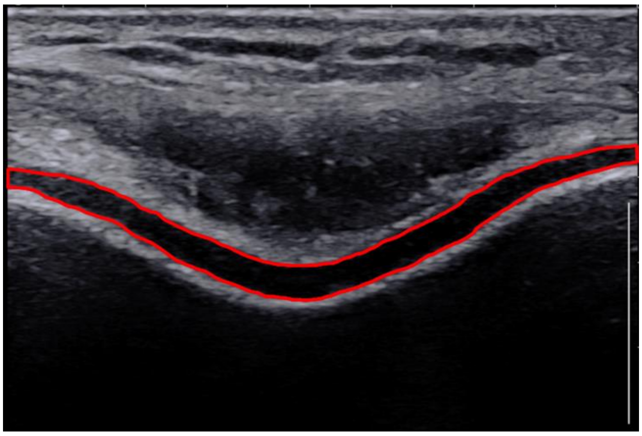

Figure 3-2. CSA of femoral cartilage

Manually segmented distal femoral cartilage CSA of each US image using OsiriX software

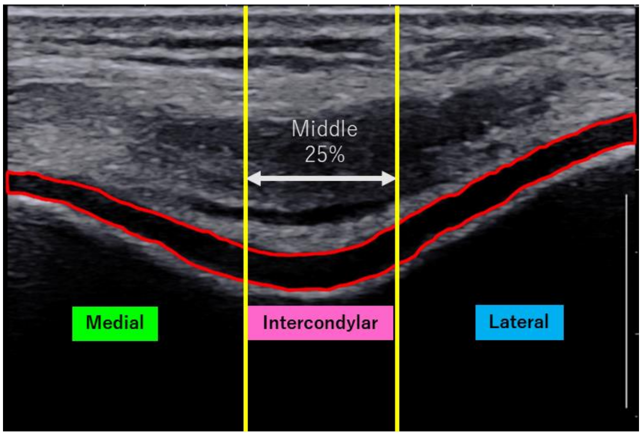

Figure 3-3. Segmentation for CSA with MATLAB program

The segmented US images were exported to a custom MATLAB program and automatically figured the mean cartilage thickness and CSA in standardized cartilage region

Statistical Analysis

Statistical analyses were performed with SPSS 27.0 (SPSS, Inc. Chicago, IL, USA). A priori alpha level was set at $p < 0.05$ for all statistical tests. We used the dominant limb, the side of kicking the ball (104) for all participants. Height, body mass, career length, and dependent valuables were compared between the T&F and rugby groups using independent t-tests. We performed between-groups comparisons of the primary outcomes using analyses of covariance, correcting for age, height, and body mass. We used Bonferroni tests for pairwise comparisons in the event of a significant analysis of covariance. Using the mean and pooled standard deviations, Cohen's *d* effect sizes and 95% confidence intervals (CIs) were calculated to assess the magnitude of differences in each femoral cartilage measurement between independent valuables for each pairwise comparison. The strength of effect sizes was interpreted as weak ($d < 0.40$), moderate ($0.40 \leq d < 0.80$), or strong ($d \geq 0.80$) (105). Equivalence was tested using the difference in mean values and 95% CIs for femoral articular cartilage morphology between rugby and T&F calculated with a paired t-test. Equivalence was assumed if the mean difference between Group A and Group B did not exceed x, and the groups were considered equivalent if the mean difference was within the CI margin.

3-3. Results

Anthropometric characteristics and the year of participating sport were provided in Table 3-1. There were significant differences in CSA of lateral femoral compartment ($p = 0.04$, $d = 0.82$, 95% CIs: 0.03, 1.61), CSA of middle compartment ($p = 0.01$, $d = 1.06$, 95% CIs: 0.25, 1.86), and intercondylar femoral articular thickness ($p = 0.01$, $d = 1.10$, 95% CIs: 0.29, 1.91) between rugby and T&F athletes. However, controlling age, height, and body mass, there were no significant differences in femoral articular cartilage morphology between rugby players and

T&F athletes. Group differences of femoral cartilage variables and rugby and T&F are presented in Table 3-2.

Table 3-1. Demographic characteristics for rugby players and track and field athletes

	Age (yrs.)	**Height (cm)**	**Body Mass (kg)**	**Body Mass Index (kg/m^2)**	**Body Fat Percentage (%)**	**Year of Participation (yrs.)**
	Mean (SD)					
Rugby (n12)	19.50 (0.91)	179.50 (8.08)	92.08 (10.28)	28.67 (3.55)	19.25 (3.91)	11.58 (3.03)
T&F (n15)	20.13 (0.64)	173.59 (5.02)	63.94 (7.92)	21.20 (2.27)	6.32 (1.96)	7.93 (2.37)
p-value	0.04*	0.03*	0.01*	0.01*	0.01*	0.01*

SD: Standard Deviation

*Significant main effects were observed ($p < 0.05$).

Table 3-2. Mean and standard deviation of the femoral articular cartilage thickness and cartilage cross-sectional area

	Compartment	Rugby	T&F	p-value	Partial η^2	Power
		Mean (SD)				
Thickness (mm)	Lateral	2.74 (0.35)	2.57 (0.20)	0.99	<0.001	0.05
	Intercondylar	2.86 (0.44)	2.48 (0.25)	0.50	0.02	0.10
	Medial	2.55 (0.49)	2.44 (0.40)	0.88	0.001	0.05
Cross-sectional area (mm^2)	Lateral	41.43 (6.02)	37.68 (2.94)	0.82	0.002	0.06
	Middle	27.31 (4.27)	23.73 (2.49)	0.50	0.02	0.10
	Medial	36.73 (7.21)	34.13 (5.72)	0.95	<0.001	0.05

SD: standard Deviation.

3-4. Discussion

This study examined morphological characteristics in the femoral articular cartilage among collegiate rugby players and T&F athletes. Collegiate rugby players had greater femoral articular cartilage thickness and CSA compared to T&F athletes. However, when controlling body mass, no differences between rugby and T&F exist in the selected-valuables. The findings of this study indicate that participation in specific sports may not be associated with distinct morphological characteristics of femoral articular cartilage. A previous study reported that values of medial femoral articular cartilage thickness in collegiate athletes were similar to those in the current study and there might not be differences in medial femoral articular cartilage thickness between rugby and T&F athletes, supported by a small effect size (means thickness rugby = 1.39mm ± 0.27, T&F = 1.39mm ± 0.29, Cohen's d effect size = 0.00, 95% CI = -0.39, 0.39) (103). They had equivalent for all femoral articular cartilage thickness (mean = -0.06mm, 95% CI = -0.75, 0.64). Our findings and the previous investigations support the result of that participating in specific sports does not alter the morphological features of articular cartilage.

Participation in competitive sports is pivotal for maintaining joint health, although certain sports may elevate the risk of future OA development (3) (4) (5). Findings from this study suggest that engagement in specific sports does not induce morphological alterations in articular cartilage. A prior investigation noted no changes in cartilage thickness among marathon runners across different competitive levels (106). Additionally, a study examining pre- and post-marathon running cartilage thickness revealed significant alterations solely in the left lateral femoral condyle cartilage thickness (107). Nonetheless, the morphological effects on cartilage from long-distance running surpass those from routine physical activities. Another study observed femoral articular cartilage thickness among younger athletes, including soccer, volleyball, weightlifter, and basketball, and sedentary individuals (108). This study reported that no significant differences in femoral articular cartilage thickness were observed between sports, except for right lateral condyle thickness in soccer players and basketball players (108). These observations suggest that participating in sports, regardless of type, level, or intensity, could potentially enhance cartilage health.

This study identified no morphological differences in femoral articular cartilage between collegiate rugby players and T&F athletes. This raised the question of whether specific sports may have a different effect on articular cartilage morphology. Both rugby and sprinting are typically categorized as the high-level of joint impact and torsional loading activities (12). Rugby involves multidirectional movements and changes of velocity, including sprinting, jumping, landing, sudden change of direction (COD), and tackling (109, 110). In rugby, sudden CODs and sudden accelerations are essential for attacking with the ball and evading opposing players or the defense line to score points (111). Additionally, defense actions require strides, sprints, and lateral movements as preparatory actions before making a tackle (112) and sudden acceleration is crucial for the tackler to collide forcefully with the opposing player (112) (113). Repeating such movements applies compressive forces and sagittal as well as transverse shear forces to the femoral articular cartilage.

T&F athletes, such as 100-meter sprinters, must perform rapid acceleration in a short period of time. At the block start, the runner extends their knee and hip joints to push off and accelerates rapidly (114), reaching top speed within 30-50 meters (115). They then maintain maximum speed and begin decelerating after crossing the finish line (115). Therefore, during a 100m sprint, compressive forces and sagittal shear forces are continuously applied to the femoral articular cartilage throughout the race. In the 200- and 400-meter sprints, athletes run on curves; however, T&F athletes are no movements involving significant lateral cutting movements and COD.

The compressive force, generated by running causes the water contained in the cartilage tissue to flow out, causing changes in cartilage thickness (66) (69) (116). The shear force, generated during cutting movements load to irregularities in the collagen arrangement within the cartilage tissue, leading to changes in cartilage thickness (66) (69) (116). The torsional force, characterized by twisting forces exerted on the joint surface, holds the potential to inflict damage on both the articular cartilage and the subchondral bone region (12). Rapid loading of articular cartilage is more likely to rupture the articular surface compared to more gradually applied loads (117). Cartilage, with its distinct layers, possesses various mechanisms for absorbing and dispersing loads contingent upon the nature of the stimuli applied (69). Participation in activities imposing diverse mechanical stimuli on the knee joint is believed to significantly influence cartilage morphology. Some researchers have previously hypothesized that the introduction of such complex stimuli may not lead to discernible changes in cartilage morphology. Consequently, the application of complex stimuli results in differential loading across various regions of the femoral articular cartilage, may potentially cause morphological changes in the knee joint.

Another possible reason that exercise loads are less likely to affect changes in the morphology of the femoral articular cartilage is that femoral cartilage has a higher compression coefficient and lower hydraulic permeability compared to tibial cartilage (118) and patella cartilage (119). Femoral articular cartilage is stiffer under compressive and shear forces compared to tibial cartilage (118); therefore, it would be predicted that femoral articular cartilage undergoes less deformation than tibial cartilage when subjected to the same magnitude of load.

Previous studies have compared the cartilage thickness of athletes with that of the non-athlete group. Some investigations have shown that femoral articular cartilage among competitive athletes is thicker compared to that of healthy individuals (2) (101) (103) (108). For instance, a study utilizing sagittal MRI images to compare the medial and lateral femoral cartilage of weightlifters with controls identified that weightlifters exhibited significantly thicker cartilage across all areas (101). Similarly, research examining the cartilage thickness of the medial femoral articular cartilage using US images revealed that athletes engaged in basketball, fencing, softball, handball, track and field, rugby, volleyball, and American football displayed thicker average cartilage thickness on both sides compared to the control group (103). A study by Babayeva et al. (108) using ultrasound images shows that the distal femoral cartilage of athletes is thicker compared to that of non-athletes. Moreover, in an MRI study comparing the knee cartilage thickness of weightlifters, bobsled sprinters, and controls, the patella cartilage was significantly thicker compared to the control group, although there was no significant difference in the thickness of the overall knee joint cartilage, including the femoral articular cartilage (2). Similarly, a study comparing the cartilage thickness of triathletes and controls found no difference in cartilage thickness, but observed that the knee joint surfaces of male athletes were wider, and the medial tibial surface of female athletes was also wider (100).

A longitudinal MRI study examining changes in knee joint cartilage during growth reported that individuals who engaged in physical activities with moderate or higher intensity in childhood had significantly larger patellar and tibial cartilage volume compared to those who engaged lower intensity physical activities (120), moreover, an individual who participated in vigorous sports had notably larger in the tibial cartilage than other individuals (120). The average number of years in participating rugby in this study has been reported 11.58 years, indicating that they started playing rugby around the age of 8. Similarly, the T&F athletes have an average of 7.93 years of experience, indicating they began participating in T&F around the age of 12. The athletes competing at a high level of competition in college have been participating in competitive sports since childhood. Therefore, the substantial mechanical load experienced during their growth period from these activities may have influenced the knee joint cartilage volume of these collegiate athletes.

Exercise is crucial for preserving joint health; however, engagement in specific sports and recreational activities may heighten the risk of knee OA (3) (4) (5). Rugby, characterized by intense impact, torsional load on the knee joint, and contact, is associated with a heightened prevalence of knee OA (12) (20). Furthermore, participation in physical activities raises the risk of knee joint injury, particularly sports-related joint injuries, which can hasten joint degeneration and increase the prevalence of knee OA (29) (75). For future research, it is imperative to investigate the cartilage morphology of rugby players and track longitudinal changes resulting from injuries to maintain the joint health of rugby players.

In addition to joint loading induced by physical activities, the cartilage morphology are influenced by other factors. Age and body mass are recognized as factors associated with cartilage thickness and are considered primary risk factors for OA. The resilience of articular cartilage is influenced by collagen density, yet aging induces cellular senescence leading to decreased collagen fiber density and degeneration (49), an inevitable aspect of aging. Considering the impact of these variables on cartilage thickness changes within the age range of the collegiate athletes studied (19-21 years old, mean ± SD: 19.85 ± 0.82), maintaining an appropriate body mass may serve as one approach to mitigate the risk of OA development. Additionally, we ran post-hoc a Pearson bivariate correlation analysis to evaluate the relationship between body mass and morphology of femoral articular cartilage. The correlation analysis indicated that body mass was significantly associated with CSA of lateral and middle compartments and intercondylar thickness of femoral articular cartilage ($r = 0.42 - 0.49$, $p = 0.05$). Previous studies have reported that the lateral femoral cartilage of overweight or obese young individuals is thinner compared to those with normal weight (121). It has also been reported that a higher rate of weight loss significantly inhibits the reduction of medial femoral cartilage thickness (122). Excess weight amplifies the mechanical stimuli resulting from exercise, consequently elevating the overall load on the articular cartilage, thus serving as a crucial indicator for preventing future OA development.

This current study has a limitation that many cross-sectional studies do not specify the timing of cartilage measurements, making it difficult to determine at which point during the athletic season the athletes' cartilage was imaged. Therefore, there are necessary to conduct a prospective study that evaluates the morphological changes of athletes' cartilage over a long time throughout the competition season and examines how participation in competition affects cartilage morphology.

We observed the cartilage morphology using US. The compositional and structural changes are linked to early cartilage degeneration, which precede visible morphological alterations. Through MRI assessment of qualitative changes in articular cartilage, it becomes possible to evaluate degeneration before morphological changes manifest. In the early stage of OA, a decrease in proteoglycan content, disruption of the collagen matrix, and increase in water content occur in the cartilage composition (123). These abnormal changes in cartilage composition were evaluated using MRI, including T2 and T1rho (18). This is a method that allows changes in cartilage structure to be observed before morphological changes in the cartilage are visualized. Future studies are needed to clarify the quality of cartilage as well as morphological changes.

3-5. Summary

Participating in sports an important role in maintaining joint health and it is known that the future incidence of OA varies depending on the sport who participate in. Therefore, this study observed the cartilage thickness between rugby players and T&F athletes.

There was no morphological different observed in the femoral articular cartilage between rugby and T&F athletes. Rugby players, high prevalence of knee OA, had greater femoral articular cartilage thickness and CSA than T&F athletes, lower prevalence of knee OA. However, femoral articular cartilage thickness was corrected for body mass, no significant difference was appeared in cartilage morphology. Rugby players had heavier body mass than T&F athletes, suggesting that body mass, which is one of the risk factors for developing OA, might affect cartilage morphology.

Chapter IV

Morphological Changes in Femoral Articular Cartilage Structure in the Competitive Athletes With or Without a Previous History of Knee Joint Injury

4-1. Introduction

Participation in regular physical activity and organized team sports has an important role in maintaining articular cartilage health and slowing the progression of lower extremity joint OA (124). However, participation in certain sports may increase the risk of OA (3) (4) (5). A high prevalence of knee OA has been reported in contact and collision sports such as soccer (3) (4) (5) and rugby (20). Given the intensity of physical contact and collision sports, the incidence of a traumatic knee joint injury is high in rugby (31) (125) (126) and may lead to long-term morbidity (20) (127), subsequently increasing the number of years living with disability. The strong association of traumatic knee intracapsular injury with symptomatic radiographic OA has been observed in rugby (20), indicating that rugby-related injury is one of the risk factors associated with the development of early onset knee OA. Therefore, understanding of the mechanisms accelerating the development of OA following traumatic knee intracapsular injury is important for effective administration of effective prevention for post-traumatic knee OA in rugby cohorts.

While the development of early onset knee OA following traumatic joint injury is a multifactorial progressive process, researchers have theorized that alterations in a cartilage response to mechanical loading may accelerate the development of knee OA. Alterations in lower extremity biomechanics resulting from knee injury may alter cartilage response to mechanical joint loads that occurs during activities of daily living and sport-related activities (34) (35) (36) (37) (38), which adversely affects tissue homeostasis and structural integrity of cartilage (73) (128) (129) (130) (131) Goldring, (59). Investigators have observed increased strain of the tibiofemoral cartilage compartment during a weight-bearing activity in individuals with medial meniscus injury (132). Contact and collision sports with high prevalence of knee OA involves a greater frequency of abrupt cutting, decelerating, and accelerating movements that exerts greater aberrant shear or torsional loading, which is theorized to contribute to reducing the fatigue life of cartilage (27). Thus, it is critical to understand how cartilage of rugby players with knee injury responds to rugby-related activities.

Quantifying the ability of the cartilage structure to appropriately respond to mechanical loading has been considered as a sensitive marker to detect the earliest alterations in cartilage structure and function (24) (26) (133). US has been utilized as a valid and reliability imaging tool to assess the femoral cartilage size and the amount of cartilage deformation after physical activity (24) (26) (63). Previous investigations have demonstrated high agreement between a US method and cross-sectional cadaver measurements (63) as well as MRI (64). Harkey et al. (134), using US, observed increased thickness of anterior femoral cartilage in individuals who have received surgical management of ACL tear compared with uninjured control individuals. Previous studies reported that acute deformation of the anterior femoral cartilage was greater after common physical activity conditions compared to non-loading conditions (24) (26). One cohort study assessed a longitudinal change in femoral cartilage thickness using MRI in physically active young adults with ACL injury and observed an increase in cartilage thickness over 5 years following ACL injury (116). However, no investigators to data have used US to assess the cartilage structure in rugby players who have previously experienced traumatic knee joint injury. Understanding associations between knee injury and the femoral cartilage response of sport-related activities will direct clinicians and researchers to determine optimal exercise-related strategies to mediate the risk of post-traumatic knee OA. Therefore, the purpose of this study was to investigate the effects of the combination of a previous history of traumatic knee joint injuries and competitive athletic activities on the anterior femoral cartilage morphology in collegiate rugby players. Based on previous investigations (116) (134), we hypothesized that collegiate rugby players with a history of traumatic knee joint injury would demonstrate a greater increase in anterior femoral articular cartilage thickness and CSA compared with control players.

4-2. Methods

Study Design

A prospective observational design was used to examine US outcome measures of anterior femoral cartilage morphology before (pre-season) and after (post-season) a 5-month competitive season (108 days of practice and 17 games) in collegiate ruby players with and without a previous history of knee joint injury (Figure 4-1). A single, unblinded investigator performed all femoral cartilage US assessments. The methods of this study were carried out in accordance with the Declaration of Helsinki. All methodological protocols were approved by the Ethical Committee on Human Research of the Ritsumeikan University Institutional Review Board.

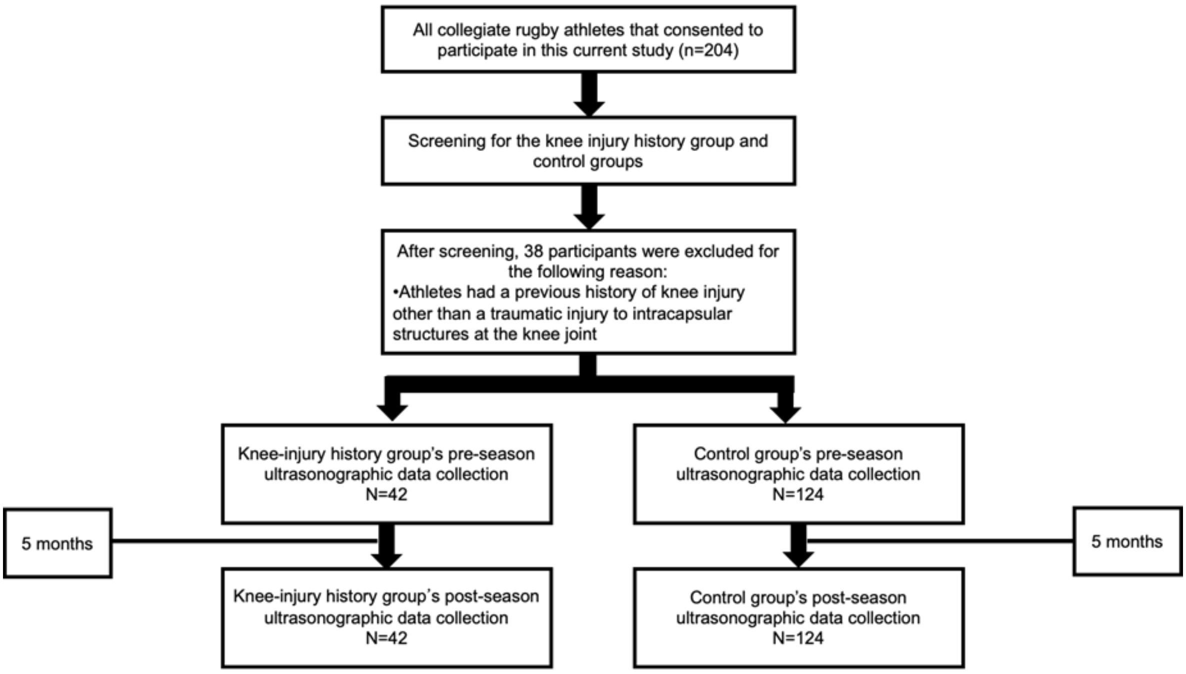

Figure 4-1. Flow chart depicting rugby players participation, data loss, and time between femoral articular cartilage ultrasonography outcome measures data collection session.

Participants

Two hundred four rugby players from a Division I collegiate team were enrolled in this study. All players were cleared for full participation by a physician. A written informed consent was obtained from all participants, along with parental or guardian consent if necessary. Enrolled players were screened with self-reported questionnaires and a medical chart recorded by an athletic trainer providing care to the rugby team to allocate the knee injury history group or control group. Rugby players in the knee injury history group were required to have a previous history of at least one significant traumatic injury to intracapsular structures (e.g., anterior and posterior cruciate ligament sprain, lateral and medial meniscus injury, femoral cartilage injury) in the knee joint resulting in swelling, pain, and temporal loss of function. No participants in the knee injury history group had acutely injured their knee in the previous 1 month of pre-season testing. As this study was focused on effects of a history of traumatic intracapsular injury at the knee joint, rugby players that have a previous history of extracapsular knee injuries (e.g., medial and lateral collateral ligament sprains) other than an intracapsular injury at the knee joint (n = 38) were not included in this current study. The type and frequency of injuries sustained by the participants are reported in Table 1. The control group participants were required to have no previous history of: 1) musculoskeletal injuries in the lower extremity; 2) back pain; and 3) surgery in the lower extremity. This resulted in a total of 166 rugby players, 42 in the knee injury history group and 124 in the control group (Table 4-1).

Additional information related to each participants' perception of their knee were collected for both groups using the Knee Osteoarthritis Outcome Scores (KOOS). The KOOS is a valid and reliable region-specific patient-generated instrument and consists of 42-items across 5 subscales that assess symptoms, pain, activities of daily living, sport and recreation, and QOL (135).

Table 4-1. List of a previous history of a traumatic knee joint injury.

Injury Type	Frequency	The Number of Surgical Management
Anterior cruciate ligament (ACL) sprain	19	13
Posterior Cruciate ligament (PCL) sprain	13	0
Medial meniscus injury (MMI)	12	10
Lateral meniscus injury (LMI)	9	7
Femoral cartilage injury	1	1

The number of participants who sustained multiple knee joint injury:

ACL + MMI = 4

ACL + LMI = 2

MMI + LMI = 4

LMI + cartilage = 1

Ultrasonographic Assessment of the Femoral Articular Cartilage

Ultrasonographic image acquisition. Participants laid supine comfortably on an examination table with the testing knee positioned in the 140° flexion, controlled using a handheld probe was used to obtain US images of the anterior femoral cartilage for this study. All system settings of the portable US unit were consistent between pre-season and post-season testing sessions as well as participants. The liner probe was placed anteriorly over the distal femoral cartilage of the medial and lateral femoral condyles in the axial position and above the superior margin of the patella and oriented to obtain the maximum reflection of the femoral trochlea and the overlying hyaline cartilage as a previous study reported (24) (26) (63) (136).

The location at which the intercondylar notch was centered on the screen was marked on the skin, enabling the examiner to return the probe to the exact location for all subsequent scans. Three images were recorded at each testing session (pre-and post-season), with the linear probe being removed and repositioned on the knee between recorded images. The US examination of the distal femoral cartilage variables has been demonstrated to be valid (63) (134) and reliable (ICC = 0.83-0.99) (26). Further, prior to data collection for the current study, we conducted a pilot study to establish a priori intratester reliability that included 6 participants and used the US image acquisition and processing as described in this study. We performed three US examination sessions separated by at least 1 week, and an US examiner (M.H.) established good to excellent intratester reliability ($ICC_{2,3}$ = 0.82-0.96).

Ultrasonographic Image processing

A single investigator (M.H.) assessed distal femoral cartilage thickness, partial area, and echo intensity using ImageJ software (National Institutes of Health, Bethesda, MD, USA). Anterior distal femoral cartilage thickness was measured at the intercondylar notch and 1 cm apart in the medial and lateral directions that were used as an estimate of the medial and lateral condyle cartilage thickness (137). The straight-line distance (mm) drawn from the hyperechoic cartilage-bone interface to the synovial space–cartilage interface was used to measure femoral cartilage thickness (Figure 4-2A) (24) (26). CSA (mm^2) of the femoral cartilage was assessed by manually tracing an area of femoral cartilage between a lateral and a medial measurement point where femoral cartilage thickness was measured (Figure 4-2B). Echo intensity was determined by the average gray-scale value across all pixels in the selected area on scale from 0 to 255 (138) (139).

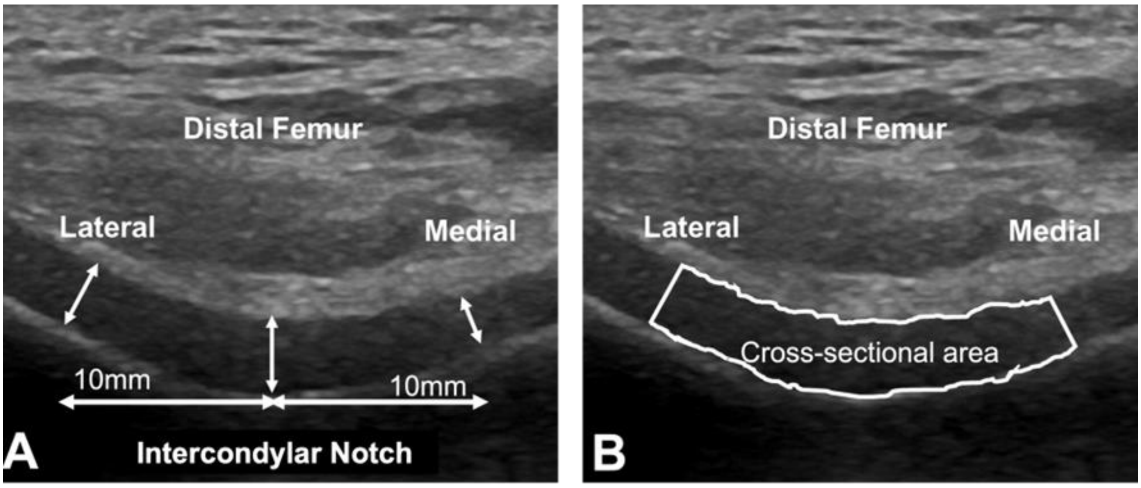

Figure 4-2. Ultrasonographic image of the anterior femoral articular cartilage:
(A) Articular cartilage thickness, (B) Articular cartilage CSA.

Statistical Analysis

An a priori alpha level was set at $p < 0.05$ using SPSS 26.0 (SPSS, Inc. Chicago, IL.) for all statistical tests. Anthropometric variables and the KOOS scores were compared between the knee joint history and control groups using independent t-test. Separate 2 × 2 (group × time) repeated measure ANOVAs were used to analyze each US femoral cartilage variable. In the case of statistically significant interactions, a post-hoc univariate analysis with pairwise comparison was conducted to ascertain the location of significant differences. For the knee joint injury history group, the previously injured limb was used. For the control group, no side-to-side differences existed for any of the US femoral cartilage measures ($p > 0.05$); therefore, the mean of both sides from the players without a history of knee joint were used. To assess the magnitude of differences in each US femoral cartilage measure between independent variables, Hedges' g using the pooled standard deviations were calculated, along with 95% confidence intervals for each pairwise comparison. The strength of the effect sizes was interpreted as small ($0 \leq g < 0.40$), moderate ($0.40 \leq g < 0.70$), large ($g \geq 0.70$) with 95% Cis (105).

4-3. Results

All included participants completed the follow-up, post-season assessment. Anthropometric characteristics were not different between the knee joint injury history and control groups ($p > 0.05$, Table 4-1). Participants with a history of knee joint injury scored significantly lower on all subscales of KOOS compared to controls ($p < 0.05$, Table 4-2).

Table 4-2. Demographic characteristics for the traumatic knee joint injury history and control groups, mean (SD).

	Intracapsular Knee Joint Injury History	**Control**	**p-value**
n	42 males	124 males	-
Age (year)	19.98 (1.22)	19.76 (1.15)	0.30
Height (cm)	173.26 (6.04)	174.40 (6.28)	0.31
Body Mass (kg)	84.64 (12.42)	84.12 (11.91)	0.81
Body Mass Index (kg/m^2)	28.15 (3.65)	27.60 (3.22)	0.36
Body Fat Percentage (%)	23.75 (17.17)	21.24 (4.37)	0.14
# of Previous Knee Injuries	1.67 (0.82) Min=1, Max=4	0.00	-
Time since the most recent knee joint injury (month)	19.94 (16.09)	-	-
Knee Injury and Osteoarthritis Outcome Score			
Pain	96.15 (5.15)	98.44 (4.33)	p=0.03*
Symptoms/Stiffness	89.18 (11.98)	95.60 (7.84)	p<0.01*
Function, Activities Daily Living	99.16 (2.39)	99.84 (0.66)	p=0.08
Function, Sports and Recreational Activities	89.40 (18.19)	98.64 (4.51)	p<0.01*
Quality of life	90.03 (16.68)	97.94 (8.25)	p<0.01*

SD: Standard Deviation

*The knee injury history group exhibited less scores on the Knee Injury and Osteoarthritis Outcome Score compared with the control groups ($p < 0.05$).

Means and standard deviations of US anterior femoral cartilage variables at pre-and post-season measurements can be found in Table 4-3. There were significant group main effects for lateral condylar thickness ($F1,164 = 4.62$; $p = 0.03$; $\beta = 0.57$), intercondylar thickness ($F1,164 = 5.14$; $p = 0.03$; $\beta = 0.62$), and partial area ($F1,164 = 5.59$; $p = 0.02$; $\beta = 0.65$). Rugby players with a history of traumatic knee joint injury had greater lateral condylar (2.37 ± 0.35 mm) and intercondylar thickness (2.51 ± 0.47 mm), as well as greater partial area (44.67 ± 7.28mm^2) compared to control players (lateral = 2.23 ± 0.35 mm, intercondylar = 2.32 ± 0.47 mm, partial area = 41.60 ± 7.26 mm2), regardless of pre-and post-season assessment time points. The magnitude of between-group differences was moderate for lateral condylar thickness ($g = 0.40$; 95% confidence intervals (CIs) = 0.05, 0.75), intercondylar thickness ($g = 0.40$; 95% CIs = 0.05, 0.75), and partial area ($g = 0.42$; 95% CIs = 0.07, 0.77). No significant group main effects were not observed for medial condylar thickness ($F_{1,164} = 2.86$; $p = 0.09$; $\beta = 0.39$; $g = 0.21$; 95% CIs = - 0.14, 0.56, knee injury = 2.07 ± 0.20 mm, control = 1.96 ± 0.59 mm) or echo intensity ($F1,164 = 0.37$; $p = 0.54$; $\beta = 0.09$; $g = - 0.11$; 95% CIs = - 0.46, 0.24, knee injury = 30.83 ± 10.32, control = 31.95 ± 10.37) with small effect sizes.

Table 4-3. Femoral articular cartilage variables pre-and post-season assessments for the knee injury history and control groups.

Mean (SD)	**Pre-season**					**Post-season**				
	Thickness (mm)			**Cross-sectional Area (mm^2)**	**Echo-intensity**	**Thickness (mm)**			**Cross-sectional Area (mm^2)**	**Echo-intensity**
	Lateral	**Intercondylar**	**Medial**			**Lateral**	**Intercondylar**	**Medial**		
Knee Injury (n=42)	2.38*# (0.48)	2.51* (0.54)	2.10# (0.43)	45.44*# (8.90)	30.33 (14.32)	2.35*# (0.45)	2.52* (0.55)	2.04# (0.41)	43.90*# (8.65)	31.32 (13.03)
Control (n=124)	2.30*# (0.40)	2.40* (0.54)	2.00# (0.37)	42.76*# (7.72)	33.30 (13.90)	2.16*# (0.36)	2.24* (0.52)	1.93# (0.37)	40.44*# (7.33)	30.60 (9.93)

SD: Standard Deviation

*Significant main effects for group ($p < 0.05$) indicate that the knee injury group had greater lateral and intercondylar thickness as well as cross-sectional area than the control group.

#Significant main effects for time ($p < 0.05$) indicate that pre-season femoral cartilage values were greater than post-season valuables, regardless of group.

The following pre-season US femoral articular cartilage variables were significantly greater than post-season variables, regardless of group membership; lateral condylar thickness ($F_{1,164}$ = 5.54; p = 0.02; β = 0.65; g = 0.18; 95% CIs = - 0.04, 0.39, pre = 2.34 ± 0.47 mm, post = 2.26 ± 0.43 mm), medial thickness ($F_{1,164}$ = 5.03; p = 0.03; β = 0.61; g = 0.16; 95% CIs = − 0.05, 0.38, pre = 2.05 ± 0.43 mm, post = 1.98 ± 0.43 mm), and partial area ($F_{1,164}$ = 11.51; p = 0.001; β = 0.92; g = 0.21, 95% CIs = 0.00, 0.43, pre = 44.10 ± 9.23 mm^2, post = 42.17 ± 8.82mm^2).

There were no significant time main effects for intercondylar thickness ($F_{1,164}$ = 2.57; p = 0.11; β = 0.36; g = 0.12; 95% CIs = - 0.10, 0.33, pre = 2.45 ± 0.61 mm, post = 2.38 ± 0.60 mm) or echo intensity ($F_{1,164}$ = 0.43; p = 0.51; β = 0.10; g = 0.06; 95% CIs = − 0.16, 0.27, pre = 31.82 ± 16.10, post = 30.96 ± 12.42). There were no significant group × condition interactions for lateral condylar thickness ($F_{1,164}$ = 1.92; p = 0.17; β = 0.28), medial thickness ($F_{1,164}$ = 0.07; p = 0.79; β = 0.06), intercondylar thickness ($F_{1,164}$ = 3.28; p = 0.07; β = 0.44), partial area ($F_{1,164}$ = 0.46; p = 0.50; β = 0.10), or echo intensity ($F_{1,164}$ = 2.00; p = 0.16; β = 0.29).

4-4. Discussion

Participation in sports and regular physical activity does not necessarily increase a risk of knee OA (27), but traumatic knee joint injury sustained during athletic activity has been recognized as a risk factor for knee OA (20) (140). The present study aimed to examine the short-term longitudinal changes in US femoral articular cartilage variables in an athletic population with and without a previous history of traumatic knee joint injury. The main findings in this study was that femoral articular cartilage thickness was associated with a previous history of knee intracapsular injury in an athletic population. Specifically, collegiate rugby players with a previous history of intracapsular knee injury demonstrated greater lateral condylar and intercondylar thickness, as well as CSA, compared to control players. The moderate effect size values for lateral thickness, medial thickness, and CSA, with CIs that did not cross zero, suggests clinically meaningful differences between groups may be present. A previous investigation has reported greater medial and lateral condylar thickness and greater CSA in individuals with a history of ACL reconstruction compared to control participants (134). Furthermore, previous authors demonstrated longitudinal increases in medial femoral articular cartilage thickness at 1 year (141), 2 years (142), and 5 years following ACL injury (116). Consistent with the findings of previous investigations of ACL injury populations, our findings from the current study indicates that traumatic knee intracapsular injury may alter the macrostructure of the femoral articular cartilage in rugby players. This study provides evidence that supports the need for future investigations to longitudinally monitor cartilage structure when determining the association between joint health and traumatic knee joint injury in competitive athletic populations.

We observed that both the knee injury history group and control groups exhibited decreased femoral cartilage thickness and partial area following a 5-month competitive season. This finding suggests monitoring changes in femoral cartilage macrostructure during an athletic season may be a critical step towards the creation of optimal exercise-related strategies to mediate the risk of knee OA. While the small effect sizes, with CIs that crossed zero, suggest differences in cartilage thickness between pre-and post-season may be not clinically meaningful, our findings of the amount of changes in femoral articular cartilage thickness on US are somewhat similar to those of a previous study in which authors demonstrated that femoral articular cartilage had longitudinal change their structures in mature volleyball athletes (102). Specifically, Eckstein et al. (102) observed mature volleyball athletes displayed a decrease in medial (- 0.32 mm) and lateral cartilage thickness (- 0.16 mm) at 2-year follow-up assessments.

These findings from the previous (102) and current studies may indicate that the effects of physical activity may lead to pronounced short-term and long-term differences in macrostructure of femoral cartilage. Participation in competitive sport might be associated with the greater frequency of shear or torsional loading induced by abrupt lateral, decelerated, and accelerated movements (20). The shear or torsional load accumulated by sport-related activities for a longer period may cause chronic changes in the femoral articular cartilage. This is on speculative and future studies are needed to examine the long-term effect of various modes of physical activities and different knee joint loading on femoral articular cartilage macrostructure.

The unique aspect to this study is that we assessed a short-term longitudinal change in the anterior femoral cartilage size following a 5-month competitive season in collegiate rugby players with a history of knee injury. However, no significant interactions between injury and time were observed for all femoral cartilage outcome measures. These findings indicate that players with a knee joint injury history may not present with a progressive thickening or thinning of anterior femoral articular cartilage during a competitive season. We speculate that no progressive changes in anterior femoral articular cartilage in rugby players with knee injury history might be due to movement strategies they utilized as an effort to protect the knee joint. Rugby athletes in the knee injury history group received supervised physical rehabilitation following a knee injury. It is possible that those with a history of knee injury have restored proper knee function and the ability to attenuate the external loads following supervised rehabilitation programs. Furthermore, previous researchers have suggested that individuals, who have received surgical management following ACL injury, may offload the injured limb to avoid placing excessive stress on injured joint in order to minimizing a risk for further tissue damage or re-injury (143). This compensation mechanism may influence mechanical joint loading of the cartilage and its structure in the injured knee. The explanations are speculative. Clearly, a prospective investigation is needed to determine if movement strategies and joint loading patterns in rugby athletes with a knee injury history are associated with femoral cartilage thickness and CSA.

While we assessed the short-term cartilage response immediately after a competitive season, we are unaware of the degree to which femoral articular cartilage could recover its macrostructure following a period of lower loading intensity and duration, such as off-season. A previous investigation observed slower recovery to baseline cartilage morphology following cartilage deformation induced by higher intense activity (24). The high resilience of cartilage can minimize the risk of cartilage failure (144), and slower recovery of femoral articular cartilage structure after removing loading accumulated by physical activity is one of factors that increase a risk for knee OA (36). Cartilage recovery following the season could differ between rugby players with and without a history of traumatic knee joint injury. Additional assessment of cartilage recovery after the athletic season (i.e., assessing cartilage macrostructure following off-season) as an indicator of cartilage resiliency may better monitor longitudinal cartilage conditioning of rugby players with a knee joint injury history following a long period of high intense activity. Future studies should examine both cartilage response and recovery following the repetitive loading induced by cutting, deceleration, and acceleration movements in athletic populations with traumatic knee joint injury.

While we observed a significant decrease in lateral and medial condyle thickness and cross-sectional area following a 5-month competitive season, these structural changes were not accompanied by alterations in echo intensity. Echo intensity may be sensitive to change in the hydration status of cartilage and cartilage loss is associated with fluid exudation (145). Previous study reported echo intensity measures were associated with a presence of arthroscopic-based cartilage damage (89). Future investigations are needed to validate US echo intensity as a clinically accessible assessment tool to detect longitudinal changes in cartilage water content and cartilage composition and monitor joint tissue health.

In the current study, there are some limitations and caveats that open the door for future research. The heterogeneity of severity in intracapsular injury, the time since the most recent injury, and injury types makes it difficult to generalize findings to one specific type of knee joint intracapsular injury, as these are factors that may influence the results of our study. The average time since knee intracapsular injury in our participants was 19.94 ± 16.09 months, and the range of time since knee intracapsular injury was wide (2-59 months). Therefore, we ran a post-hoc exploratory Pearson product moment correlation analyses to evaluate the association between time since intracapsular injury and anterior femoral articular cartilage morphology using data from the current study. We found negligible negative correlations of the anterior femoral articular cartilage thickness and CSA with time since injury in players with a history of intracapsular injury ($r = -0.03 \sim 0.11$, $p > 0.05$). However, future studies are required to examine interaction between injury and competitive athletic activities for anterior femoral articular cartilage macrostructure in a more homogenous injury cohort.

While all players participated in the same practice, conditioning, and training sessions, we did not consider other possible influential factors such as game hours played, position played, and other physical activities in which players participated beside rugby team activities. While there were no differences in game and practice days participated between players with and without a history of knee injury ($p > 0.05$), the cumulative load during this investigation may be different among participants. Furthermore, we did not assess knee alignment (e.g., knee valgus) that influences the femoral articular cartilage thickness and contributes to the progression of knee OA (146). Future investigations will need to include the factors identified above to comprehensively understand the effects of knee joint intracapsular injury on morphological characteristics of anterior femoral cartilage in this athletic population. Observer bias may be introduced into the US image analysis by an unblinded examiner performing all of the US image analyses. Future studies should conduct with a blinded design as examiners are blinded to group memberships. Lastly, an US does not allow for capturing the enter cartilage at the tibiofemoral and patellofemoral joint, and we could determine effects of knee joint injury and competitive season on thickness and size in other femoral cartilage compartments. While an US method utilized by this study has been accepted for viewing the lateral and medial femoral condyle as well as intercondylar notch (63) (64), anatomically this method is limited to capture the most anterior position of the femoral articular cartilage, which may represent both patellofemoral and tibiofemoral articulations.

4-5. Summary

We observed that collegiate rugby athletes with a previous history of knee intracapsular injury had greater anterior femoral articular cartilage lateral condyle thickness, intercondylar thickness, and CSA compared with uninjured control players. Traumatic intracapsular injury at the knee joint may alter the macrostructure of the femoral cartilage in rugby players, which may affect the long-term joint and cartilage health. Short-term longitudinal changes in anterior femoral articular cartilage structure were observed in collegiate rugby athletes following a competitive season, regardless of the presence of a knee injury history. These findings suggest regular femoral cartilage health assessments during an athletic season may be a critical step towards the creation of optimal exercise-related strategies to mediate the risk of knee OA.

Chapter V

Morphological Changes in the Femoral Articular Cartilage Caused by Participating in the Two-Year Consecutive Collegiate Rugby Seasons With and Without a History of Knee Joint Injury During Two-Year Consecutive Competitive Seasons

5-1. Introduction

Participating in sports helps maintain cartilage health but increases the likelihood of sustaining joint injury. In the physically active population, the knee ranks among the most commonly injured joints (75) (147). Injuries to the knee joint alter joint structures and negatively affect the joint's capability to attenuate loading. As a result, the changes in articular cartilage macrostructure and cartilage composition contribute to the early onset of OA. Although OA is a common condition among the elderly, OA associated with joint injuries is prevalent among the young populations (147). The term for OA caused by joint injuries is post-traumatic osteoarthritis (PTOA) (75). People who suffer from knee joint injury are four to six times more likely to develop PTOA than those who have no previous history of knee joint injury (4) (75) (77) (78). Therefore, knee joint injury becomes a significant risk factor for developing PTOA among the young populations.

Intracapsular knee joint injuries are also the most common (148) (149) (150) and severe type of joint injury among rugby players (149) (151). Rugby has a high prevalence of developing symptomatic radiographic knee OA related to a traumatic knee joint injury (20). In a previous study focusing on former rugby players, approximately 40% of the participants experienced a practice reduction of 4 or more weeks due to their injuries (20). Additionally, 39% of all participants had been diagnosed with knee OA, and 12% had undergone knee replacement (20). Symptoms of PTOA, such as joint pain (8) (152) and functional limitations (152), interfere with their performances and daily activities (8) and lead to reduced quality of life (8) (152).

Articular cartilage has a resilience that demonstrates a change in its structure when subjected to mechanical stress on the joint and recovers to its baseline (69) (144). A previous study has observed the acute response of femoral articular cartilage thickness following physical activities (26). This US study demonstrated femoral articular cartilage thickness changes in response to the mechanical load during walking and drop-landing, and its thickness gradually returned to its baseline thickness over time after ceasing physical activities (26). Several studies observed longitudinal changes in femoral cartilage structures for collegiate athletes (18) (92). A MRI study has observed compositional and structural changes in the femoral articular cartilage during a competitive season and an off-season for collegiate basketball players (18). This study used quantitative MRI to compare compositional and structural changes in knee articular cartilage between the first pre-season and the second pre-season. However, this study did not target populations with a previous history of knee joint injuries (18). An early stage of OA progression may be associated with an increase in articular cartilage thickness (62) (153). The increase in articular cartilage thickness may be attributed to lower US image quality, which could potentially manifest as the blurring of the cartilage margin in the US (154). However, it is insufficient to examine morphological changes in the articular cartilage occurring within a single season. By clarifying the changes in femoral cartilage structure at multi-season, it is necessary to understand the effects of participating in competitions on femoral articular cartilage and the changes in femoral articular cartilage that occur after competitions.

Attending supervised rehabilitation following a traumatic knee joint injury is important to prevent the development of PTOA. Supervised rehabilitation after a knee injury has been shown to restore sensorimotor function and functional joint stability (98) (155) (156), potentially mitigating the risk of PTOA. Furthermore, a previous investigation has demonstrated supervised rehabilitation improved articular cartilage composition in patients with a traumatic intracapsular knee joint injury (98). These findings from previous studies indicate that supervised rehabilitation may be beneficial to articular cartilage health in the secondary and tertiary prevention of PTOA after a traumatic intracapsular knee joint injury (98) (155) (156). However, it is unclear how receiving and successfully completing rehabilitation at the time of a traumatic knee joint injury affects the changes in femoral articular cartilage at multi-season. This could be useful information for maintaining femoral cartilage health in athletes who continue participating in sports after injury. Therefore, we aimed to evaluate femoral articular cartilage thickness over two consecutive seasons in collegiate rugby players with a history of intracapsular knee joint injuries that had received supervised rehabilitation after their knee injury and those without a history of knee joint injury. We hypothesized that collegiate rugby players with a previous intracapsular knee joint injury, who had received supervised rehabilitation after their traumatic knee injury, would demonstrate no remarkable change in femoral articular cartilage thickness and CSA following two competitive rugby seasons compared with the control players. Furthermore, the thickness of femoral articular cartilage has recovered to the original state at the beginning of the second season compared to the end of the first season.

5-2. Methods

Study Design

This prospective observational study was approved by the Ethics Review Committee for Medical and Health Research Involving Human Subjects at Ritsumeikan University. Before participating in the study, all participants provided informed written consent, and parental or guardian consent was obtained if necessary. This study assessed US outcomes measures of distal femoral cartilage morphology before and after two consecutive rugby seasons in collegiate rugby players with or without a previous history of knee joint injury (Figures 5-1, 5-2).

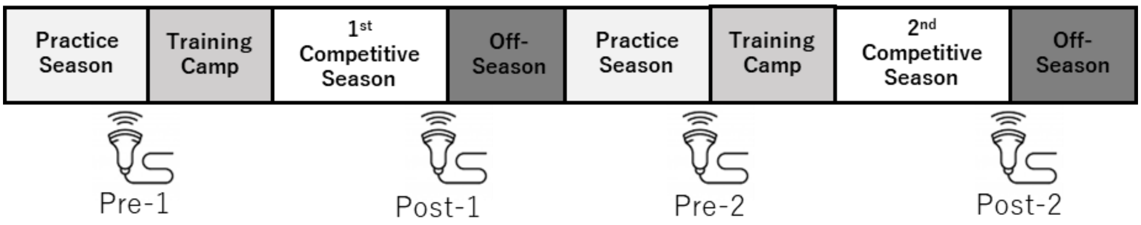

Figure 5-1. Data collection timeline: Pre-season assessments were performed prior to summer training camp, and post-season assessments were conducted immediately after the conclusion of the seasons.

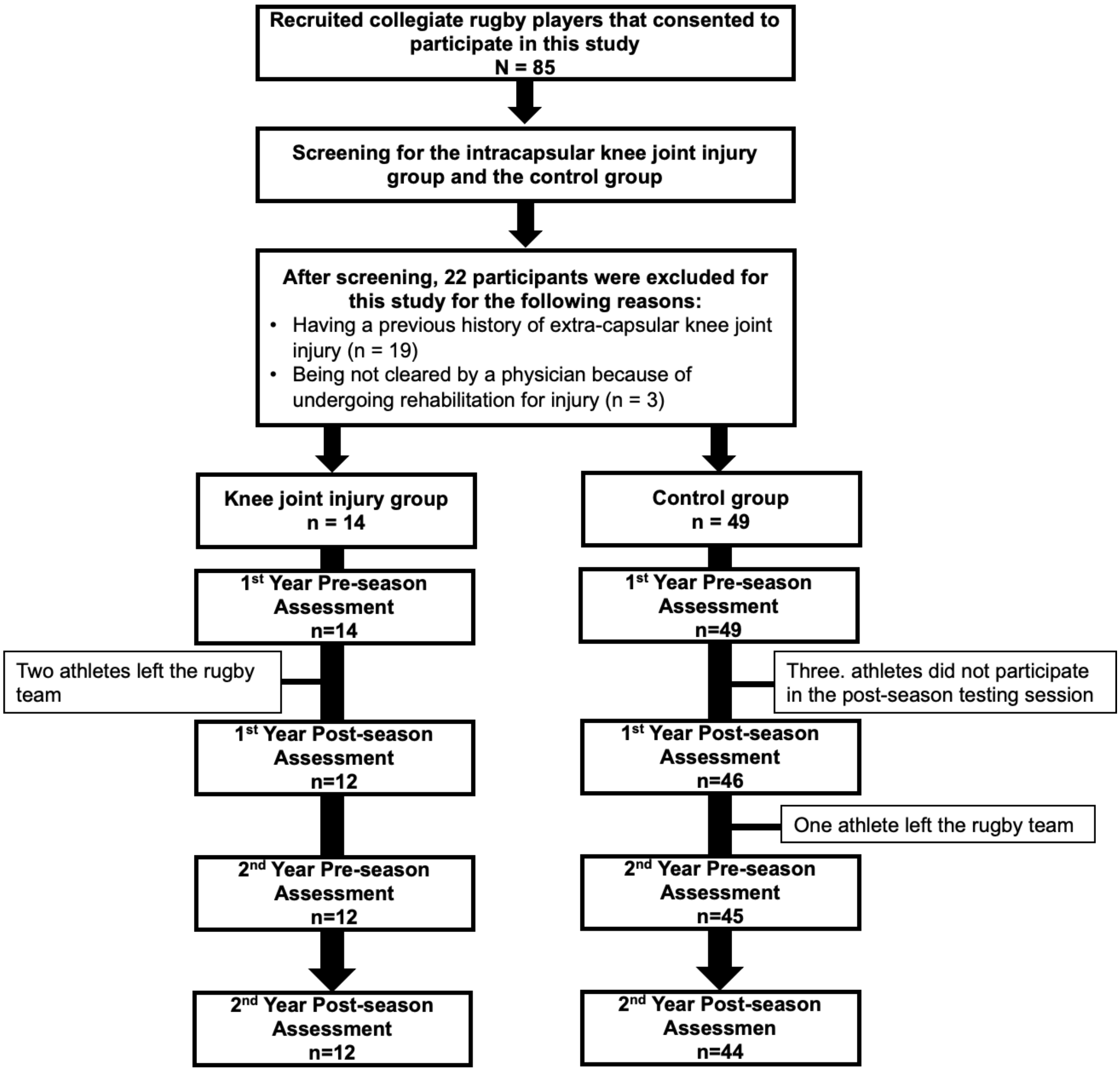

Figure 5-2: A flow chart for study participants

Participants

Eighty-five male rugby players from a Division I collegiate team were recruited for this study. After examining each player's qualifications, 63 players were included in either the injury-rehabilitation history (n = 14) or the non-injury history group (n = 49). The knee joint injury-rehabilitation history (Injury-Rehab) group was required to (1) have at least one significant previous history of traumatic intracapsular knee joint injuries (e.g., anterior and posterior cruciate ligament sprain, lateral and medial meniscus injury, and femoral cartilage injury) and (2) have received and completed supervised rehabilitation after their knee injury. Because this study focused on the effect of traumatic intracapsular knee joint injury, players with a previous history of extra-capsular knee joint injury (e.g., medial and lateral collateral ligament sprain) were excluded. The control group consisted of rugby players with no previous history of knee joint injury.

During the competitive rugby season, players participated in regular practices and games once a week. Daily practices were held on artificial turf, 1.5 to 2 hours per session, six times a week, and players also had 1 to 1.5 hours of strength training sessions four to five times a week. Some athletes were absent from team practices and games due to injury or sickness; however, all participants joined team practices and competitions throughout the seasons. The team's athletic trainer provided all medical records.

Data Collection Procedures

A single investigator (M.H.) took all femoral cartilage US assessments and analyzed the US examinations. The examinations were performed before (pre-season) and after (post-season) at each competitive season. The pre-season tests were conducted before summer training camp in late July, while the post-season examinations were conducted immediately after the season ended in late December. The following four points of measurement were established in this study: pre-season 1 (Pre-1), post-season 1 (Post-1), pre-season 2 (Pre-2), and post-season 2 (Post-2) (Figure 5-1).

Ultrasonographic Assessment of the Femoral Articular Cartilage

We asked participants not to engage in any type of exercise the day before taking the US image. Upon arriving at the examination room, participants rested in a long-sit position with both knees in full extension for 30 min to unload the cartilage (26). After 30 min of rest, participants laid supine comfortably on an examination table with both knees positioned at 140° of flexion, using a handheld goniometer (24). An ultrasound unit (LOGIQe V2; General Electric Co., Fairfield, CT, USA) with a 12-MHz linear probe was used to obtain US images of the femoral articular cartilage for this study. The system settings of the portable ultrasound unit were consistent between pre-season and post-season testing sessions and among participants. The linear probe was placed anteriorly over the distal femoral articular cartilage of the medial and lateral femoral articular condyle in the axial position and above the superior margin of the patella. The liner probe had oriented to obtain the maximum reflection of the femoral trochlea and the overlying hyaline cartilage as a previous study reported (24) (26) (63). The location at the intercondylar notch was centered on the screen was captured the image. Three images were recorded at each testing session and the liner probe being removes and repositioned on the knee between images.

Ultrasonographic Image Processing

A single investigator (M.H.) assessed all femoral articular cartilage image processing by using ImageJ software (National Institutes of Health, Bethesda, MD, USA). The medial and lateral cartilage thickness was estimated at the intercondylar notch, as well as 1cm apart in the medial and lateral directions (137). The femoral cartilage thickness was measured as the straight-line distance (mm) drawn from the hyperechoic cartilage-bone interface to the synovial space-cartilage interface (24) (Figure 4-2A). The CSA (mm^2) of femoral articular cartilage was assessed by tracing an area of femoral cartilage between a lateral and a medial measurement points where femoral cartilage thickness was measured (Figure 4-2B). The US examination of the distal femoral cartilage variables has been demonstrated to be valid (63) (64) and reliable (ICC = 0.83-0.99) (24). Furthermore, before data collection for the current study, the US examiner established good to excellent intratester reliability ($ICC_{1,3} = 0.82$-0.96).

Statistical Analysis

Anthropometric valuables were compared between the Injury-Rehab and control groups using independent t-tests ($p < 0.05$). For the Injury-Rehab group, the injured limb was used to assess the articular cartilage structures. The control group used the dominant limb, the side of kicking the ball (18). A separate 2 × 4 (group × time) repeated measures ANOVAs was used to analyze each US femoral cartilage valuables. In the case of the statistically significant interactions and main effects, a Bonferroni post-hoc univariate analysis with a pairwise comparison was conducted to ascertain the location of significant differences. Statistical significance was set at $p < 0.05$ using SPSS 27.0 (SPSS, Inc. Chicago, IL.). Cohen's d effect sizes using pooled standard deviations were calculated to assess the magnitude of differences in each femoral cartilage measurement between independent valuables, along with 95% confidence intervals (CIs) for each pairwise comparison. The strength of effect sizes was interpreted as weak ($d < 0.40$), moderate ($0.40 \leq d < 0.80$), or strong ($d \geq 0.80$) (105).

5-3. Results

Fifty-six out of 63 participants (89%) completed all four-time points of femoral cartilage US assessments (Figure 5-2). The type and frequency of intracapsular knee joint injuries sustained by the participants were reported in Table 5-1.

Table 5-1. List of previous intracapsular knee injury history

Injury Type	Frequency	The Number of Surgical Management
Anterior cruciate ligament (ACL) sprain	4	3
Posterior Cruciate ligament (PCL) sprain	6	0
Medial meniscus injury (MMI)	2	1
Lateral meniscus injury (LMI)	2	2
Femoral cartilage injury	1	0

The number of participants who sustained multiple knee joint injuries:

ACL + LM = 1

PCL + PCL = 1

MM + LM = 1

Rehabilitation was overseen by orthopedic surgeons, physical therapists, athletic trainers, or a team specializing in conditioning and strength. The supervised rehabilitation lasted for an average of 152 days (minimum: 30 days, maximum: 323 days). The rehabilitation regimens encompassed exercises to restore ROM, build strength, engage in functional training (balance, proprioception), weight training, plyometrics, aerobic exercises (walking, cycling, running, sprinting), agility, and sport-specific training (tackling, scrimmage, kicking, etc.). The frequency of rehabilitation sessions was tailored to the individual athlete's status and progression. Anthropometric characteristics were not statistically different between the Injury-Rehab and the control groups ($p < 0.05$, Table 5-2).

Table 5-2. Demographic characteristics for the intracapsular knee injury and control groups

	Knee injury	Control	p-value
	Mean (SD)	Mean (SD)	
N	12 males	44 males	
Age (yrs.)	19.33 (1.15)	19.43 (0.82)	0.74
Height (cm)	173.17 (6.06)	174.48 (6.67)	0.54
Body Mass (kg)	84.33 (12.44)	84.29 (11.43)	0.99
Body Mass Index (kg/m²)	28.10 (3.67)	27.66 (3.29)	0.69
Body Fat Percentage (%)	21.79 (4.92)	21.15 (4.38)	0.66
# of previous intracapsular knee joint injuries	1.33 (0.49) Min=1, Max=2	0.00	-
# of knee joint surgeries	1.58 (0.52) Min=1, Max=2	-	-
Time since the most recent inter-capsular knee joint injury (month)	16.96 (13.88) Min=2, Max=47	-	-

SD: Standard Deviation

The group mean and standard deviation of femoral cartilage variables at the four assessment time points are shown in Figure 5-3 and Table 5-3. There were no significant Injury-Rehab main effects for the lateral condylar thickness ($p = 0.17$), the intercondylar thickness ($p = 1.10$), the medial condylar thickness ($p = 0.51$) and CSA ($p = 0.15$). There were no significant Time × Injury-Rehab interactions for the lateral condylar thickness ($p = 0.47$), the intercondylar thickness ($p = 0.55$), the medial condylar thickness ($p = 0.25$), and CSA ($p = 0.67$). Significant Time main effects were observed for the lateral condylar thickness ($p < 0.001$), the intercondylar thickness ($p = 0.001$), the medial condylar thickness ($p < 0.001$), and CSA ($p < 0.001$) (Tables 5-3, 5-4, 5-5). Compared to Pre-1, there were significantly less lateral condylar cartilage thickness at Post-1, Pre-2, and Post-2 with medium to large effect sizes; intercondylar cartilage thickness at Pre-2 and Post-2 with medium effect sizes; medial condylar cartilage thickness at Post-1, Pre-2, and Post-2 with small to medium effect sizes; as well as CSA at Post-2, Pre-2, and Post-2 with medium effect sizes ($p < 0.05$) (Table 5-4, 5-5).

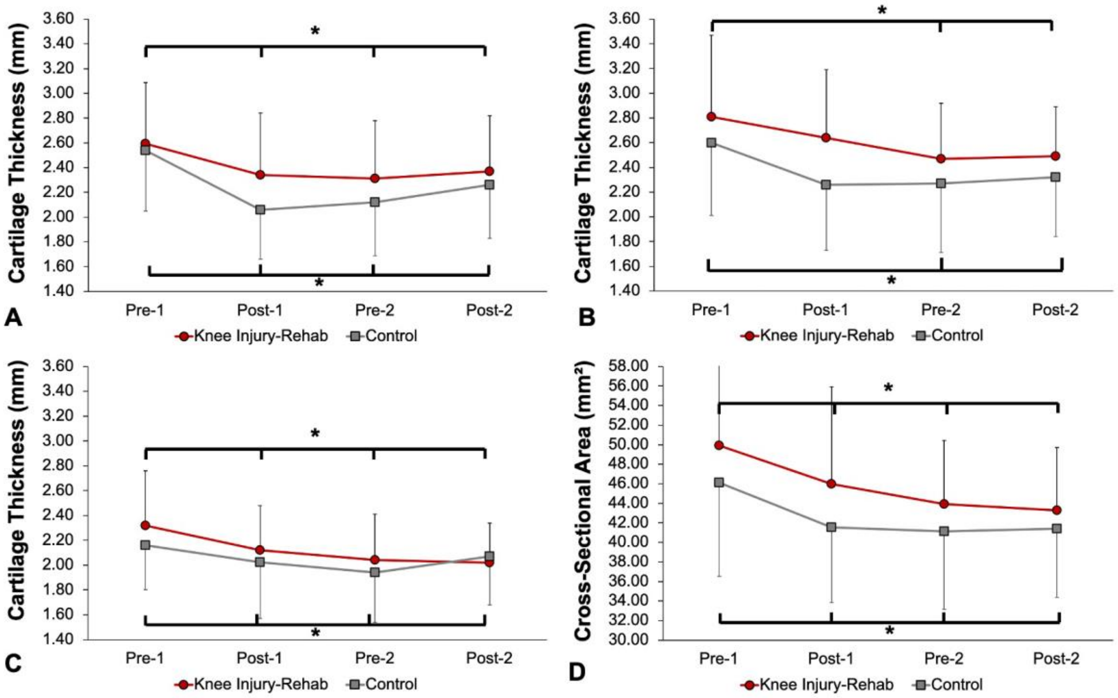

Figure 5-3. Changes in femoral articular cartilage variables over two-competitive seasons:

A) Lateral Condylar Cartilage Thickness,

B) Intercondylar Cartilage Thickness,

C) Medial Condylar Cartilage Thickness,

D) Cross-sectional Area

*Significant time main effects were observed (P <0.05). The femoral articular cartilage variables at the pre-season assessment in the first year (Pre_1) were significantly lower compared to the post-season assessment at the conclusion of the first year season, the pre-season assessment in the second year (Pre_2), and the post-season assessment in the second year (Post_2) (P<0.05).

Table 5-3. Mean femoral articular cartilage outcomes: pre- and post-season measurements for two-consecutive years.

Outcomes	**Time**	**Injury** Mean (SD)	**Control** Mean (SD)	**Main effect/Interaction**	**p-value**	**Effect size (η^2)**	**Power (β)**
Lateral Condylar Thickness (mm)	Pre-1	2.60 (0.49)	2.54 (0.49)	Time	<0.001*	0.16	1.00
	Post-1	2.34 (0.50) #	2.06 (0.40) #	Knee Injury	0.17	0.04	0.28
	Pre-2	2.31 (0.47)	2.12 (0.43) #	Time * knee injury	0.47	0.02	0.21
	Post-2	2.37 (0.45) #	2.26 (0.43) #				
Intercondylar Thickness (mm)	Pre-1	2.81 (0.66)	2.60 (0.59)	Time	0.001*	0.11	0.95
	Post-1	2.64 (0.55)	2.26 (0.53)	Knee Injury	1.10	0.05	0.38
	Pre-2	2.47 (0.45) #	2.27 (0.56) #	Time * knee injury	0.55	0.01	0.17
	Post-2	2.49 (0.40) #	2.32 (0.48) #				
Medial Condylar Thickness (mm)	Pre-1	2.32 (0.44)	2.16 (0.36)	Time	<0.001*	0.12	0.98
	Post-1	2.12 (0.36) #	2.02 (0.45) #	Knee Injury	0.51	0.01	0.10
	Pre-2	2.04 (0.37) #	1.94 (0.40) #	Time * knee injury	0.25	0.03	0.34
	Post-2	2.02 (0.32)	2.07 (0.39) #				
Cross-sectional Area (mm²)	Pre-1	49.91 (9.87)	46.12 (9.57)	Time	<0.001*	0.15	1.00
	Post-1	45.98 (9.96) #	41.53 (7.71) #	Knee Injury	0.15	0.04	0.31
	Pre-2	43.92 (6.51) #	41.13 (7.99)	Time * knee injury	0.67	0.01	0.13
	Post-2	43.27 (6.43) #	41.38 (6.98) #				

SD: Standard Deviation

*Significant time main effects were observed ($p < 0.05$).

#The femoral articular cartilage variables were significantly lower compared to these variables at the pre-season assessment in the first year (Pre-1) regardless of group membership ($p < 0.05$).

Table 5-4. Pairwise comparison between ultrasonography assessment time points and between the injury-rehab and control groups.

Outcomes	Time		p-value	Effect size (d)	95% CIs		Between-group comparison		p-value	Effect size (d)	95% CIs	
					Lower	Upper					Lower	Upper
Lateral Condylar Thickness	Pre-1	Post-1	0.001*	0.93	0.54	1.32	Injury-rehab vs. Control	Pre-1	0.74	0.12	-0.52	0.76
		Pre-2	<0.001*	0.84	0.45	1.22		Post-1	0.05	0.66	0.01	1.31
		Post-2	0.03*	0.58	0.20	0.95		Pre-2	0.19	0.43	-0.21	1.08
	Post-1	Pre-2	1.00	-0.09	-0.46	0.28		Post-2	0.41	0.25	-0.39	0.89
	Pre-2	Post-2	0.47	-0.27	-0.64	0.10						
Intercondylar Thickness	Pre-1	Post-1	0.07	0.54	0.16	0.92	Injury-rehab vs. Control	Pre-1	0.30	0.35	-0.29	0.99
		Pre-2	0.01*	0.60	0.22	0.97		Post-1	0.03	0.71	0.06	1.36
		Post-2	0.02*	0.54	0.16	0.92		Pre-2	0.26	0.37	-0.27	1.01
	Post-1	Pre-2	1.00	0.06	-0.32	0.43		Post-2	0.27	0.37	-0.28	1.01
	Pre-2	Post-2	1.00	-0.10	-0.47	0.27						
Medial Condylar Thickness	Pre-1	Post-1	0.03*	0.36	-0.02	0.73	Injury-rehab vs. Control	Pre-1	0.21	0.42	-0.22	1.07
		Pre-2	<0.001*	0.60	0.21	0.97		Post-1	0.50	0.23	-0.41	0.87
		Post-2	0.01*	0.34	-0.03	0.71		Pre-2	0.26	0.25	-0.39	0.89
	Post-1	Pre-2	0.85	0.20	-0.17	0.57		Post-2	0.24	-0.13	-0.77	0.51
	Pre-2	Post-2	1.00	-0.26	-0.63	0.11						
Cross-sectional Area	Pre-1	Post-1	0.01*	0.49	0.12	0.87	Injury-rehab vs. Control	Pre-1	0.23	0.39	-0.25	1.04
		Pre-2	0.01*	0.59	0.20	0.97		Post-1	0.10	0.54	-0.10	1.19
		Post-2	0.01*	0.61	0.24	0.99		Pre-2	0.65	0.36	-0.28	1.00
	Post-1	Pre-2	1.00	0.09	-0.28	0.47		Post-2	0.40	0.28	-0.37	0.92
	Pre-2	Post-2	1.00	-0.01	-0.38	0.36						

CI: Confidence Interval

*the femoral articular cartilage valuables were significantly lower compared to theses variables at the pre-season assessment in the first year (Pre-1) regardless of group membership ($p < 0.05$).

5-4. Discussion

This study aimed to examined structural changes in the femoral articular cartilage in collegiate rugby players with a previous history of intracapsular knee joint injury who have received and successfully completed supervised rehabilitation after their previous injury and those without a history of knee joint injury for two competitive rugby seasons. The findings of this study were that rugby players demonstrated a decrease in femoral articular cartilage thickness and CSA from Pre-1 to Post-2, supported by medium to large effect sizes with 95% confidence intervals that did not cross zero. These findings indicate that engaging in collegiate rugby induces alterations in femoral articular cartilage structure, and the reduction in cartilage thickness and CSA were clinically relevant. It prompts consideration of necessary measures to preserve athletes' cartilage health and mitigate the risk of knee OA. Additionally, we found no differences in lateral condylar thickness, the intercondylar thickness, the medial condylar thickness, and CSA between those with and without a previous history of traumatic knee joint injury. These results indicate attending supervised rehabilitation after a knee injury may reduce impacts of a previous history of intracapsular knee joint injury on the changes in femoral articular cartilage thickness among collegiate rugby players. However, it is important to acknowledge that these findings are limited to the short term, and we cannot extend the implications of attending supervised rehabilitation on cartilage health beyond the study duration.

In this study, we observed a decrease in femoral articular cartilage thickness among participants during competitive rugby seasons. There were medium and large effect sizes for lateral condylar and intercondylar cartilage thickness and CSA at Post-1, Pre-2, and Post-2 compared to Pre-1, and 95% confidence intervals around the effect sizes did not cross zero. The effect size for medial condylar thickness between Pre-1 and Pre-2 was medium with 95%

confidence intervals that did not overlap zero. The medium and large effect sizes with narrow 95% confidence intervals bolstered our results by highlighting the moderate to large magnitude of reduction in articular cartilage thickness and CSA during two consecutive rugby seasons. It has been assumed that differences in loading environment may have a significant influence on the morphological characteristics of articular cartilage (2). During rugby matches, players engage in a spectrum of activities, including low- and high-intensity running, rapid accelerations and decelerations, and high intensity contact and set plays (2) (110) . The increased repetitive shear loading from sudden lateral movements, braking, and acceleration in rugby may reduce femoral articular cartilage thickness and the fatigue life of cartilage (62), potentially contributing to an elevated risk of knee OA (157). Femoral articular cartilage thickness seems to be load-dependent (132) (158). Previous studies observed that exercises, such as walking, running, and drop-landing produced greater femoral cartilage deformation compared to a non-exercise condition (24) (26). However, it is unclear how different movement and joint loading patterns would be in terms of changes in cartilage thickness. Therefore, future investigation is required to examine morphological responses of femoral articular cartilage to different movement conditions, such as sudden lateral movements.

The observed decrease in femoral cartilage thickness over two-year competitive seasons in this study suggests a potential alteration in the distribution of mechanical stresses within the knee joint. Changes in femoral articular cartilage thickness are often considered indicative of alterations in its ability to withstand mechanical stress (116). A reduction in articular cartilage thickness may lead to a change in the loading area on the knee joint, suggesting it may not be fixed to a specific point (159). Additionally, a previous study demonstrated that this reduction in articular cartilage thickness led to a decrease in the contact area between the cartilage and meniscus (160), impacting the meniscus's capacity to efficiently transmit load. Thus, the findings may explain why participation in rugby is linked to a heightened risk of OA. Further research is needed to inform our understanding of types of loading for rugby players to reduce the risk for knee OA and to design prevention programs that facilitate cartilage health.

Changes in biomechanics due to injuries can potentially lead to modifications in contact mechanics, possibly increasing stress within the knee joint (159). Previous studies have reported an increase in femoral articular cartilage thickness after six months (94), one year (141), three years (161), four years (159), and five years after ACL reconstruction (116). However, the current study did not observe significant differences in any femoral articular cartilage thickness variables between rugby players with and without a previous history of knee joint injury. This study encompasses all intraarticular knee joint injuries and not just an ACL injury. It is possible that different changes in cartilage thickness, distinct from ACL injuries, may have influenced the absence of significant differences in the morphological changes of the femoral cartilage.

Participants in this study, who sustained a knee joint injury, were undergoing rehabilitation programs by a team physician and a team athletic trainer to return to play at the competitive level. The rehabilitation program included various components to restore knee joint function, such as strengthening, ROM exercises, and neuromuscular training (29) (161) (Appendix 2). Several studies have reported that an increase in knee extensor muscle strength effectively reduces pain (162) (163) and improves physical function (163). By continuing training after returning to rugby, participants can enhance muscle strength and functional performance, potentially delaying the early onset of PTOA (152). It is speculated that such rehabilitation programs and training may have prevented significant changes in femoral articular cartilage structures, even in individuals with a previous history, thus explaining the absence of observed alterations.

While no significant Injury-Rehab main effects were observed for the selected cartilage variables, some of the non-significant between-group difference in cartilage thickness were associated with medium effect sizes, with 95% CIs that did not cross zero. Specifically, the Injury-Rehab group exhibited greater lateral condylar and intercondylar cartilage thickness at Post-1 compared to the control group. The medium effect sizes indicate that these differences are likely of moderate magnitude, suggesting clinically meaningful differences between groups. In instances of medium effect sizes with 95% CIs extending across zero, these associations may be prone to statistical error and could benefit from a larger sample size to strengthen the findings. Previous studies reported greater femoral cartilage thickness in individuals with a history of a traumatic intracapsular knee joint injury compared to control participants (26) (92). Researchers have hypothesized that an increase in articular cartilage thickness may be attributed to cartilage swelling and/or hypertrophy of the extracellular matrix (164) (165) as well as be related to a progression of OA (62) (153). If the articular cartilage in the Injury-Rehab group is thicker compared to the control group, it appears that rehabilitation did not confer any benefits to the cartilage. Therefore, developing more effective interventions is crucial to modifying the adverse impacts of knee joint injuries on cartilage health.

This present study also examined the changes in femoral articular cartilage thickness and CSA during an off-season (Post-1 to Pre-2). A previous MRI study observed structural and compositional alterations in femoral articular cartilage during an off-season (18). Additionally, a study investigating acute change in femoral articular cartilage reported that thinning cartilage due to 30 min of exercise returns to baseline thickness over time after the completion of exercise (26). Based on these studies (18) (26), a hypothesis was proposed that undergoing off-season with reduced exercise load would allow the thinning cartilage during the competitive season to recover to baseline. However, in this study, the cartilage thickness did not recover during the off-season. We observed that the changes in femoral cartilage thickness ranged from -0.08 mm to 0.04 mm during the off-season. Small effect sizes were noted, accompanied by 95% CIs that overlapped zero when comparing US cartilage variables between Post-1 and Pre-2. The presence of small effect sizes, coupled with wide 95% confidence intervals, indicates that the reduction in articular cartilage thickness and CSA during the off-season was of minor magnitude. Notably, these changes in femoral articular cartilage thickness did not exceed the established minimal detectable change (0.09 mm-0.15 mm) (24) (26).

The previous study targeting collegiate basketball players defined the period without team practice as the off-season (18). On the other hand, we defined the off-season as the period from the end of the competitive season to the beginning of the next competitive season. As described in our study, the off-season encompassed a time when regular team practices were suspended, and athletes engaged in fundamental training and skill development in preparation for the upcoming competitive season. Although not on a regular basis, practice matches were also conducted. The strain exerted on the cartilage during such training sessions might explain the absence of morphological recovery in the cartilage. While articular cartilage possesses resiliency (69), the accumulation of training load on the joint can lead to overload, impeding the generation of essential components required to maintain the cartilage structure (49). A study on collegiate basketball players found altered femoral cartilage composition during the off-season, suggesting that the off-season strength and conditioning training may place more stress on the femoral cartilage (18). However, the authors observed some recovery in the anterior region of the femoral articular cartilage during the off-season (18). This finding implies that incorporating a designated period of rest and recovery, such as the off-season, can aid in restoring the cartilage structure altered by physical activity. In competitive sports, the off-season does not necessarily denote complete inactivity. Therefore, investigating the types of exercises and exercise loads that aid in maintaining and enhancing cartilage health would significantly contribute to extending the athletic careers and longevity of competitive athletes. For example, self-paced running has demonstrated its potential to stimulate improvements in cartilage health without elevating the risk of knee OA (27). It remains a challenge to determine the specific exercises and joint load most beneficial for training during the rest period.

A limitation of this study was that we did not consider participants' self-reported function. Even though participants actively engaged in team practices and games, we acknowledge the possibility that some may have experienced pain and functional limitations in both rugby and daily activities. Prior studies utilized self-reported questionnaires, such as KOOS, to assess the impact of knee joint injuries (29) (166). These studies highlighted significantly lower KOOS scores in the knee ligament injury group, particularly in the symptoms and knee-related quality of life subscales (29) (166). Long-term tracking and assessment using knee-related self-reporting questionnaires may shed light on structural changes in the degenerated knee joint of individuals with a history of knee joint injuries.

5-5. Summary

This study observed a significant decrease in femoral articular cartilage thickness and CSA during two consecutive rugby seasons in a collegiate rugby population. Participating in a competitive rugby season appeared to significantly impact the change in femoral articular cartilage thickness and CSA among active rugby players. Furthermore, we did not find an association between changes in femoral articular cartilage morphology and a previous history of intracapsular knee joint injury in collegiate rugby players. Attending supervised rehabilitation at the time of their knee joint injury may have a positive effect on femoral articular cartilage health in collegiate rugby players with a previous history of intracapsular knee joint injury. Future research should continue to monitor multi-season changes in femoral articular cartilage thickness, focusing on both knee joint injury status and participation in rugby competitions.

CHAPTER VI

Morphological Changes in the Femoral Articular Cartilage Response and Recovery from Various Exercises

6-1. Introduction

Articular cartilage is a mechanical component that reduces compressive and frictional force on the joint (68). Articular cartilage, a resilient structure, can adapt to absorb and distribute forces in the knee joint effectively (69), and the amount of the cartilage structure change depends on the magnitude of the force on articular cartilage (99). All physical activities in both daily living and sports activities create mechanical load on the joint. Physical exercises create greater force on the knee joint than daily living activities, such as walking, climbing stairs, and sitting or standing in a chair (133). Excessive stress can lead to the degeneration of cartilage structures (19) (68), even though the exact threshold for when stress becomes excessive is not established yet.

Recently, physical activities have been beneficial in maintaining cartilage health (69). Driban et al. (4) reported elite soccer players had a higher prevalence of knee osteoarthritis than handball and basketball. These sports activities combine several physical demands and different type of loading conditions on the joint; for example, acceleration and deceleration (22) (167), twisting (4) (19), and turning (4) motions have a significant effect on changes in cartilage structures. Previous studies have observed the acute response of femoral articular cartilage structure change following some physical activities, including walking, running, and drop-landing (23) (24) (25) (26). MRI study reported running condition had greater acute femoral articular cartilage thickness change than drop-landing when compared to the baseline (23), and a previous study with ultrasonography reported running became thinner than walking conditions at medial femoral cartilage thickness (24) and drop-landing task had greater femoral articular cartilage deformation than walking at medial, intercondylar, and lateral compartments (26). A previous US study reported acute femoral articular cartilage deformation and recovery after physical activities (26). In the study, femoral articular cartilage gradually returned the structure to the baseline after removing the mechanical force induced during walking and drop-landing (26). Previous investigations had quantified femoral articular cartilage thickness deformation only in unidirectional movement, such as walking and running; however, no study examined effects of cutting movement on femoral articular cartilage structure change.

Exercise is vital for maintaining optimal cartilage health; at present, it is not clear how different types of exercises and mechanical loading influence femoral cartilage morphology. Therefore, the purpose of this study was to compare femoral cartilage deformation in response to different movement conditions, cutting and running, using ultrasonography.

6-2. Methods

Study Design

A randomized cross-over study with repeated measures design was conducted to examine US outcome measures of distal femoral cartilage thickness in healthy individuals (Figure 6-1). Each participant reported to the research laboratory on three separate occasions, seven days apart. Participants completed two loading conditions (cutting and running conditions) and a control condition. The order of the three condition tasks was counterbalanced between participants. We provided participants with athletic shoes (ASICS, JOLT 3 extra wide, 22.5 to 28.0 cm) to minimize the effects of shoes. The US examinations were performed at pre-loading and two post-loading time points. A single investigator performed all femoral cartilage US assessments and analyses. This study was approved by the Ethics Review Committee for

Medical and Health Research Involving Human Subjects at Ritsumeikan University. All participants provided informed written consent before engaging in this study.

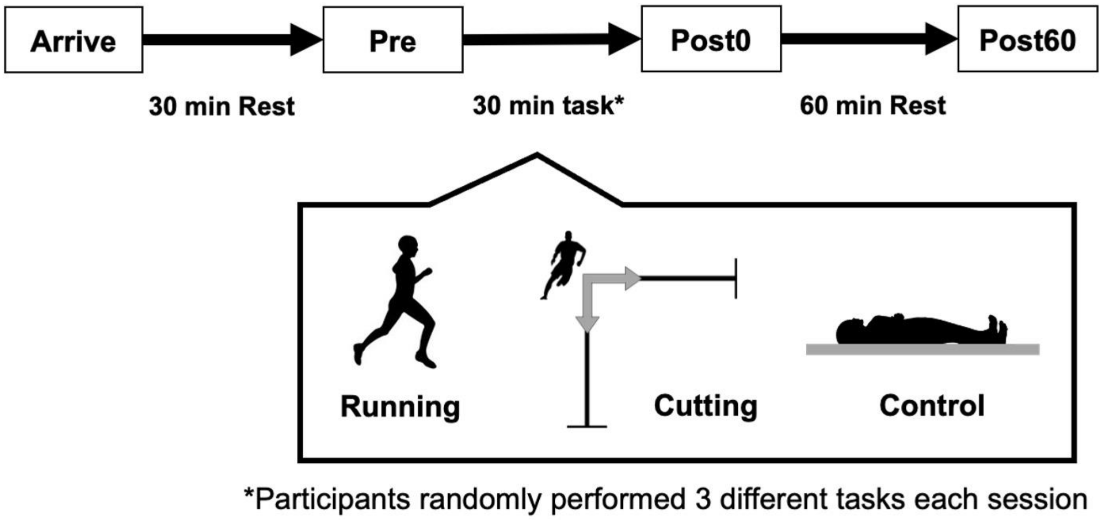

Figure 6-1. Schedule for the randomized data collection

Participants

Twenty-five healthy collegiate participants were recruited for this study. This study focused on femoral cartilage structure change; therefore, our exclusion criteria for participants were 1) a previous history of traumatic knee joint injury (e.g., anterior and posterior cruciate ligament injury, medial and lateral collateral ligament injury, meniscus injury, and cartilage injury), 2) a previous history of musculoskeletal injuries in the lower extremity, and 3) a history of surgery and fractures of the lower extremity. To eliminate the impact of competition characteristics, we excluded participants who participated in competitive sports in the three months before participation.

Data collection procedures

A single investigator (M.H.) performed all femoral cartilage US assessments and analyzed the US images. The examinations were performed before (pre-task) and after (post-task) the running, cutting, and control conditions. This study focused on the participants' dominant limb, specifically the side used for kicking the ball (24).

Pre-task ultrasonographic image acquisition

Participants arrived at the research laboratory for each of the three data collection sessions at the same time of the day. Participants wore athletic shoes and were positioned on the treatment table in a supine position, with both knees fully extended for 30 minutes to relieve pressure from the cartilage (24). Following a 30-minute rest period, we captured pre-task images of the femoral articular cartilage on both knees (Pre). The procedure for capturing US images of the participants' femoral articular cartilage was conducted as follows: The top of the head was aligned with the edge of the table, and both knees were flexed at approximately 140°, utilizing a handheld goniometer (24) (25) (92). A tape measure was secured to the treatment table to ensure consistency in positioning across all sessions, and we measured the distance from the end of the athletic shoes to the posterior portion (25). A single investigator (M.H.) took all femoral cartilage US images in the dominant leg (baseline). A US unit (AIXPLORER; Super Sonic Imagine, Aix en Province, France) with a 10-MHz linear probe was used to obtain US images of the anterior femoral cartilage for this study. All system settings of the US unit were consistent in all sessions and participants. The linear probe was placed anteriorly over the distal femoral cartilage of the medial and lateral femoral condyles in the axial position and above the superior margin of the patella and oriented to obtain the maximum reflection of the femoral trochlea and the overlying hyaline cartilage as a previous study reported (24) (25) (63) (92). A transparency grid was placed over the US monitor for image reproducibility (24) (25) (26). Three images on both knees were recorded at each US assessment, with the linear probe being removed and repositioned on the knee between recorded images (24) (92).

Post-task ultrasonographic image acquisition

Immediately after completing the 30-minute tasks, participants were supine on the treatment table. We captured the femoral cartilage and recorded three post-task US images (Post 0) in the same procedure already described. Participants rested for 60 minutes with both knees fully extended to relieve pressure on the cartilage for subsequent assessments. We captured the distal femoral cartilage and obtained the US images at 60 min (Post 60) following each condition. All post-task US images were acquired within 3 minutes from the initiation of the US assessment.

Loading conditions

Following the pre-loading US assessment, participants completed 30-minute running, cutting, and control conditions described and shown in Figure 1. All loading and unloading conditions were performed on the same floor in the research laboratory.

Cutting condition. Participants undertook cutting tasks on a 10 x 10 m L-shaped course within the research laboratory (Figure 6-2). The cutting task involved a 10-meter forward sprint, followed by a rapid 90-degree pivot on the dominant foot, transitioning onto the opposite foot, and then resuming the sprint. They were instructed to swiftly change their direction by 90 degrees. In total, participants completed 200 trials, covering a cumulative distance of 4 km for 30 minutes. To control sprinting speed, participants listened to sound signals guiding them to maintain a pace of 9 seconds per lap, roughly equivalent to a speed of approximately 8 km/h. The pace was monitored using the photoelectric cell system.

Running condition. Participants ran a 40.34 m circuit within the research laboratory for a total of 100 repetitions, covering approximately 4 km (23). Participants initiated the run with both feet positioned behind the start line (Figure 6-3). Participants commenced running upon hearing a sound signal and maintained a speed of 8 km/h speed for 30 minutes on the track. To regulate the pace, sound signals were provided every 18 seconds per lap, ensuring the consistent running speed. The timing and repetitions were monitored and recorded using a photoelectric cell system (Witty, Microgate, Bolzano, Italy). This system was installed at a height of 1.2 m above the floor, positioned 2 m apart and facing each other on either side of the starting line. Timing commenced as participants crossed the photoelectric cell system and ceased as they crossed the plane of the sensors.

Control condition. During the control condition, participants lay on the treatment table supine with both knees in fully extended allowing for unloading of the femoral articular cartilage for a duration of 30 minutes.

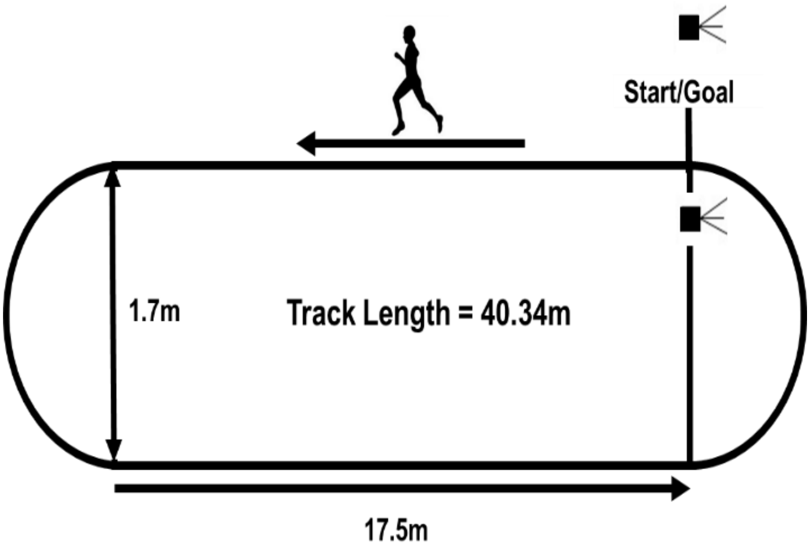

Figure 6-2. Experimental set up for running task

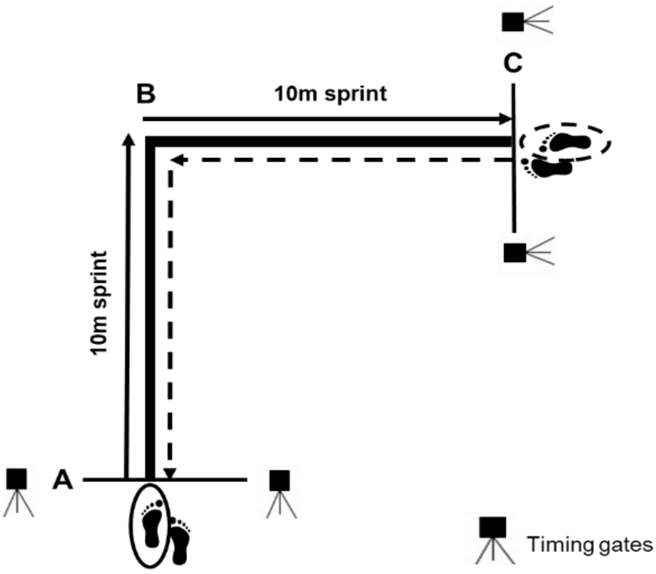

Figure 6-3. Experimental set up for cutting task

Ultrasonographic image processing

The single investigator (M.H.) manually segmented the femoral cartilage of each US image (26) using OsiriX software (Pixmeo SARL, CH-1233 Bernex, Switzerland) (Figure 3-2). These segmented US images were then exported to a custom MATLAB program (version 9.10, The Math Works, Natick, MA, USA) designed to automatically calculate the mean cartilage thickness and CSA in a standardized cartilage region, following the steps outlined in a previous study (26). Initially, the program identified the central point of the US image. It then loaded the manually segmented cartilage region images from the US image and positioned the central point at 50% of the total width of the cartilage region within the overall US image. The program proceeded to divide the entire cartilage cross-sectional area into distinct regions: medial, intercondylar, and lateral. The intercondylar region was defined as the middle 25% of the cartilage region, constituting 12.5% of the length from the center to the central point. The medial and lateral regions encompassed the areas on either side of the intercondylar region, with each region's cross-sectional area being calculated (Figure 3-3). The mean femoral cartilage thickness was determined by dividing the cross-sectional area by the length of the cartilage-bone interface for each respective area (Figure 3-3) (Appendix 1). The previous study reported that US examination of the distal femoral cartilage variables has been demonstrated to be valid (63) (168) and reliable (ICC = 0.83-0.99) (24). Further, prior to data collection for the current study, the US examiner established good to excellent intratester reliability ($ICC_{1,3}$ = 0.86-0.99, SEM = 0.06-0.12, MDC = 0.06-0.21).

Statistical Analysis

Statistical analyses were performed with SPSS 27.0 (SPSS, Inc. Chicago, IL, USA). A separate repeated measures ANOVA was used to compare pre-loading femoral cartilage thickness between each condition. Separate 3 × 3 (condition × time) repeated measure ANOVAs were utilized to analyze each femoral cartilage thickness. In the case of the statistically significant main effect and interactions, a Bonferroni post-hoc pairwise comparison was conducted to identify the location of significant differences. Using the mean and pooled standard deviations, Cohen's d effect sizes and 95% CIs were calculated to assess the magnitude of differences in each femoral cartilage measurement between independent valuables for each pairwise comparison. The strength of effect sizes was interpreted as weak ($d < 0.40$), moderate ($0.40 \leq d < 0.80$), or strong ($d \geq 0.80$) (105). A priori alpha level was set at $p < 0.05$ for all statistical tests. Equivalence was tested using the difference in mean values and 95% CIs for femoral articular cartilage morphology between running and cutting conditions calculated with a paired t-test. It was assumed that the groups were equivalent if the difference in means between Group A and Group B did not exceed x. If the mean difference between groups fell within the CI margin, they were considered equivalent.

6-3. Results

Twenty-five participants completed all three-time points of femoral cartilage US assessments for all conditions. The anthropometric characteristics information on this study is provided in Table 6-1. There were no significant condition main effects for pre-loading femoral cartilage thickness ($p = 0.84$, $\eta^2 = 0.01$, ß = 0.07) (Table 6-2). Mean and standard deviations of each femoral cartilage thickness are shown in Table 6-2.

Table 6-1. Participant Demographics

	Mean (SD)
Sex (Male/Female)	19/6
Age (yrs.)	22.40 (2.61)
Height (cm)	169.22 (8.77)
Body Mass (kg)	62.59 (9.78)
Body Mass Index (kg/m^2)	21.79 (2.59)
Body Fat Percentage (%)	17.28 (6.31)
Dominant Leg (Rt/Lt)	17/8

SD: Standard Deviation

Table 6-2. Femoral articular cartilage variable at pre and post-loading conditions

Compartment	Condition	Pre	Post 0	Post 60	Main effect/Interaction	p-value	Effect size (η^2)	Power (β)
Lateral (mm)	Running	2.40 ± 0.31	2.37 ± 0.34	2.34 ± 0.31	Time	0.49	0.03	0.17
	Cutting	2.41 ± 0.35	2.37 ± 0.30	2.40 ± 0.33	Condition	0.46	0.03	0.18
	Control	2.37 ± 0.35	2.36 ± 0.35	2.39 ± 0.35	Time * Condition	0.49	0.04	0.27
Intercondylar (mm)	Running	2.46 ± 0.40	2.58 ± 0.49[§¶]	2.43 ± 0.38[§^]	Time	0.14	0.08	0.40
	Cutting	2.46 ± 0.37[*†]	2.62 ± 0.43[*#]	2.54 ± 0.42[†^]	Condition	0.01[‡]	0.19	0.83
	Control	2.48 ± 0.43	2.42 ± 0.39[§¶#]	2.49 ± 0.42[§]	Time * Condition	0.01[‡]	0.16	0.94
Medial (mm)	Running	2.10 ± 0.26	2.12 ± 0.28	2.02 ± 0.24	Time	0.33	0.05	0.24
	Cutting	2.08 ± 0.31[†]	2.07 ± 0.27	2.12 ± 0.30[†]	Condition	0.89	0.01	0.07
	Control	2.12 ± 0.29	2.06 ± 0.27	2.06 ± 0.27	Time * Condition	0.08	0.08	0.63

[*]Significant differences between Pre and Post 0 ($p<0.05$).

[†]Significant differences between Pre and Post 60 ($p<0.05$).

[§]Significant differences between Post 0 and Post 60 ($p<0.05$).

[¶]Significant differences between the running and control conditions ($p<0.05$).

[#]Significant differences between the cutting and control conditions ($p<0.05$).

[^]Significant differences between the cutting and running conditions ($p<0.05$).

[‡]Significant interaction and main effect were observed ($p < 0.05$).

Intercondylar cartilage thickness

Significant condition by time interaction ($p = 0.002$, $\eta^2 = 0.16$, ß = 0.94) and condition main effect ($p = 0.007$, $\eta^2 = 0.19$, ß = 0.83) were observed for intercondylar cartilage thickness (Table 2). Intercondylar cartilage thickness at Post 0 were greater in the cutting condition compared to the control ($p < 0.001$, d = 049, 95% CIs: 0.10, 0.30) and the running compared to the control condition ($p = 0.01$, d = -0.36, 95% CIs: -0.26, -0.05), and the difference approached statistical significance. At Post 60, the cutting condition exhibited a greater intercondylar thickness compared to the running condition ($p = 0.01$, d = 0.27, 95% CIs: 0.03, 0.21). There were significant different in intercondylar thickness at the cutting condition between Pre and Post 0 ($p = 0.01$, d = 0.40, 95% CIs: -0.28, -0.04) and Post 0 and Post 60 ($p = 0.04$, d = 0.19, 95 %CIs: 0.004, 0.15), the running condition had a significant difference in intercondylar thickness between Post 0 and Post 60 ($p = 0.01$, d = 0.34, 95% CIs: 0.04, 0.27). No significant time main effect was not observed for intercondylar thickness ($p = 0.14$, $\eta^2 = 0.08$, ß = 0.40).

Medial cartilage thickness

The cutting condition had a greater medial cartilage thickness at Post 60 compared to the running ($p = 0.03$, d = -0.36, 95% CIs: 0.01, 0.18). There were significant different in medial thickness at the cutting condition between Pre and Post 60 ($p = 0.02$, d = -0.20, 95% CIs: 0.01, 014).

Lateral cartilage thickness

There were no significant time or condition main effects, or time by condition interactions for lateral cartilage thickness ($p > 0.05$, Table 2).

6-4. Discussion

The mechanical load applied to the knee joint during physical activities plays a crucial role in preserving and improving knee joint health. However, certain activities, including acceleration, deceleration, and COD, may have a negative impact on cartilage structures (4) (22) (167). While previous studies have observed acute changes in femoral cartilage immediately following high- and low-impact tasks (23) (26), no investigation has definitively identified whether activities involving deceleration and COD influence morphological changes in femoral cartilage. In this study, we investigated how femoral articular cartilage responds to different physical tasks during running, cutting, and control conditions. Our primary findings are that while no significant difference in pre-loading intercondylar femoral articular thickness existed among the conditions, the cutting and running conditions had greater thickness of the intercondylar femoral articular cartilage immediately following each task compared to the control. The intercondylar femoral articular cartilage increase thickness after 30-minute of cutting condition; however, no difference between pre- and post-loading intercondylar femoral articular cartilage thickness was observed in the running condition. In the cutting condition, the intercondylar femoral articular cartilage thickness was still significantly greater at the pre-loading time point compared to 60 minutes post-loading time point. A previous study reported that the femoral articular cartage thickness decreased following 30 minutes of walking and running conditions (24). Taken together, findings from this current and previous studies indicate physical activities have an acute impact on the femoral articular cartilage thickness immediately following the conditions.

Previous research has examined changes in cartilage thickness before and after walking or running, and significant changes were observed in the thickness of the medial femoral articular cartilage of the knee joint (24) (25). Unlike running and walking, cutting conditions showed changes in the intercondylar femoral articular cartilage, suggesting that different areas of joint cartilage in may be affected. According to Havens and Sigward (169), the cutting movement consists of two phases: a deceleration phase and a redirection phase for turning 90-degree. During the deceleration phase, the knee joint is subject to extensor moment and backward ground reaction force as it prepares for the change of direction (169). During the redirection phase, the knee joint experiences abduction and internal rotation moments (79) and the hip joint also experiences abduction moments at initial contact of 90-degree cutting movement (169). In this study, the directional change involved stepping with the outside leg to perform a 90-degree redirection. Additionally, it has been shown that participants' trunk lean angle also moves toward the direction of movement during the stepping phase of this directional change (169).

There is a possibility that the load position on the knee joint shifted from the medial to the intercondylar compartment of the femoral articular cartilage due to the combined involvement of the abduction moment of the knee and hip joints generated by the directional change movement, as well as the stepping with one leg and movement of the upper trunk. Nevertheless, the observed differences in cartilage thickness before and after the cutting movements, unlike running conducted over the same distance and time, suggest that cutting movements may have a significant impact on knee joint cartilage compared to running.

In this study, the intercondylar femoral articular cartilage thickened after the cutting condition. An increase in cartilage thickness following cutting conditions can be explained by swelling of the cartilage that is created by an influx of water attributed to cartilage extracellular matrix break-down (ie, proteoglycan depletion or collagen network disorganization). Swelling of the cartilage may also lead to lower US image quality, which could potentially manifest as the blurring of the cartilage margin in the US and create an increase in cartilage thickness (154). Cartilage swelling occurs due to undesirable osmotic pressure, resulting from decreased levels of glycosaminoglycans (GAGs) within the joint cells, particularly in the middle and deep zones of the cartilage (170). It is hypothesized that the reduction in GAG levels is caused by repeated loading towards the intercondylar compartment of the femoral articular cartilage. Maroudas (170) reported that the local GAGs content is minimally influenced by the water content in healthy cartilage. The findings of this study suggest that even short-term repeated cutting conditions may lead to an increase in cartilage thickness. Hence, it is crucial to recognize swelling as one of the initial symptoms of OA, where collagen fatigue and breakdown may represent the earliest stages of the destructive onset of OA (170). Cutting conditions have been observed to thicken cartilage even over short durations, examining changes in cartilage thickness resulting from prolonged exercise would provide crucial insights for maintaining cartilage health.

This current study observed no significant morphological differences in femoral articular cartilage between the cutting and running conditions. This result may be attributed to intercondylar femoral articular cartilage between the method of running condition. In this study, running was conducted by setting up a short track in the laboratory, allowing participants to perform both running and cutting conditions on the same surface. While running on the short track, rapid cyclic forces were exerted on the articular cartilage, similar to the quick torsional loads experienced during cutting condition. Rapid loading, compared to gradual loading, is associated with the rupture of the articular cartilage surface (117). Participants may have employed similar knee kinematics during cutting and running conditions, to respond to higher-magnitude loading. The comparable mechanics during these two tasks may have exposed the anterior femoral cartilage to both axial loading, occurring during foot contact with the floor, and compressive loading created by tension in the quadriceps mechanism. As the extent of cartilage macrostructure can be interpreted as a measure of cartilage function, these results suggest the potential for evaluating cartilage health via cutting condition that is feasible in knee joint injury populations that have diminished quadriceps function and limited ability of higher-magnitude loading during cutting condition. Furthermore, both cutting and running conditions were executed with the same exercise duration, running velocity, and working surface. Previous study that examined changes in medial femoral articular cartilage thickness before and after walking and running with the same exercise duration and surface found no differences in cartilage thickness changes due to the type of exercise (24). As the loading response of cartilage is dependent on both magnitude and duration of loading (171), the similar cartilage thickness observed following cutting and running conditions may be due to a similar load per unit distance across the two tasks. Considering this, it is suggested that the type of exercise and the speed and intensity of the loads on the femoral articular cartilage may influence the morphology of the intercondylar femoral articular cartilage.

The significant difference was observed immediately after running condition and 60 minutes following running instead of there was no significant change in intercondylar articular cartilage thickness before and after running, The value of cartilage thickness 60 minutes after running was smaller compared to the pre-loading time point. Articular cartilage possesses resilience (69), and when the load on articular cartilage is removed, it tends to return to its original state (66) (69). A previous study examined changes in femoral cartilage thickness before and after drop-landing and walking. It found that drop-landing, which caused a greater change in femoral articular cartilage thickness immediately after exercise, had a slower recovery rate 45 minutes post-exercise compared to walking (25). This current study and previous research suggest that the speed of cartilage recovery is proportional to the magnitude of the load applied to the cartilage, suggesting that the mechanical stimulation from running is lower compared to cutting condition. Verifying the changes and recovery of femoral articular cartilage thickness due to exercise is crucial for maintaining cartilage health.

Previous research comparing cartilage thickness has indicated that the medial and lateral femoral articular cartilage tends to differ after engaging in physical activities such as walking, running, and drop-landing (23) (24). In contrast, our present study observed an increase in the thickness of the intercondylar femoral articular cartilage following both running and cutting conditions, with no significant differences noted in the medial and lateral femoral articular cartilage. We hypothesized that the notable change in thickness of the intercondylar femoral articular cartilage could be attributed to the consistent loading experienced during cutting and running conditions. While running condition, vertical compressive forces and sagittal plane shear forces are applied to the femoral articular cartilage (172) continually. During cutting condition, transverse plane forces impact the intercondylar articular cartilage region, which serves as the pivot point of the knee joint. The intercondylar cartilage thickens due to continuous compressive forces during cutting and running conditions, leading to a more pronounced increase in load on the intercondylar femoral articular cartilage compared to the medial and lateral compartments, resulting in significant thickening of the intercondylar femoral articular cartilage. Additionally, in experiments examining cutting conditions, typically, only a few trials are conducted (79). Unlike the previous study, this study involved performing repeated COD movements with consistent timing for 30 minutes. It is inferred that the increased load on the intercondylar femoral articular cartilage over time caused the cartilage to thicken.

As a result of this study, it should be noted that the rate of cartilage thickness change in the control group was greater than the pre-value at lateral and intercondylar thickness. A previous study that compared the femoral cartilage thickness of the control group after 30 minutes of walking and running reported that the rate of cartilage change in the control group was lower than that of the exercise group (26). A previous study introduced 60 minutes of cartilage unloading before imaging to establish a baseline femoral cartilage thickness (26). This study also documented a lower femoral cartilage thickness change rate in the control group compared to the 30-minute walking and drop-landing groups (26). Unlike the results of the two previous studies, this present study indicated that the rate of medial compartment cartilage thickness change in the control group was greater compared to the exercise group. For this study, participants were requested to use same transportation, such as walking or taking a bus, for each measurement session. However, considering that changes in cartilage thickness were still observed even after 30-minute rest period, it has been indicated that future considerations should not only focus on the rest duration but also the method of arrival to the facility.

6-5. Summary

Previous research on cutting techniques has primarily focused on enhancing athletic performance and preventing sports injuries. However, there has been no research exploring the relationship between cutting techniques and changes in the structure of knee cartilage. The result of this study demonstrated that cutting conditions may exert a greater influence on acute intercondylar femoral articular cartilage deformation. This indicates that cutting conditions can have a greater impact on morphological changes in articular cartilage compared to running. Regular physical activity is crucial for extending healthy life expectancy. It is difficult to engage in any exercise or sport that does not involve cutting operations. Therefore, it will be important in the future to investigate the relationship between movement patterns and joint health, to recommend safe, low-impact activities that minimize stress on the femoral articular cartilage.

CHAPTER VII

General Discussion

7-1. Summary of the main finding

The primary purpose of this dissertation was to investigate the attributes and influence of rugby participation on the morphology of femoral articular cartilage, laying the groundwork for developing innovative approaches to preserve joint health in collegiate rugby players. In this dissertation, we set out four specific aims.

First, the morphological characteristics of femoral articular cartilage were examined between collegiate rugby players, who have a high prevalence of knee OA, and T&F athletes, who have a lower prevalence (Chapter 3). Rugby players exhibited greater intercondylar femoral articular cartilage thickness and larger CSA in the lateral and middle compartments compared to T&F athletes. However, when adjusted for body mass, there were no significant differences in the femoral articular cartilage thickness and CSA between the two groups. These findings suggest that body mass may have a greater influence on cartilage morphology than the type of sport activity.

The next study aimed to examine the characteristics of femoral articular cartilage morphology in collegiate rugby players with and without a history of knee joint injuries (Chapter 4). The main findings were that collegiate rugby players with a history of intracapsular knee joint injury had greater lateral condylar and intercondylar thickness, as well as larger CSA, compared to those without a knee joint injury. Traumatic intracapsular knee joint injury may alter the macrostructure of femoral articular cartilage in collegiate rugby players, potentially affecting long-term joint and cartilage health.

Next, the study observed changes in the femoral articular cartilage morphology of collegiate rugby players over two competitive seasons to understand how participation in rugby competitions affects cartilage morphology (Chapter 5). Participation over two consecutive season led to a decrease in femoral articular cartilage thickness and CSA, regardless of a previous history of knee joint injury. This result suggests that participating in a competitive rugby season significantly impacts changes in femoral articular cartilage thickness and CSA among collegiate rugby players.

Finally, we investigated the influence of running and cutting conditions, both of which are performed in rugby, on femoral cartilage (Chapter 6). The femoral articular cartilage thickness increased immediately after both running and cutting conditions compared to control group, but there were no significant differences in cartilage thickness between the running and cutting conditions. In the cutting condition, femoral articular cartilage thickness immediately after exercise was significantly higher compared to the pre-exercise value. Sixty minutes after the exercise, the femoral articular cartilage thickness was lower than immediately after the exercise but remained higher compared to the pre-exercise value. In running condition, there were no significant differences in femoral articular cartilage thickness values between pre- and post-exercise, and 60 minutes after running ceased, femoral articular cartilage thickness returned to its pre-exercise value. This result suggested that cutting condition has a greater influence on changes in femoral articular cartilage morphology compared to running.

7-2. Analysis of femoral articular cartilage morphology in collegiate rugby players

The key to maintaining and improving joint health is engaging regular physical activity. The load on the femoral articular cartilage depends on the type of exercise, its intensity, and its duration. Articular cartilage changes morphology in response to these factors, adapting the mechanical stresses placed on the joint (99) (100). Additionally, participating in competitive sports places varying loads on the joints based on the characteristics of the sport and the level of competition. Certain sports activities carry a greater risk to developing knee OA (3). Therefore, understanding movement and exercise intensity based on the specific characteristics of the sport is crucial for developing strategies to maintain cartilage health.

7-2-1. Comparison of femoral articular cartilage morphology between rugby players and track and field athletes

We investigated the influence of participation of rugby on the femoral articular cartilage morphology of collegiate rugby players, comparing them with T&F athletes, who have different physical task characteristics and knee OA prevalence. The collegiate rugby player had thicker femoral articular cartilage compared to T&F athletes; however, these difference ware not observed when controlling for body mass (Chapter 3).

As stated in Chapter 6, the femoral articular cartilage thickened after 30 minutes of running and cutting conditions. The changes in femoral articular cartilage thickness before and after the cutting condition ware significantly greater compared to the running condition. While significant changes in cartilage thickness were observed after the cutting condition, no significant differences were noted in the running condition. A study that examined the transient changes in cartilage thickness due to walking and running conditions also found that the changes in femoral articular cartilage thickness before and after the drop-landing condition showed a significant difference compared to the changes before and after the walking condition (24).

Cartilage morphology changes depending on the magnitude and type of load applied, with areas subjected to greater loads becoming thicker to withstand the stress (56). This suggesting that the differences in load applied by different types and magnitude of exercise have varying effects on the cartilage. A previous study reported that the compressive load on the knee joint during walking increases by approximately 4 kg for every 1 kg increase in body mass (173). The exercises performed during competition involve higher-intensity movements compared to walking, so the stress on knee joint cartilage due to increased body weight also proportionally increases. Since rugby players are heavier than T&F athletes and perform movements that place a greater load on their joint cartilage, it is believed that the femoral articular cartilage of heavier rugby players becomes thicker compared to that of T&F athletes.

The imaging period of this study was set for the latter half of the off-season to minimize the impact of the competition season. Consequently, measurements had to be conducted within a limited time frame, but this period was considered appropriate to eliminate the influence of recent competitive activity on femoral articular cartilage morphology. However, due to the small sample size, it was difficult to categorize players based on the presence or absence of a history of knee joint injuries and years of competition experience. Therefore, the subjects of this study were selected as players without knee joint injuries in the year before measurement, but the influence of knee joint injuries could not be eliminated. As a result, the sport and a history of joint injuries may have had some impact. To further investigate the characteristics of femoral articular cartilage morphology by different sports, it is important to increase the number of sports being compared, examine the differences in cartilage morphology among athletes from various sports, and verify the relationship with future OA incidence rates.

7-2-2. Femoral articular cartilage morphology and knee injury history in collegiate rugby players

Primary prevention and reduction of the risk of knee OA involves preventing joint injuries in competitive athletes (152) (174). Knee joint injury is a high-risk factor for the onset of knee PTOA (75). Individuals with a previous history of knee joint injury in youth have a higher incidence of PTOA compared to those without a joint injury (29).

In rugby, elite-level players have a higher injury incidence rate and greater injury severity compared to amateur rugby players and non-contact sports (43) (44). Additionally, the number of reported surgeries is higher for rugby players than for non-contact sports athletes (43). Knee joint injuries are common during practices and games (149) (150), and the severity of those injuries is higher compared to injuries at other body sites (149) (151). Injuries frequently occur during tackle situations (149), and the incidence rate of injuries for tacklers is higher compared to ball carriers (150). The prevalence of OA is higher in athletes with a history of joint injuries compared to those without such a history (20) (43). A study on former international rugby players found that 61.5% had developed OA, and 27% had undergone total joint replacement. Among these cases, knee OA had a higher incidence rate compared to hip OA, with rates of 38.5% and 19.6% respectively (20).

This study examined changes in femoral articular cartilage morphology in collegiate rugby players with a history of intracapsular knee joint injuries compared to those with no history of knee injuries (Chapter 4). To eliminate the influence of other knee joint injuries, we specifically compared and analyzed players with a history of intracapsular injuries to those with no knee joint injury. The results revealed that collegiate rugby players with a history of intracapsular knee injury had greater lateral condylar and intercondylar femoral articular cartilage thickness and larger CSA compared to uninjured rugby players. An increase in femoral articular cartilage thickness has also been observed in individuals with a history of ACL injury (95) (116) (134) (142). An increase in cartilage thickness following knee joint injury can be explained by swelling of the cartilage that is created by an influx of water attributed to cartilage extracellular matrix break-down, such as damaged and degeneration of the collagen network (170), decreased matrix proteoglycan and cellular loss. At the time of the initial ACL injury, acute chondral injury occurs in the knee articular cartilage at the lateral tibial plateau and lateral femoral condyle (175). The degeneration of cartilage at the lateral compartment occurs due to the shift of load to the lateral side of the joint after an ACL injury (175). Therefore, it is important to examine the changes in cartilage at the lateral compartment as well as the medial compartment. However, Calvo et al. (165) reported that this cellular loss was due to tissue damage, not trauma. This study suggested that changes in joint structure due to trauma, along with subsequent joint instability and altered functional movement, modified the structure of the femoral articular cartilage. Damage to the ligament, especially ACL and meniscus significantly impact knee joint biomechanics have the risk of developing PTOA (48). Injuries to the knee joint produce high levels of mechanical loading across the joint (70). Injury-induced changes in joint structure and function alter the location and magnitude of the load on the articular cartilage. Abnormal loading on cartilage areas that were previously non-weight-bearing before the injury accelerates cartilage degeneration, increasing the risk of developing OA (56). Prolonged exposure to such excessive loads may result in negative degeneration of the cartilage tissue and adversely affect the structural integrity of the cartilage (73) (128) (130) (131) (176) (177).

Considering that knee joint instability is a factor in the risk of developing PTOA, it is necessary to investigate changes in femoral articular cartilage morphology due to both intracapsular and extracapsular knee joint injuries. Previous studies on professional rugby players indicated that medial collateral ligament (MCL) injuries had the highest incidence rate among knee joint injuries (43) (44) (151), compared to ACL injuries and chondral and meniscal injuries (151). Unlike other ligamentous injuries, MCL injuries do not typically require surgical treatments such as reconstruction (151). Therefore, examining the influence of MCL injuries on femoral articular cartilage morphology can provide insights into the relationship between extracapsular injuries and the risk of OA. Additionally, MCL injuries often occur alongside ACL and meniscal injuries, commonly known as the "unhappy triad", making it essential to investigate not only the effects of isolated MCL injuries but also the impacts of multiple injuries and re-injuries on femoral cartilage morphology. This comprehensive examination can deepen our understanding of the correlation between knee injury and the risk of developing knee OA. Consequently, this is proposed as a subject for future research.

7-2-3. Changes in femoral articular cartilage morphology due to competitive rugby seasons

No significant differences were observed in the femoral articular cartilage morphology characteristics between rugby players and T&F athletes. We investigated how participation in competitive season affects the morphology of the femoral articular cartilage in collegiate rugby players over two years. After two consecutive competitive rugby seasons, the femoral articular cartilage thickness and CSA of collegiate rugby players decreased compared to before the competition (Chapter 5). Changes in femoral articular cartilage thickness and structure due to sports participation have also been reported in volleyball (102) and basketball (18), indicating that participation in any sport can affect cartilage morphology.

Rugby matches and practices require players to engage in diverse and repeated high-intensity activities, including sudden CODs (111), rapid accelerations (111), sprints (112), and tackling (109). Even during less intense periods, rugby players move around the field by walking or jogging (109). Studies examining changes in femoral articular cartilage thickness due to short-duration exercises have shown that changes after cutting conditions, which are repeatedly performed in rugby, were greater compared to running (Chapter 6). Additionally, the time required for the cartilage thickness to recover to its pre-exercise state is longer for cutting conditions than for running. Previous studies investigating acute changes in femoral articular cartilage thickness due to walking and running have observed greater changes in cartilage thickness after running compared to walking (24). These results suggest that transient morphological changes in cartilage due to exercise differ depending on the type of exercise, and that recovery of cartilage morphology after exercise may be proportional to the magnitude of those changes.

During the competitive rugby season, players participated in regular daily practices, each lasting 1.5 to 2 hours, and had 1 to 1.5 hours of strength training sessions four to five times a week, with games once a week. As participating time in rugby increases, the load on femoral articular cartilage also becomes greater. This study did not verify changes in femoral cartilage thickness resulting from the repeated combination of these exercises. It can be inferred that during the competitive rugby season, the femoral articular cartilage of collegiate rugby players did not get enough time to rest and maintain its thickness, the accumulation of load on the femoral articular cartilage can lead to overload, hindering the generation of essential components needed to maintain cartilage structure (49), which may reduce thickness and overall cartilage health. To minimize changes in cartilage morphology, it is important to provide sufficient rest periods proportional to the load experienced during training and games. To determine the necessary rest time for rugby players, it is desirable to track changes in femoral articular cartilage morphology throughout the year. This requires measuring femoral articular cartilage morphology in conjunction with the team schedule and closely examining the effects of practices and games.

7-3. Practical application of cartilage health for collegiate rugby players

Daily physical exercise is an important factor in improving healthy life expectancy, and mechanical stimulation from exercise is crucial for maintaining and improving joint health (69). Mechanical stimulation on joint cartilage can induce functional adaptation, resulting in positive changes when appropriate. However, exposure to stimuli beyond the adaptive range can lead to cartilage fatigue and negative changes (5). Participating in competitive sports places a greater load on the cartilage compared to regular exercise. Femoral articular cartilage thickness altered by exercises tend to return to its original state when the load is removed (66) (69). Change in femoral articular cartilage thickness are influenced by the magnitude of load applied to the cartilage (56), and the time it takes to return to its original state is proportional to the load placed on the cartilage (66).

Considering the results that verified change in cartilage thickness due to acute exercises, it is necessary to provide sufficient rest time to restore cartilage thickness to its original state. However, given the high and complex loads experienced by the femoral articular cartilage during practice and game, it is difficult to determine the exact recovery time needed. Therefore, future research should focus on developing recommendations for maintaining athletes' cartilage health, such as devising training structures to reduce the accumulation of cartilage fatigue and providing rest periods to recover from fatigue.

This dissertation has elucidated that participation in rugby competitions induces changes in cartilage morphology (Chapter 4 and 5), and there were no significant differences in femoral articular cartilage morphology due to sports participation (Chapter 3). Furthermore, no differences were observed in the changes of femoral cartilage thickness due to different exercise components (Chapter 6). These findings suggest that regardless of the mechanical load generated by exercise elements such as cutting and running conditions, the frequency and magnitude of load on the femoral articular cartilage during exercise may have an impact on changes in cartilage morphology.

When comparing femoral articular cartilage thickness across different sports, Babaveya et al. (108) did not observed significant differences between sports, and no significant differences were observed in cartilage thickness among marathon runners of different skill levels (106). It is necessary to investigate the hypothesis that physical activity itself influences the morphological changes in the femoral articular cartilage, irrespective of the type of exercise. While this study did not directly examine the magnitude of load on articular cartilage resulting from different sports or exercises, elucidating these relationships could serve as a guide for making informed choices about physical activity and sports participation, considering changes in articular cartilage morphology. This could also lead to important insights for preventing future OA development.

Changes in femoral articular cartilage vary among athletes and are influenced by various factors such as the athlete's physical characteristics (2), the anatomical structure of the knee joint (56) (70), and a previous history of knee injuries (4) (20). General OA risk factors, such as overweight and aging, must also be considered. The femoral articular cartilage thickness of rugby players was greater than that of T&F athletes. However, after adjusting for body mass, no difference was observed, indicating that body mass is a factor influencing femoral articular cartilage morphology. In rugby, a sport involving physical contact, having a heavy body mass is often an advantage, as BMI increases, so does the load on the knee joint cartilage (182). Rugby players tend to be heavier than those playing other sports, making weight management the key to maintaining cartilage health and preventing knee OA (11) (49) (72) (74) (152). Given that rugby is associated with a high risk of developing OA (20) (43), maintaining an appropriate weight for rugby competition is essential to reduce stress on the knee joint and support cartilage health in rugby players.

Aging is also the main risk factor for the onset of OA (48) (178). According to Jones et al. (120) knee joint cartilage is thickest during Tanner stage two and then decreases in thickness in proportion to bone growth thereafter. It can be inferred that the joint cartilage of collegiate athletes, who have already exceeded their growth period, might affected by aging. OA is rare in young adults, but abnormal stress due to injury can lead to an early onset of OA (29). Since rugby involves repeated movements that significantly alter femoral articular cartilage morphology, we cannot overlook the fact that the cartilage's regenerative capacity decreases with age (179).

During the competitive rugby seasons, femoral articular cartilage thickness decreases regardless of the presence or absence of injury. However, players with a history of knee joint injury consistently showed thicker femoral articular cartilage across all measurement periods over both one and two consecutive rugby seasons compared to those without a history of injury (Chapter 4 and 5). This suggests that the cartilage may become chronically thickened due to structural changes. Since cartilage morphology changes in response to the load generated by exercises (158) (180), likely, cartilage morphology changes proportionally to the load extended on it during competition.

Collegiate rugby players with a history of knee joint injury participated in a rehabilitation program aimed at returning to competition, under the supervision of a team physician and a team athletic trainer immediately after their injury. The team rehabilitation program included restore strengthening and ROM and included neuromuscular training (152) (161). The program was not only focused on the recovery of the injured area but also included position-specific routines designed for returning to competitive rugby practices and games. These programs addressed aspects such as contact, muscular power, and the biomechanical demands of each position in rugby (181) (Appendix 2). Additionally, to reduce the risk of reinjury, reinforcement training was conducted after returning to competition as needed, under the supervision of a team athletic trainer and strength training specialists. Implementing such programs to restore knee joint and lower limb function likely helped control abnormal loads on the knee joint and delayed cartilage degeneration.

In this study, all collegiate rugby players with a knee joint injury participated in the team rehabilitation program; therefore, it is difficult to evaluate the specific effects of rehabilitation. However, the decrease in femoral cartilage morphology during the competitive season did not show a significant difference compared to players with and without a history of knee joint injuries. Additionally, no players with a history of knee joint injury sustained a reinjured knee joint during this study period. Future research is needed to examine how participation in rehabilitation and differences in rehabilitation content impact femoral articular cartilage. The result of this study suggests that restoring and improving knee joint function through rehabilitation tailored to the demands of the sport may help delay the degeneration of femoral articular cartilage in collegiate rugby players.

Cartilage thickening is a primary indicator of morphological changes in the femoral articular cartilage that can lead to the development of OA (182). However, it has been reported that in early or mid-stage knee OA, changes in femoral articular cartilage thickness are not unidirectional; the thickness can either increase or decrease. According to Buck et al. (62) two-year observational study, medial tibiofemoral cartilage that progressed to KLG 2 showed approximate proportions of thinning and thickening from the baseline. In contrast, KJG 3 knees exhibited more thinning than thickening of the medial cartilage. Understanding the transitions from thickening cartilage to reductions after injury provides insights into the mechanisms of OA progression. Therefore, longitudinal examination of femoral articular cartilage morphological changes in individuals with a history of knee joint injury is crucial for understanding cartilage health and preventing onset and progression of future OA. We believe that valuating and verifying the qualitative structure using MRI could elucidate the mechanisms of OA onset and progression in the future.

In this dissertation, we used US imaging to conduct a comprehensive examination of morphological changes in the femoral articular cartilage of collegiate rugby players. Rugby is a sport with a high incidence of knee OA (20), and knee injuries, which are risk factors for OA, occur more frequently in rugby compared to other non-contact sports (44). Changes in femoral articular cartilage thickness are one of the symptoms of early OA (60) (61). However, no studies have examined the characteristics of femoral articular cartilage morphology in active rugby players or how playing rugby affects this cartilage morphology. We pursued this research believing that investigating changes in femoral articular cartilage thickness in athletes would provide important insights into cartilage health. MRI is the primary method for evaluating knee joint cartilage (83) (84), US has also become a standard method for assessing the morphological characteristics of femoral articular cartilage (64). The measurement method used in this study employed anatomical landmarks, allowing for relatively easy determination of measurement sites and high reproducibility (63), which enabled repeated measurements over a long period.

Since US measures from the surface, the range of cartilage that can be assessed is limited. Therefore, when evaluating the tibiofemoral joint using US, only the femoral articular cartilage can be measured (64). In this study, the US probe was placed on the distal femoral articular cartilage of the medial and lateral femoral condyles and the superior margin of the patella (24) (26) (63). This method allows for a broad capture of the femoral articular cartilage. The transverse approach can image and evaluate the femoral articular cartilage extensively, focusing on the intercondylar fossa of the femur (63). While this method can comprehensively evaluate the femoral articular cartilage surface, it has limitations in measuring the lateral portion of the medial and lateral femoral condyles. The medial femoral articular cartilage, a weight-bearing area, can be imaged by placing the probe on the medial femoral condyle (64). When a deeper examination, this method allows for imaging with the same transverse approach as in the current study. Since it has been shown that this measurement site includes the weight-bearing area of the medial femoral condyle (24), we consider this measurement method appropriate for the objectives of our study.

7-4. Limitations

This dissertation has limitations that need to be acknowledged. The study focused on collegiate rugby players and included only male athletes as subjects. The collegiate rugby team targeted in this study included several female players who trained alongside the male players in most non-contact practices. Female rugby players have a higher incidence rate of knee injuries, including ACL injuries, than males (183). Additionally, women have a higher incidence of knee OA compared to men (48), highlighting the importance of establishing preventive measures for OA. However, there are fewer studies on female rugby players than males, indicating a need for further research to gain new insights.

Another limitation of this dissertation, a single examiner conducted all tasks, from collecting US data to assessing femoral articular cartilage. All data were anonymized to prevent individual identification, and maintaining blinding throughout the study was another challenge. Future studies should include measurements and investigations across multiple teams at the same competition level to facilitate a more comprehensive analysis.

7-5. Future perspectives

For further advancement in research, the future perspectives are outlined below. US imaging was used to examine changes in the femoral cartilage morphology of collegiate rugby players. By using a portable US device and selecting a highly reproducible imaging method for our measurements, we were able to track changes in the cartilage morphology of rugby players over two years. The reliability (26) and validity (24) (25) (35) (108) of measuring cartilage morphology using US imaging has been demonstrated in many previous studies. However, qualitative assessment using US has not been performed throughout the study. Qualitative evaluation of cartilage using US images was performed using echo intensity (89). Articular cartilage withstands the load applied to the joint by allowing the water contained within to flow out (69). Since echo intensity is influenced by the body's water content and the water contained in the cartilage (145), it is possible that the water expelled from the cartilage in response to the load on the joint could affect the results of echo intensity. Structural changes in articular cartilage occurred before morphological changes, so examining qualitative changes along with morphological changes may further clarify the effects of exercise on cartilage structure. However, to quantitatively examine changes using echo intensity, it is necessary to validate the measurement methods. Therefore, alongside US measurements, it is also essential to consider conducting morphological and qualitative evaluations of the femoral articular cartilage using MRI.

Measuring cartilage thickness after the initial assessment, regardless of continued participation in rugby, will provide valuable data for verifying changes in femoral articular cartilage morphology during adulthood, even after 5 or 10 years have passed. Also, it proposes examining changes in the femoral articular cartilage in athletes after concluding competitive sports. This can lead to an understanding of how mechanical stimuli on the femoral articular cartilage from competitive sports might influence it in the future. Understanding the longitudinal changes in morphological aspects of the femoral cartilage that were not observed during competition can be instrumental in determining the timing of future OA development.

Further investigation is needed regarding the changes in cartilage morphology of collegiate rugby players during the off-season. In a two-year longitudinal study, we observed changes in cartilage thickness between two competitive seasons (Chapter 5), it was not possible to confirm recovery of cartilage thickness. This may because including the preseason in the off-season may have hindered the confirmation of cartilage thickness recovery. Between the two competitive seasons, collegiate rugby players participate in fundamental and sports-specific training. Additionally, collegiate rugby players compete in spring matches. A previous study comparing the femoral articular cartilage structure of collegiate basketball players during the competitive season and the subsequent off-season reported that T2 and T1rho relaxation times significantly decreased before and after the competitive season (18). Additionally, the relaxation times of the femoral articular cartilage increased after the competitive season and the off-season, approaching the pre-season values (18). This result suggests that spending the off-season, a period without practice or games, may help restore the femoral articular cartilage structure in collegiate athletes. Reviewing the measurement periods and re-examining how the off-season affects the femoral articular cartilage morphology of collegiate rugby players is a future research task.

CHAPTER VIII

Conclusions

Regular physical exercise is an important factor in improving cartilage health. However, the specific types of exercise that best maintain and improve cartilage health remain unclear. Previous research has shown that certain sports can increase the future incidence rate of OA; however, it is not clear how participation in sports affects athletes' cartilage morphology. Therefore, the objects of this dissertation were to clarify the effects of exercise and sports participation on the femoral articular cartilage and to verify the morphological changes in the femoral articular cartilage of collegiate rugby players using US imaging.

This dissertation followed the morphological changes in the femoral articular cartilage of collegiate rugby players over two years and revealed that the cartilage morphology showed lower values due to participation in the sport, regardless of the presence or absence of a history of knee joint injuries. It is challenging to determine whether the femoral articular cartilage morphological changes observed in collegiate rugby players will lead to chronic changes based on a two-year follow-up. However, demonstrating that continued participation in rugby results in a decrease in femoral articular cartilage can contribute to research on cartilage health in collegiate athletes.

Participation in rugby competitions alters knee joint cartilage morphology, but maintaining physical function suitable for the sport suggests the potential to control the cartilage health in the knee joint. Additionally, since changes in femoral articular cartilage morphology are early signs of knee OA, observing these changes in cartilage morphology can serve as a tool to assess cartilage health in athletes and verify the relationship between exercise and knee OA risk.

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Appendix 1. The program code of MATLAB for calculating the femoral articular cartilage morphology

```
%% -------- Preprocessing for analysis -------- %%

% Import the analyzed ultrasound images into the MATLABA program.
image = dicomread('US_file.dcm');

% Extract the red line drawn in the ultrasound image.
[BW] = createMask(image);

% Fill in the red frame with white.
ClippingBW = imfill(BW,'holes');

%% -------- Detecting the left and right edges and determining the central point of the image -----
--- %%

% Obtain the number of pixels arranged vertically.
vertical_pixel = zeros(1, size(BW, 2));
for num = 1:1:size(BW, 2)
    vertical_pixel(num) = sum(BW(:, num));
end

% Detect the left and right edges of the image.
Left_edge = find(vertical_pixel(1, 1:round(num/2)) == max(vertical_pixel(1:round(num/2))));
Right_edge = find(vertical_pixel(1, round(num/2):end) ==
max(vertical_pixel(round(num/2):end))) + round(num/2);

% Align left and right edges to the image.
ClippingBW = ClippingBW(:, Left_edge(1):Right_edge(end));

% Detect the center point of the image.
x = round(size(ClippingBW, 2) / 2);

% Trimming 12.5% from the left and right sides of the central point.
WidthRatio = 12.5 / 100;
```

```
% Calculate the width of the entire image and determine 12.5% to the left and right from the center of the image.
Width = size(ClippingBW, 2);
Width_ratio = round(Width * WidthRatio);
Range_Center_L = x - Width_ratio;
Range_Center_R = x + Width_ratio;

%% -------- the calculate the average cross-sectional area (CSA) of the femoral articular cartilage -------- %%

% Obtain the number of pixels of the lateral, Intercondyle, and medial compartments of knee cartilage from the images.
tickness_left = sum(ClippingBW(:, 1:Range_Center_L));
tickness_center = sum(ClippingBW(:, Range_Center_L+1:Range_Center_R-1));
tickness_right = sum(ClippingBW(:, Range_Center_R:end));

% Convert one pixel to millimeters (10mm/references length (pixel): Annotation.1).
pixel_to_mm = 10 / XXX.XX;

% the calculate the average CSA of lateral, intercondyle, and medial compartments.
area_left = sum(tickness_left) * pixel_to_mm * pixel_to_mm;
area_center = sum(tickness_center) * pixel_to_mm * pixel_to_mm;
area_right = sum(tickness_right) * pixel_to_mm * pixel_to_mm;

%% -------- the calculate the average thickness of femoral articular cartilage -------- %%

% Detect the contour of the femoral articular cartilage.
length_picture = edge(ClippingBW);

% Separate the upper and lower ends of the femoral articular cartilage.
Upper = zeros(size(length_picture, 1), size(length_picture, 2));
Lower = zeros(size(length_picture, 1), size(length_picture, 2));

for Num = 1:1:size(length_picture, 2)
    Edge = find(length_picture(:, Num) > 0);
```

```
    Edge_Row = Edge - Edge(1);
    for Num2 = 1:1:size(Edge_Row, 1)
        if ( Edge_Row(Num2) < 20 )
            Upper(Edge(Num2), Num) = 1;
        elseif ( Edge_Row(Num2) >= 20 )
            Lower(Edge(Num2), Num) = 1;
        end
    end
end

% Calculate the length of the lower end of the femoral articular cartilage for each compartments:
lateral, intercondyle, and medial.
length_left = sum(sum(Lower(:, 1:Range_Center_L))) * pixel_to_mm;
length_center = sum(sum(Lower(:, Range_Center_L+1:Range_Center_R-1))) * pixel_to_mm;
length_right = sum(sum(Lower(:, Range_Center_R:end))) * pixel_to_mm;

% calculate the mean thickness of femoral articular cartilage in the lateral, intercondyle, and
medial compartments.（CSA/cartilage length of the lower end）.
mean_tickness_left = area_left / length_left;
mean_tickness_center = area_center / length_center;
mean_tickness_right = area_right / length_right;
```

Annotation 1. The 10mm/references length (pixel) were analyzed using the average number of pixels obtained by drawing ten 10 mm lines on sample images taken with each ultrasound device. In this study, the following number of pixels were used for calculations: 141.16 pixels were used for image analysis captured by a portable US unit (Cahpter3) and 222.91 pixels were used for an US unit (Chapter 6).

Appendix 2 Basic rehabilitation protocol for knee joint injury to return to the team practices

	Level 0	Level 1	Level 2	Level 3	Level 4	Level 5
	Acute	Rehabilitation		Athletic Rehabilitation		Pre-return to practice
Knee joint Rehabilitation	PROM Isometric training Tube training	AROM Tube training Machine training	Body-weight training 1/4 ~ 1/2 Squat Two legs ~ one leg	1/2 ~ Full Squat Lunge: one-way, two-way, multidirectional	Balance training: agility disk, balance ball Plyometrics training: box jump	Sports specific training
Aerobic training	Hand pedal bike Seated upper-body dumbbell circuit training Seated Battle rope training	Stationary bike Walking	Jogging & Running Long distance running Pace-up running	Pace-up running Acceleration/Deceleration Change-of-pace *change running shoes to spike shoes	Sprinting: w/wo turn Sidestep: w/wo turn Change of direction: single ~ multidirectional	Sports specific training Reaction drill
Weight training	Upper-body exercise Core training Non-weight bearing ~	Open to Closed Kinetic training Body weight bearing	Low ~ Medium intensity Restore basic muscle strength	Medium ~ High intensity Single- ~ multi-joint training	No limitation Balance training Plyometrics training	Transition to team training
Contact Skills					With Dummy	With People
Positional Skills				Scrum: Machine only	Scrum: 1vs1 ~ 3vs3 Line-out Kicking the ball	Scrum: 5vs 5 - Full