

Synthesis of Neurolysin (EC 3.4.24.16) in the Rat Corneal Endothelium

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Neurotensin (NT) is a 13 amino acid peptide that displays a wide range of biological activities including hypotension, hypertension, hyperglycemia, hypothermia and increased vascular permeability. In the eye, NT has been detected in the iris-ciliary body and in aqueous humor. The anterior chamber of the eye is bounded anteriorly by the corneal endothelium, except at its far periphery, and it is filled with aqueous humor. The purpose of this report was to verify the rat corneal endothelium capacity to express neurolysin (EC 3.4.24.16), a metalloendopeptidase involved in neuropeptide metabolism, including NT. To achieve this aim, total RNA from rat corneal endothelium was processed for RT-PCR using neurolysin specific primers and the amplification products were sequenced. The whole protein extract of corneal endothelium was electrophoresed in a 10% SDS-PAGE gel and transferred onto the PVDF membrane to be analyzed using the Western Blot technique with a rabbit anti-neurolysin antibody. The cornea was also processed for neurolysin immunolocalization using the same antibody. The RT-PCR and sequencing experiments detected the presence of neurolysin mRNA in the rat corneal endothelium. The Western Blot assays demonstrated immunochemical cross-reactivity with a protein of the expected molecular mass to neurolysin (~78 kDa). The reactivity to neurolysin, following immunohistochemical processing, was observed in the corneal endothelium. Taken together, these results indicate that the rat corneal endothelium expresses neurolysin. Its expression indicates that the corneal endothelium could participate on the neuropeptide processing in the anterior segment of the eye especially that related to the neurotensin inactivation.

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