

MORPHOLOGICAL CHANGES IN THE OSTEO-CARTILAGINEOUS ENDPLATE AND ANULUS FIBROSUS OF CERVICAL AND THORACIC SPINE AFTER TRAUMA – CASE REPORTS.

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INTRODUCTION: The aim of the study was to evaluate typical morphological changes in the endplate and the AF in relation to time after trauma. Animal study group: The annulus fibrosus (AF) and the endplate of lower cervical spine from healthy 6 to 8 months old pigs was used to evaluate vertebral bone and disc architecture as well as cell death by necrosis. Patients study group: Specimens of the AF and endplate from the lower cervical and thoracic spine of four traumatically injured patients were taken during operation. Anterior fusion was performed in between four months after trauma. Recent investigations have shown that chondrocytes undergo apoptosis to regulate cell proliferation in the hypertrophic zone of the growth plate in the maturation process 1. Evidence has been presented so far that apoptotic cell death occurs in the AF of degenerative discs 2+3. The possibility that necrosis is even more important in traumatic injured discs, has not yet been published.

METHODS: Animal study group: Specimens from AF were investigated after one, twelve and twenty four hours after death, to study necrosis in chondrocytes. Endplates were collected and embedded in different positions to evaluate different angles of cutting planes (sagittal to horizontal planes). Patient study group: Specimens from four patients (17 – 58 a) have been investigated so far. One taken at the day of trauma, the other three at one week, three weeks and four months. Tissue samples were taken during operation and fixed immediately (less than 15 minutes) for histological and ultrastructure evaluations. Investigations were done by light microscopy (LM) and transmission electron microscopy (TEM). The specimens were fixed for LM investigations with Schaffer solution for two days followed by dehydration with Ethanol and embedded in Methylmethacrylat. The tissue blocks were sectioned at 4-6µm. Staining was performed with Goldner and Methylenblue. LM sections were examined on an AX70 Olympus-Microscope.

If possible specimens were taken for TEM. From outer AF tissue samples were diced in one millimetre cubes. They were fixed in 0,1 M Glutaraldehyd in Cacodylat buffer for twelve hours.

The specimens were rinsed with Cacodylat buffer and postfixed with Osmiumtetroxyd. Embedding was procedured in the routine manner with Araldit. Semithin (6µm) and ultrathin (0,90µm) sections were cut and stained with Uranyl acetat and lead nitrat (Reynolds). Sections on 200µm mesh grids were examined in a TEM 10 (Zeiss). Five different areas of each patient and each specimen of the pig were studied.

RESULTS, DISCUSSION & CONCLUSIONS:

Animal study group (LM and TEM): In TEM morphological changes could be detected after 12 hours, starting with swelling of the organelles and sometimes also with leaking in the cell membrane. At 24 hours same lesions became more intense. Osmiophilic cell detritus was identified in the disc tissue same time. In LM horizontal planes of sections gave a better overview, than sagittal sections. Morphological changes in LM have not been as obvious as in TEM in the time range of twenty four hours.

Patient study group (LM and TEM): Not surprisingly, in specimens obtained on the day of trauma (patient 1) ample signs of massive bleeding and cracks within both, hard and soft tissue components of the samples were the predominant features. In TEM cells appear swollen, next to osmiophilic cells.

At one week after the event (patient 2), irregularities especially at the bone-(fibro)cartilage interface including first signs of vascular proliferation and bone remodelling, bleeding due to vascular damage and dead chondrocytes were conspicuous. Electron micrographs showed that cell death is evident as well as swollen cells with enlarged organelles.

Cartilage cell clusters as a sign of focal remodelling was evident three weeks after trauma (patient 3) in both, at the osteochondral junction and AF.

Four months after trauma (patient 4) vascular ingrowths into bone marrow, endplate and AF was paralleled by interposition of fibrous connective tissue containing large amounts of cellular components. New areas of fibrocartilage were seen next to damaged tissue. Ultrastructural investigations demonstrated strongly osmiophilic

chondrocytes and on the other hand sign of necrosis.

In preliminary results, necrosis seems to be more important in the first two weeks after trauma. Cartilage cell clusters could be seen first three weeks after trauma. Four months after trauma vascular ingrowths into bone marrow and anulus fibrosus is evident.

We expect with our morphological investigations of the posttraumatic period to show the specific reaction of the chondrocytes in discs and endplate to comparable traumata. This might influence the clinical procedure in some cases.

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