Establishment and characterization of an oral squamous carcinoma cell line from a patient with no apparent habits associated with oral cancer

Type: Meeting Abstract

Content:

Introduction: We have established a cell line ORL-48(T) from a surgically resected specimen of an untreated primary human oral squamous cell carcinona of the mandible. This patient did not appear to have any oral habits which have been reported to be associated with oral cancer Materials and Method: The in vitro growth characteristics, epithelial origin, in vitro anchorage independency, HPV infection, microsatellite instability status, karyotype and the status of various cell cycle regulators and gatekeepers of the ORL-48(T) cell line were investigated. **Results**: The ORL-48(T) cell line is immortal, 3T3- independent and grew as a monolayer with the doubling time of 48h. Immunohistochemistry staining of cytokeratins confirmed the cell line is of epithelial origin. Soft agar assays demonstrated that ORL-48(T) expressed a low degree of anchorage independency (CFE<6%). Karyotyping analysis revealed that ORL-48(T) is an euploidy in which the number of chromosomes varied between 67 and 73. There is consistent trisomies of chromosomes 1, 2, 3, 5, 7, 9, 12, 15, 16 and 18 and homogenously staining region on 4q suggesting gene amplification in this region. Sequencing of exons 5, 6, 7, 9 and 10 of p53 revealed a mutation resulting in a stop codon. Molecular analysis of p14INK4a indicates hypermethylation of the p14INK4a promoter. In contrast to normal human keratinocytes, MDM2 is overpressed in ORL-48(T). Microsatellite analysis of BAT26 indicated that ORL-48(T) is microsatellite stable and this was confirmed by western blots demonstrating the expression of both hMSH2 and hMLH1 mismatch repair proteins. Polymerase chain reaction demonstrated that ORL-48(T) was not infected with human papillomavirus (HPV). Conclusion: This well characterized cell line will be useful tool in the understanding of the molecular changes associated with oral cancer particularly in cases where no clear aetiological factors are present.

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