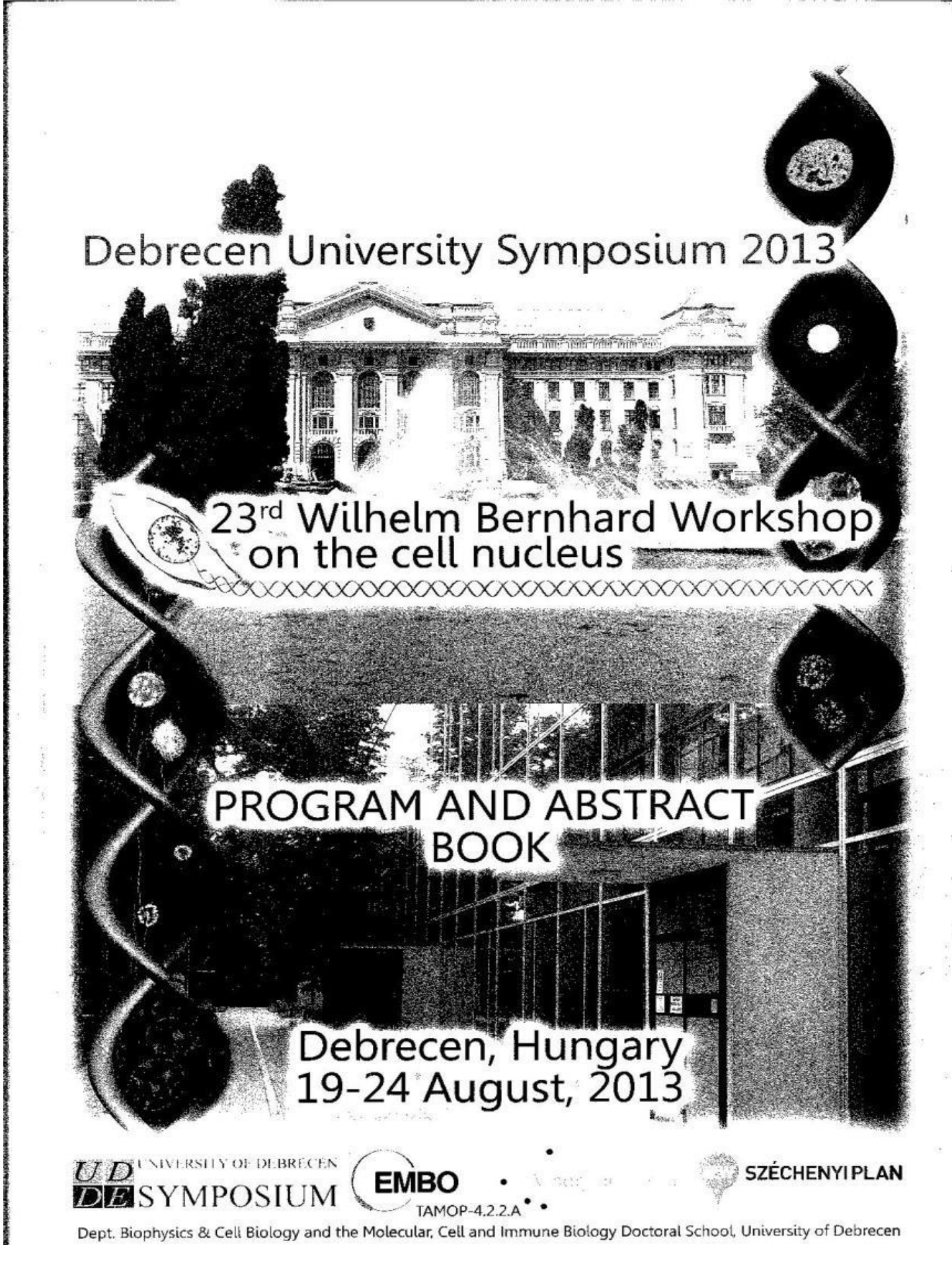




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SZÉCHENYI PLAN

Dept. Biophysics & Cell Biology and the Molecular, Cell and Immune Biology Doctoral School, University of Debrecen

P38. TRANSIENT RECEPTOR POTENTIAL VANILLOID TYPE 1 AND CHONDROGENESIS

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Our major interest is to study *in vitro* chondrogenesis, focusing on the relation of various extracellular signals, plasma membrane ion channels, intracellular Ca^{2+} -concentration changes and protein phosphatases in chondrifying high density cell cultures (HDC). Though, chondrocytes are non-excitabile cells, they express a large set of ion channels (e.g. purinergic P2X₄, glutamate receptor NMDARs, Kv channels) maintaining a dynamic relationship with the acidic and hypoxic extracellular environment in cartilage. Transient Receptor Potential Vanilloid (TRPV) subfamily of non-selective cation channels of plasma membrane has not been studied in a detailed way during cartilage formation, except TRPV4 activity that contributes through the Sox9 pathway in the process of chondrogenesis.

Our experimental model is a primary chondrogenic HDC system, in which chondrogenic cells are isolated from limb buds of 4-day-old chicken embryos. Spontaneous cartilage formation occurs within 6 days in HDCs.

Chicken chondrogenic cells express mRNAs of various TRPV members during the course of cartilage differentiation. We focused on the polymodal thermo- and mechanosensitive TRPV1 membrane receptor and were able to detect several bands, including the one with the expected MW with two different TRPV1 antibodies in Western blots in samples of chicken articular cartilage or samples from chicken HDCs, as well as in chicken brain and dorsal root ganglia, applied as positive controls. Therefore, we also plan to identify the interested protein and its possible splice variants with mass spectrometry. These experiments are in progress. As avian TRPV1 contains a truncated capsaicin-binding region, it is insensitive to capsaicin or related pharmacological modulators. Therefore, we applied heat treatment on chicken HDCs to prove functionality of the channel and monitored temporal pattern of TRPV1 mRNA expression in a 4 hours period. To clarify the precise function of TRPV1 during chondrogenesis we need to perform transient gene silencing in HDCs.

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