TRANSITION OF EPIGLOTTAL EPITHELIUM DURING HUMAN DEVELOPMENT

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Our previous results have shown that human epiglottal epithelium changes during development, from single-layered squamous epithelium in the 6-week-old embryo to two-layered cuboidal epithelium in the early 8-week-old embryo; in the newborn, pseudostratified epithelium with goblet cells predominates and after the air-flow is established, stratified squamous epithelium predominates. Seven-week-old and 9-week-old embryo epiglottal epithelium was now analyzed in more detail and compared. Embryos from the celloidin collection of human embryos belonging to the Archive of the Department of Histology and Embryology were stained by HE, Azan, Masson trichrome stain, Verhoeff iron hematoxylin and PAF-Halmi. Epiglottal epithelia were investigated by light microscopy. In the 6-week-old embryo, epiglottal swelling was covered by a single layer of cuboidal cells. In the 7-week-old embryo, two-layered cuboidal epithelium was discovered. In 9-week-old fetus, epiglottal epithelium was multilayered columnar without cilia and goblet cells. It has now been confirmed that single-layered epiglottal epithelium has changed to the two-layered epithelium already in the 7-week-old embryo. Developmental studies like this one might be of importance for tissue engineering purposes.

DETECTION OF CLONAL T-CELL RECEPTOR GAMMA CHAIN GENE REARRANGEMENTS IN SUSPECTED CUTANEOUS T CELL LYMPHOMAS

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Molecular analysis of rearranged T cell receptor (TCR) represents an important diagnostic tool in diagnosing cutaneous T cell lymphomas. We report here three cases suspected of cutaneous T cell lymphomas, for which dermatopathologists were unable to give final diagnosis without the use of T cell clonality analysis. Working diagnoses in study patients were mycosis fungoides, cutaneous lymphoma and pseudolymphoma. DNA was extracted by NucleoSpin Tissue XS kit (Macherey-Nagel, Germany) designed for extra small amount of material (less than 100 cells), and suspected cells were selected by laser-microdissection from microscopic slides of paraffin embedded biopsy materials. Using modified BIOMED multiplex nested PCR and primers specific for gamma chain of the T cell receptor (TCR-γ), we were able to amplify the specific gene region. The PCR products of 65 to 95 bp in length, labeled with 6-FAM flourescent dye, were separated by capillary electrophoresis on the ABI Prism 310 Genetic Analyzer; results were analyzed by use of GeneMapper software (Appl. Biosystems). In all experiments, positive and negative controls were included. Multiplex nested PCR analysis was performed in triplicates. Specific amplification products of 65 to 95 bp were obtained in all experiments, and separation and analysis of amplicons revealed polyclonal rearrangement patterns in all three cases. The results obtained showed absence of clonal rearrangement and indicated reactive proliferation. In conclusion, capillary electrophoresis sensitivity reached down to1 bp, so we were able to easily distinguish different clones. The multiplex nested PCR method and capillary electrophoresis proved to be a highly sensitive screening tool for clonal TCR-γ chain gene rearrangements.