

collapse of the femoral head with degenerative changes. It has been estimated that approximately 10 000 to 20 000 new cases are diagnosed in the USA each year and there are 300 000 – 600 000 people diagnosed with AVN.

Aim of the study. To elucidate the actual status in etiology of AVN of femoral head.

Material and methods. The following databases were used for articles search: Pubmed, Embrase, Hinary, Web of Science, Medline, Sciencedirect, for searching articles. We have selected and studied 74 articles containing the keywords: AVN of the femoral head, etiology of AVN, genetic disorders in AVN.

Results. Traumatic aseptic necrosis of the femoral head appears as results of mechanical disruption of blood flow to the femoral head. The non-traumatic causes of secondary AVN of the femoral head are: chronic alcohol consumption (20–40%), corticosteroid therapy (35–40%), after organ transplant, haematologic disease (anemia, polycythemia, hemophilia, thalassemia), clotting diseases, connective tissue disease, infiltrating diseases; some endocrine diseases (Cushing disease, hyperparathyroidism), metabolic diseases (gout, hyperuricemia, high cholesterol), congenital diseases (congenital sprain hip joint, Legg–Calvé–Perthes disease), Caisson disease, pancreatitis, chronic renal failure, hemodialysis, chronic liver disease, HIV infection, pregnancy, chemo- and radio- therapy, thrombophlebitis. Approximately 10 to 20% of cases do not have any identifiable risk factors and are therefore considered to be idiopathic in nature. It has been shown that some genes are involved in the pathogenesis of AVN: ADH2, ADH3, ALDH2 and P450E1. These genes are involved in the alcohol metabolism and polymorphisms of these genes have been associated with the risk of AVN. Jones et al. found that approximately 82% of patients in their study had at least one coagulation factor abnormality. Familial forms of AVN of the femoral head appear to be very rare, with only a few families reported in the medical literature. Liu et all. noted that a COL2A1 gene mutation in certain families predisposed to development of AVN of the femoral head by autosomal dominant transmission.

Conclusions. 1. Avascular necrosis of the femoral head is especially common among young people, affecting mainly men. Often an underlying cause cannot be determined. 2. Aseptic necrosis of the femoral head is a disease whose etiology is not completely elucidated while the actual role of the genetic disorders in this pathology is to be determined.

Key words: avascular necrosis of the femoral head, etiology of avascular necrosis, genetic disorders in avascular necrosis

266. THE THREE-DIMENSIONAL LIVER MATRIX FOR TISSUE ENGINEERING

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Introduction. According to The World Health Organization in 2012, about one-third of the world's population has serological evidence of hepatitis B infection (VHB). Terminal stage liver disease or hepatocellular carcinoma caused by VHB, leads to 0.5-1 million deaths per year. Worldwide viral hepatitis B is considered the 9th cause of death and represents 5-10% of all liver transplantation. That's why the phenomenon is perceived as significant global issues in public health. The growing of people number who need the liver transplant and the insufficiency of organ donors, as the advancement in bioengineering has enabled the development of new therapeutic strategies which involve generation of functional artificial organ, obtained by the decellularization technology and create extracellular matrix and their subsequent recellularisation.

Aim of the study. To obtain a liver matrix by decellularization and to maintain its vascular tree.

Materials and methods. As the object of this study served rat livers (n=9) which were subjected to decellularization with sodium dodecyl sulfate solution (SDS) 0.1 and 0.5% and the combination of sodium dodecyl sulfate 0.1% to 0.5% and anticoagulant. Subsequently, the extraction of nucleic acids was performed according to the protocol QIAamp Blood Mini Kit (2003).

Results. After the liver tissue decellularization we obtained the liver matrix. The quantification of nucleic acids revealed the existence of a small amount of DNA 1.04 ± 0.43 ng/ μ l, * p<0,05 in decellularised matrix with SDS solution and anticoagulant. In case of decellularization by SDS exclusively, we obtained a larger amount of nucleic acids which revealed a less efficient decellularization 5.2 ± 2.19 ng/ μ l, * p<0.05.

Conclusions. The use of detergent SDS with anticoagulant for decelularisation is more effective method in comparision with only SDS solution, which was proved by quantification of nucleic acids content in decellularised matrix. A more efficient decellularized liver tissue represent a 3D bioconstruction for future recellularisation.

Key words: decellularization, recellularisation, liver matrix.

267. OBTAINING OF A SUITABLE OSTEOCHONDRAL GRAFT FOR ARTICULAR CARTILAGE ENGINEERING

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Introduction. Chondral injuries are common following a knee trauma. There are numerous studies with different ways to obtain a suitable graft for articular cartilage regeneration, but without imposing results.

Material and methods. From two freshly sacrificed rabbits the distal femurs were harvested and frozen at -84°C for one week. From each distal femur all tissues except cartilage and subcondral bone were removed and small pieces of normal osteochondral tissue (NOCT) were taken. The remaining osteochondral tissue has been demineralized in 0,6M HCl (Chem-Lab, Belgium) over night and again small pieces of demineralized osteochondral tissue (OCDT) were cutted with a scalpel and placed in a PBS solution for 24 hours. The remaining OCDT were separated in 4 groups. Two groups were decellularized in 0,5% and 1% SDS (Sigma, UK) and another two in 0,5% and 1% Triton X-100 (HiMedia, India). The decellularization lasted for 24 hours. At the next day the decellularized and demineralized osteochondral tissues (OCDDT) were washed with distilled water and PBS for 24 hours. All tissues were dessicated through centrifugation at 4000 rpm for 10 min (Hettich, Germany). From all types of OCT were cutted from three to nine pieces 20 mg each and quantification of DNA was performed with GeneJET Genomic DNA Purification Kit (Thermo Fisher, Lithuania). The results were read with spectrophotometer NanoDrop 2000c at wavelength of 260 nm (Thermo Fisher, USA). The best decellularized tissue and OCDT were tested for cytotoxicity with MTT test (ISO 10993-5) with mesenchymal stem cells and chondrocytes.

Results. The average of DNA content in a rabbit NOCT is 36 ng/ μ l, in OCDT 4,23 ng/ μ l, OCDDT with 0,5% and 1% SDS is 3,23 ng/ μ l and 2,16 ng/ μ l respectively and in OCDDT with 0,5% and 1% TritonX-100 is 1,96 ng/ μ l and 0,96 ng/ μ l. At the MTT assay with mesenchymal stem cells and chondrocytes on the OCDT and OCDDT with 1% TritonX-100, we obtained a higher cell viability in both cases more than 80%.