MOLECULAR AND BIOCHEMICAL ANALYSIS OF APOLIPOPROTEIN E EXPRESSION BY THE RAT CORNEA

Bertazolli-Filho, R., Haddad, A. & Laicine, E.M.
Departamento de Biologia Celular e Molecular e Bioagentes Patogênicos, FMRP/USP

Apolipoprotein E (ApoE) plays a role on the delivery of plasma cholesterol to the cells. In the eye, ApoE is synthesized by the Müller cells of the retina and it is detected in the aqueous humor. The present study investigated the expression of the ApoE by the cornea. This was carried out using different approaches: ***i.*** explants of rat corneas were incubated in presence of ^{35}S-methionine and the culture media were immunoprecipitated using antiApoE antibody-Protein A Sepharose CL-4B, and the resulting extracts were analyzed by SDS-PAGE plus fluorography; ***ii.*** histological sections of cornea were processed for ApoE immunocytochemistry; ***iii.*** whole RNA samples from isolated rat corneas were processed for RT-PCR, using primers made on the basis the known sequence of rat liver ApoE mRNA; the RT-PCR product was analyzed in agarose gel, then purified and sequenced. From the immunoprecipitation experiments, it was detected a labeled fraction with electrophoretic mobility of ApoE. The immunocytochemical analysis showed that corneal stroma and endothelium were positive for ApoE. The size of the fragment obtained from the RT-PCR was the one expected; its sequence matched 100% when compared to the liver one. These data show that the cornea is able to synthesize and secrete ApoE, indicating that it may be the source for at least part of the aqueous humor ApoE.

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