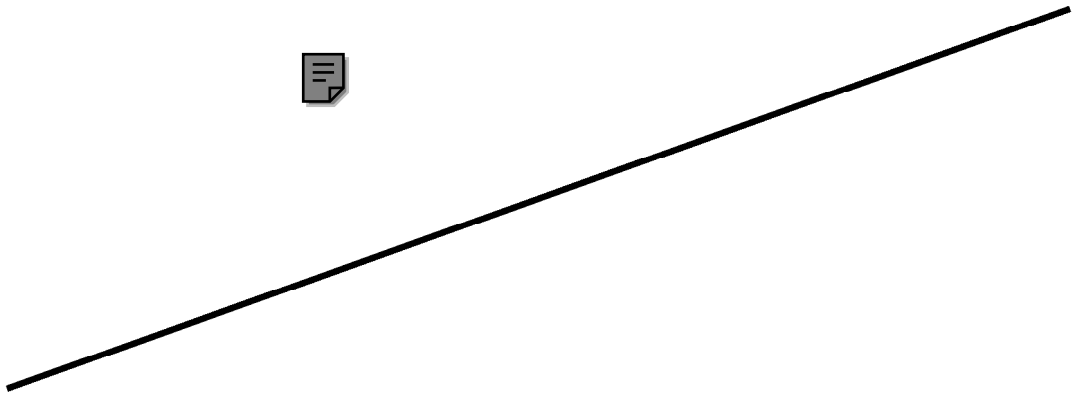

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219 **Establishment of a spontaneously immortalized rat bronchial epithelial cell line with basal cell-like characteristics**

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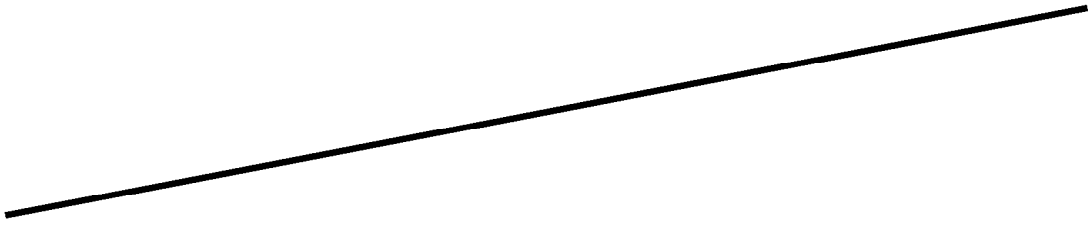
We isolated lung epithelial cells from an adult male Wistar rat by pronase digestion and cultured them in Ham's F12 medium containing 5% FBS, insulin, transferrin, EGF, aFGF and cholera toxin. Out of a mixed cell population containing type II pneumocytes and ciliated cells as the major cell types, one cell type was established as a spontaneously immortalized cell line. This adherent cell line expresses different cytokeratins (CK) as intermediate filaments, which is specific for epithelial cells. The predominant appearance of CK 14 and 15, determined by western blotting at passage 42, points to a possibly bronchial basal cell origin. During ageing the postconfluent monolayers produce a squamous cell layer that reacts with a desmoplakin antibody, which recognizes desmosomal structures that are typical for epithelial cells. In addition, markers for type II cells, Clara cells and fibroblasts are lacking. The cell line was passaged more than 40 times within 9 months with population doubling times that decreased from 31 h to 19 h with increasing passage number. These rat bronchial epithelial cells (RaBE) are able to grow at cloning density on a plastic surface with a CFE of 36 ± 3.6 % (mean±SD, n=10, 200 cells/100 mm dish). Chromosome analysis revealed a shift from 62 to 75 chromosomes (median, n=20-50) between passage 9 and 19. Although immortalized, the cell line did not change to a transformed phenotype, as indicated by the strong dependency on serum and growth factors. Whether these cells represent pluripotent stem cells that are able to differentiate under complex culture conditions is part of our current investigation. This rat bronchial epithelial (RaBE) cell line is now well established and might be useful for cell communication or toxicity tests, and possibly for mutation assays, which remains a subject for future studies (This work was supported by Philip Morris, USA).



Eur. J. Cell Biol. 79: 78-78 (2000)