

Benzalkonium Chloride Homologs in Ocular Surface Cells in Vitro and in Vivo

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Purpose Benzalkonium chloride (BAC), a common preservative in ophthalmic products, is a mixture of homologs. The cytotoxicity of three different homologs and BAC mixture (containing 64.1% C₁₂, 33.8% C₁₄, 2.1% C₁₆) were studied in corneal and conjunctival epithelial cell cultures. The distribution of homologs in ocular surface tissues of rabbit eyes was examined.

Methods Human corneal epithelial (HCE) and conjunctival epithelial (IOBA-NHC) cell cultures were exposed to homologs or mixture for one hour. Cytotoxicity was assessed with the WST-1 assay. BAC solution was applied into rabbit eyes daily for 14 days and homologs in corneal and conjunctival tissues were analyzed.

Results *In vitro*, conjunctival cells appeared to be more sensitive to BAC exposure than corneal cells. In corneal cells, the cytotoxicity of tested preservatives was rather similar, with the EC₅₀ values 0.00130% for C₁₆, 0.00127% for BAC mixture, 0.00101% for C₁₂, and 0.00097% for C₁₄. In conjunctival cells, the EC₅₀ values were 0.00065% for C₁₆, 0.00047% for BAC mixture, 0.00041% for C₁₄, and 0.00038% for C₁₂. The amounts of C₁₂, C₁₄ and C₁₆ in rabbit corneal and conjunctival tissues were respectively (mean ± SEM, n=5): 0.37 ± 0.08 and 2.64 ± 0.27 ng/mg, 0.42 ± 0.07 and 4.77 ± 0.43 ng/mg, 0.04 ± 0.01 and 0.54 ± 0.05 ng/mg.

Conclusions Besides different potency of toxicity, BAC homologs possess different absorption in ocular surface tissues. The greater sensitivity of conjunctival cells than corneal cells to homologs *in vitro* may be explained by higher absorption of homologs into the conjunctiva *in vivo*.