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nal areas are composed of cells that present fibroblast-like morphology and phenotype. Interestingly, there is no expression of type II collagen throughout the whole young meniscus. In the adult meniscus, however, the three analyzed areas did not show the differences in cell morphology noted for the young tissue: all cell populations showed an intermediate morphology between chondrocytes and fibroblasts. Moreover, all cell populations were positive for type II and type I collagen presenting a gradient of type II versus type I: from the inner to the outer area, type II collagen decreases while type I increases.

**Conclusions:** These results lead to the conclusion that meniscus maturation, from young to adult, is accompanied by changes in cell phenotype; in the early stages of life the cells from the intermediate and outer part are still immature and far from a chondocyte-like phenotype; in adult life all meniscus cells assume a mature and specialized phenotype: cells of the external area maintain a fibroblasts-like phenotype, while cells of the intermediate and inner meniscus develop a chondrocyte-like phenotype.

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## CALORIMETRIC INVESTIGATION OF NORMAL AND DEGENERATIVE HUMAN MENISCUS

**G. Sohár**, K. Wellinger, Z. Aigner, P. Szabó-Révész, K. Tóth Univ. of Szeged, Szeged, Hungary

**Purpose:** Degeneration of human knee joint menisci has been the subject of numerous morphological studies, but detailed studies on the chemical composition and the metabolism of menisci are limited. Thermoanalytical techniques measure the change in physical or chemical properties of the sample as a function of temperature.

The main purpose of this study was to further characterize the altered metabolism in matrix composition during the late stage of meniscus degeneration that promotes disease progression. Based on previous studies, we hypothetized that enthalpy change of the process, initiated by the temperature change, might represent potential marker of the disease activity. Patients with different degree of meniscal degeneration were chosen for our investigations to find correlation between the enthalpy changes and the severity of disease.

**Methods:** The human knee joint meniscectomy specimens were received from the Orthopedic Department, University of Szeged. After surgical removal, the menisci were dissected free of their attachments and 5 mm thick disc was produced under sterile conditions from the middle 2/3 of each meniscus.

The calorimetric properties of samples were determined by DSC method (Mettler-Toledo DSC 821e apparatus). Samples were heated from 0 to 80 °C. The heating rate was 0.3 °C/min. Conventional Hastelloy batch vessels were used with 40  $\mu$ l sample volume. All the DSC measurements were preceded in Ar atmosphere, and the flow rate was 100 ml/min.

**Results:** The enthalpy change of the process initiated by the temperature change showed marked difference between the normal and pathological groups. With the rise of temperature an endothermic reaction was observed in all of the cases.

Change in the enthalpy was observed in normal cartilage as 1632.48 J/g (SD = 50.55). In case of early degeneration a greater change at 1707.83 J/g (SD

= 112.46), while in the severely degenerated samples at 1677.30 J/g (SD

= 182.48) was measured. Therefore, denaturation caused by heating was largest in the early stage of degeneration of human meniscus. Consequently these samples required the largest amount of energy for decomposition. Denaturation peak in normal samples was at 56.06 °C (SD = 1.91), however it was lower in the early stage at 52.55 °C (SD = 6.45) and similar in sever degeneration at 50.08 °C (SD = 7.14).

**Conclusions:** In summary, we examined the thermal properties of human meniscus tissue of normal origin in young adults and in patients with early and late stages of degeneration. We were the first ones who used normal samples that were extracted from live surgeries for the investigations.

A newly established calorimetric protocol was used for our experiments. This method proved to be suitable for compositional thermoanalytical study of normal and degenerative human meniscus samples.

Our results showed clear evidence that complex deviations from the normal matrix composition during degeneration correlated with changes in thermal properties. Correlation was found between the enthalpy changes and the severity of degeneration. All samples that were extracted for this study were obtained during live surgeries. A new protocol was established, using simple saline solution instead of the previously used phosphate buffer. This new method proved to be suitable for the thermoanalytical investigations.

Meniscus damage may play an important role in osteoarthritis pathophysiology. Whether meniscus damage or cartilage degradation occurs first is unknown, however. A torn meniscus and extrusion seem to be strong risk factors for the development and progression of knee osteoarthritis.

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## PLATELET-RICH PLASMA INJECTION TO IMPROVE TENDON HEALING PROCESS

**J.-F. Kaux**<sup>1</sup>, P. Drion<sup>1</sup>, J. Renouf<sup>1</sup>, F. Pascon<sup>1</sup>, V. Libertiaux<sup>1</sup>, C. Le Goff<sup>1</sup>, A. Gothot<sup>1</sup>, S. Cescotto<sup>1</sup>, J.-O. Defraigne<sup>1</sup>, M. Rickert<sup>2</sup>, J.-M. Crielaard<sup>1</sup> <sup>1</sup>Univ. of Liège, Liège, Belgium; <sup>2</sup>Univ. of Heidelberg, Heidelberg, Germany

**Purpose:** It is well known that injured tendons do not heal easily. For example, tendinopathy is a condition which often becomes chronic in the case of bad or late management. Recently, several studies, essentially in vitro and, more recently, a few in clinical practice, have demonstrated the positive effects of platelets on the healing process of different tissues. In fact, platelets contain lots of growth factors which can be released after a local injection. These growth factors have the potentiality to enhance the tendon healing process, for example after rupture or tendinopathy.

The aim of our experiment was to ascertain whether the use of Platelet-Rich Plasma (PRP) was of interest for accelerating the healing process of Achilles tendon after surgical induced lesion.

**Methods:** All experimental procedures and protocols used in this investigation were reviewed and approved by the Institutional Animal Care and Use Committee of the University of Liège.

60 rats were divided into 2 groups: A: control (no injection) and B: PRP injection. A 5mm defect was surgically induced in the rats' Achilles tendon after resection of plantaris tendon. Rats of group B received a PRP injection in situ after the surgery. Afterwards, rats of both groups were placed in their cages without immobilization.

After 5, 15 and 30 days, the traumatized Achilles tendons of 10 rats of both groups were removed and dissected during their healing process. Immediately after sampling, tendons were submitted to a biomechanical tensile test up to rupture, using a "Cryo-jaw". Rats were then euthanized.

Statistical analyses were made with an ANOVA. Values are significant when p-value is below 0.05.

**Results:** We observed that the force necessary to induce tendon rupture during biomechanical tensile testing increased with time in both groups; that this force was greater for tendons which had been submitted to an injection of PRP. The ratio between force and weight increased with time in both groups; that this ratio was greater for tendons which had been submitted to an injection of PRP too. There is also a significant interaction between time and the group.

The surface area of the section of the tendons increased between 5 and 15 days followed by a stabilization. After 30 days, sections in both groups were similar. Thus, the constraint was similar after 5 and 15 days but is significantly better for PRP group after one month.

**Conclusions:** We demonstrated that the force necessary to induce tendon rupture during biomechanical tensile testing was greater for tendons which had been submitted to an injection of PRP. These results were observed and significant (p<0.05) from day 5 onwards. We observed too that the section of the tendon was the same in both groups after 30 days. Thus the quality of the healing tendon is better with an injection of PRP, as shown with the increase of the constraint until rupture.

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### Pain: Clinical and Pathophysiology

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# SOMATOSENSORY ABNORMALITIES IN PATIENTS WITH KNEE OSTEOARTHRITIS

V. Wylde<sup>1</sup>, I.D. Learmonth<sup>1</sup>, S. Palmer<sup>2</sup>, P. Dieppe<sup>3</sup> <sup>1</sup>Univ. of Bristol, Bristol, United Kingdom; <sup>2</sup>Univ. of West of England, Bristol,

United Kingdom; <sup>3</sup>Univ. of Exeter, Exeter, United Kingdom

Purpose: The aim of this study was to use Quantitative Sensory Testing