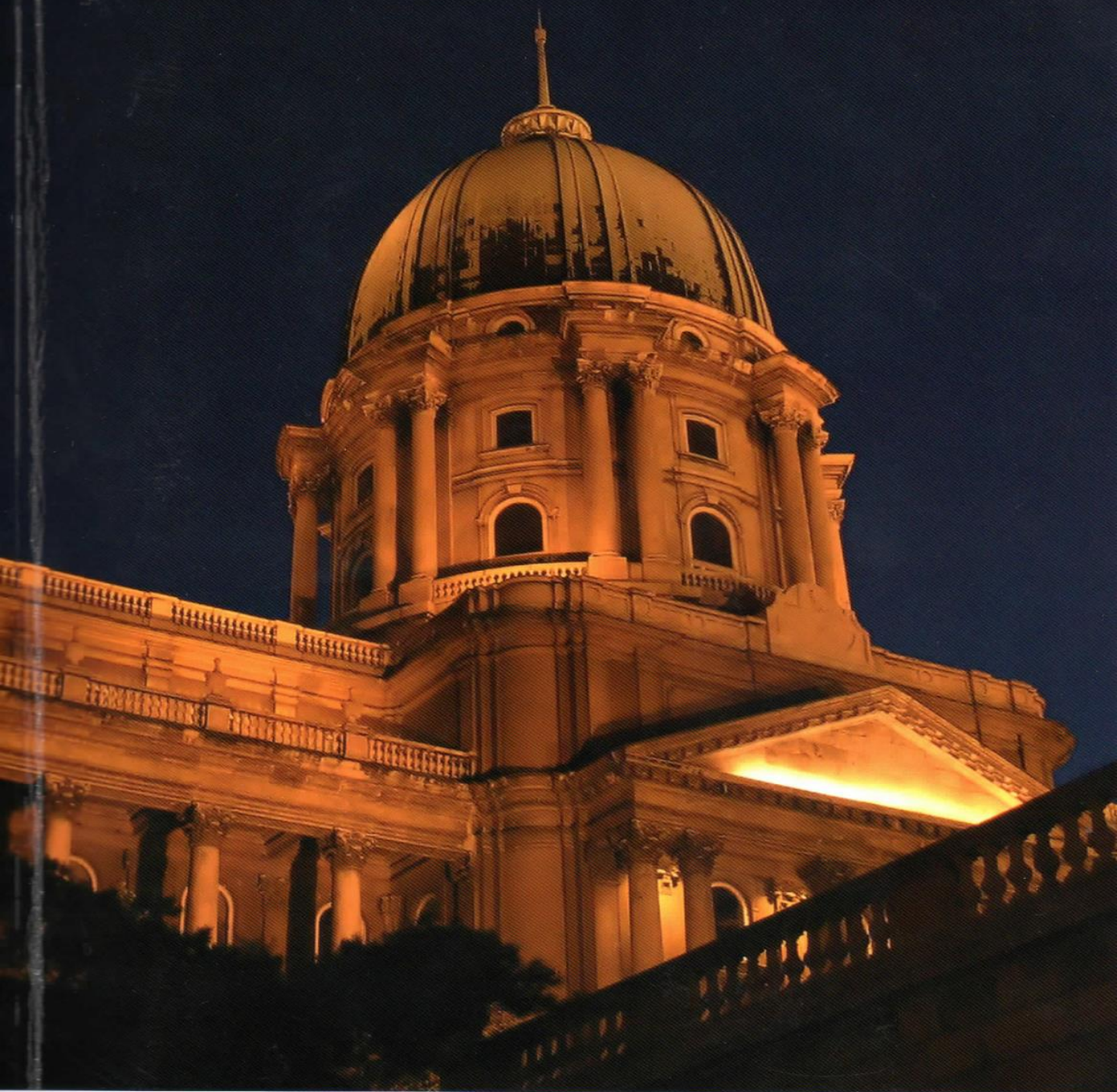




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**P-67****STUDYING THE MEMBERS OF ENDOCANNABOID SYSTEM IN MOUSE MICROMASS CULTURES**

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Background: More and more countries legalize cannabis-based products for treating various pathological conditions of the skeletal system (e.g. osteoarthritis, rheumatoid arthritis) due to their pronounced analgesic and anti-inflammatory effect. Cannabinoid 1 and 2 Receptors (CB1, CB2) are known to be expressed in synoviocytes and adult articular chondrocytes; but presence and possible role of this system during embryonic formation have not been investigated yet.

Methods: Mouse MicroMass cultures (MMMC) were established from distal limb buds of 11.5-day-old mouse embryos. Chondrogenesis spontaneously occurs during the 6-7 days of culturing. The expression of EndoCannabinoid System (ECS) receptors and enzymes were monitored by reverse-transcription PCR technique through the 6 days of culturing. Protein expression of CB1 was examined using immunohistochemistry in the distal limb buds of 15-day-old mouse embryos and also in MMMC. Chondrogenesis was monitored by metachromatic staining. Expression pattern of ECS members was investigated by RT-PCR after CB receptor agonist and antagonist treatments.

Results: The most important members of the ECS are expressed at mRNA level in chondrifying MMM. CB1 receptor is expressed in both MMMC and in the distal limb buds of mouse embryos at a protein level. According to our preliminary results an agonist of the ECS, anandamide positively influences chondrogenesis when it was applied at low concentrations to MMMC. Measurements of the changes of intracellular Ca^{2+} concentration under effect of CB receptor agonist and antagonist are in progress.

Conclusion: Mature and differentiating chondrocytes express members of ECS, and endocannabinoids may influence embryonic cartilage formation.

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