

Histopathological study in acute rupture of the anterior cruciate ligament of the knee

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ABSTRACT

Introduction: The histological structure of ligaments, including the anterior cruciate ligament involves collagen and elastin wrapped by a matrix of water and proteoglycans. Collagen is the main component of tendons and ligaments—it represents 65-75% of their dry weight. The aim of this study was to carry out a histopathological evaluation of the fragments of broken anterior cruciate ligaments taken at the moment of arthroscopy.

Materials and Methods: Prospective observational study conducted in 50 cases with rupture of the anterior cruciate ligament. We took samples from it during arthroscopy and carried out the histopathological evaluation of the ligament.

Results: We outline histopathological results and postoperative outcomes.

Conclusion: There were histopathological degenerative changes; it cannot be said whether these changes were previous or posterior to the ligament rupture.

Key words: Anterior cruciate ligament; ACL; histopathology.

Level of evidence: IV

ESTUDIO HISTOPATOLÓGICO EN ROTURA AGUDA DEL LIGAMENTO CRUZADO ANTERIOR DE RODILLA

RESUMEN

Introducción: La estructura histológica de los ligamentos, inclusive el ligamento cruzado anterior, está compuesta por colágeno y elastina envueltos en una matriz de agua y proteoglicanos. El colágeno es el componente principal de los tendones y ligamentos, representa el 65-75% de su peso en seco. El objetivo de este estudio es realizar una evaluación histopatológica de los fragmentos de ligamento cruzado anterior rotos, tomados en el momento de la cirugía artroscópica.

Materiales y Métodos: Estudio prospectivo observacional sobre 50 casos con rotura de ligamento cruzado anterior. Se tomaron muestras de ligamento cruzado anterior en el procedimiento artroscópico y se evaluó la histopatología del ligamento.

Resultados: Se mencionan los resultados de la anatomía patológica y la evolución posoperatoria.

Conclusión: Se observaron cambios degenerativos en la histopatología; no puede demostrarse si estos cambios son anteriores o posteriores a la rotura ligamentaria.

Palabras clave: Ligamento cruzado anterior; LCA; histopatología.

Nivel de Evidencia: IV

Conflict of interests: The authors have reported none.

Introduction

The anterior cruciate ligament (ACL) is an important structure which keeps the knee stable.^{1,4} Its rupture is more frequent in sport people and, therefore, it affects healthy persons who want to keep an active lifestyle.⁵⁻¹⁰ In the US nowadays there are more than 200,000 injuries of this kind per year.¹¹

Females are fourfold more prone to ACL injury than males.¹² There are several predisposing factors: hormone variations in women,¹³⁻¹⁵ proprioceptive deficits, anatomic variants such as an excessive tibial slope^{16,17} or narrow intercondylar notch,¹³ and mutations in collagen genes which cause abnormal articular laxity.¹⁸

Due to the frequency and potential seriousness related to these lesions, it is necessary to get thoroughly acquainted with ACL histopathology, realizing when it is injured.

The histological structure of ligaments, including the ACL, involves collagen and elastin fibres wrapped by a matrix of water and proteoglycans. Collagen is the main component of tendons and ligaments, representing 65-75% of their dry weight. Collagen fibres are 150-250 nm in diameter, looping themselves to form a complex web.^{7,19} Several of such fibres get together to form subfascicular units, each one surrounded by a thin band of lax connective tissue—the endotenon.

Danylchuk et al.,¹⁹ using scanning electron microscopy, verified a more complex structure where the straight collagen bundles are formed by a “complex of interweaved fibrils”.

Electronic microscope studies have showed that, between the proximal, middle and distal thirds of the ACL there are differences with respect to the distribution of the collagen fibrils, as well as their diameter.^{7,20}

The aim of this study is to make a histopathological evaluation of the fragments of broken ACLs taken at the time of carrying out arthroscopy.

Materials and Methods

This is a prospective, observational study of a series of cases. Between 2014 and 2016 we evaluated 50 patients with medical and MRI diagnosis of complete ACL rupture. All of them were operated on using the same arthroscopic technique.

Forty-two of them were male, and eight were females, averaging 30.94 years old (ranging from 17 to 46). Thirty-one lesions affected patients' left legs and 19, patients' right legs. No patient had history of ligament rupture.

The ACL sample that was made to analyze by the histopathology department was got during the surgery.

We took two fragments right from both ends of the ruptured ACL. The samples sent for analysis were fixed in 10%-formaldehyde and, then, stained using haematoxylin

and eosin for ulterior evaluation.

The inclusion criteria were: 1) isolated injury of the ACL, 2) MRI preoperative diagnosis, 3) >16 and < 50 years of age and 4) closed ruptures.

The exclusion criteria were: 1) previous knee surgery, 2) multi-ligament injury, 3) immature skeleton, 4) previous local or systemic therapy which might have weakened the ligament (for example, local infiltration with anaesthetic or steroid agents in the knee region, immunosuppressive treatment in transplanted patients, autoimmune diseases, etc.). We registered associated meniscal injuries in every patient, their location (medial or lateral) and the treatment administered in every case (meniscectomy or meniscal repair).

Results

All the patients had suffered complete rupture of the ACL. Average time between traumatism and surgical intervention was 42.5 days (ranging from 20 to 180).

Histopathology analysis showed: fibrous connective tissue (42% of the patients), fatty tissue (4%), hyaline changes in collagen (46%), collagen degenerative changes (defined as the loss of febrile structure, which makes collagen homogeneous and hyaline) (36%) (Figure 1), congested blood vessels (10%), new formation of capillary vessels (3%), lymphoid infiltration (80%) (Figure 2), hemosiderin deposition (18%), foci of calcification and laminar bone tissue making up part of the ligament attachment area (6%) (Figure 3).

Forty-seven patients (94%) did satisfactorily (with no pain and resumption of previous activities, and MRI ACL osseointegration) and three (6%) developed arthrofibrosis.

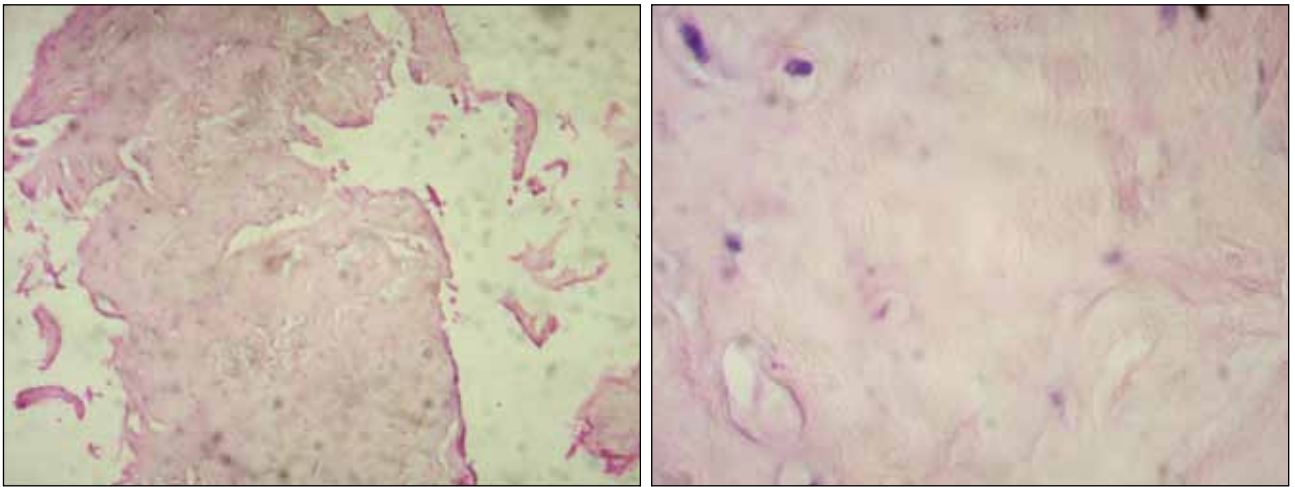
Discussion

The ACL has a complex structure. Its organization and biological features are directly related to its function.²¹ Due to its complex morphology, patients find it difficult to recover from its complete rupture.²²

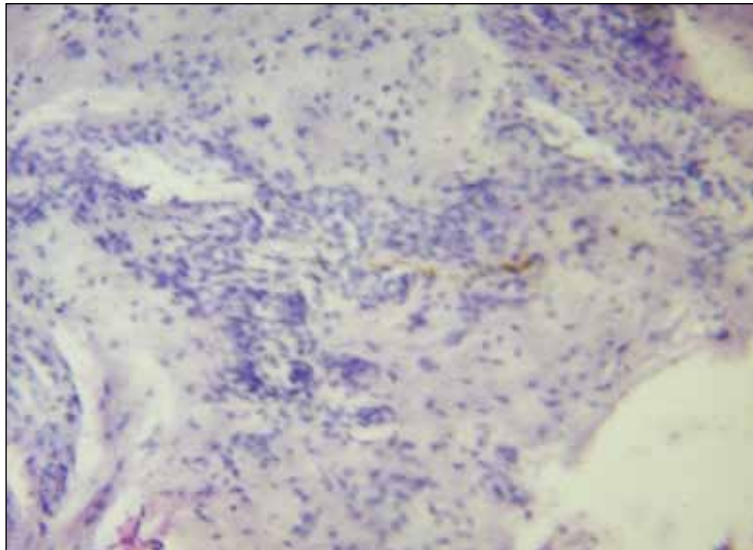
The arthroscopic reconstruction of the ACL is a surgery acknowledged worldwide.^{11,23} Parry et al.²⁴ reported that adults' tendons and ligaments have a bimodal distribution of collagen fibres, which are considerable wide in diameter and represent a great percentage of the tendon on transverse section. Collagen's crossed fibres account for collagen's high resistance to traction.⁷

The fibres of the injured ACL show intrinsic potential for healing due to blood supply given by synovial structures and the capability of fibroblasts to produce collagen.^{4,25}

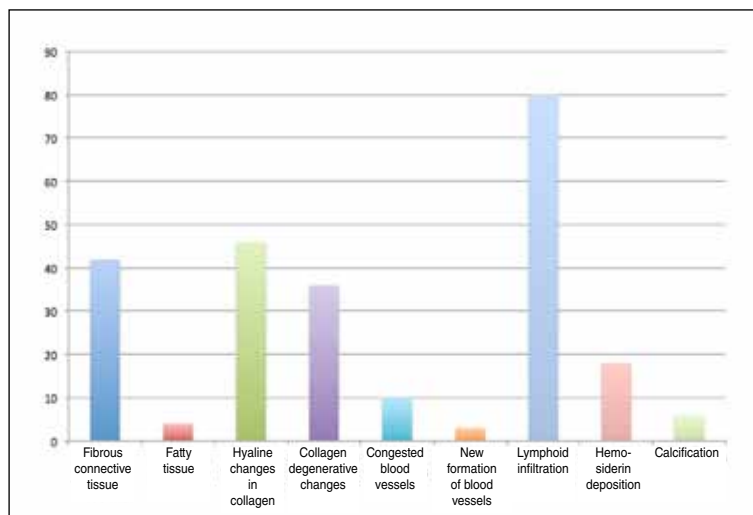
Strocchi et al.⁸ published that the bundles of collagen are irregularly aligned and that their ultrastructural or-



▲ **Figure 1.** Collagen degenerative changes, abundant collagen matrix and scarce fibroblast nucleus.



▲ **Figure 2.** Lymphoid infiltration in the ligament.



▲ **Figure 3.** Histopathology department report.

ganization is varied and complex. They verified that, in consecutive sections, most fascicles are orientated in different angles with respect to the long axis of the ligament, whereas just few fascicles run in parallel fashion. All these fascicles were studied while in rest position.

The mechanical properties of the connective tissues are directly related to their structural integrity. The disorganization and histopathological disorders of the collagen fibres can lead to the decrease of mechanical resistance and, thus, increase the likelihood of ligament rupture.²⁰

Bayat et al.²¹ studied the collagen fibres at the level of the deepest region of the ACL and fibrocartilage somewhere near the ACL bone attachment. In an observational study of the ACL ultrastructure, apart from finding fibro-

blasts in dense and spindle-like populations, and areas of collagen organized in bundles, HoChen et al.²⁰ verified the presence of areas with wavy fibres and irregular disposition of collagen fibres with increase in the number of cells and blood vessels.

Conclusion

In all ACL samples gathered for our study, there was evidence of disordered collagen structure, fibrous tissue and lymphoid infiltration. However, it could not be pinpointed whether these changes were previous or posterior to the ligament rupture.

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