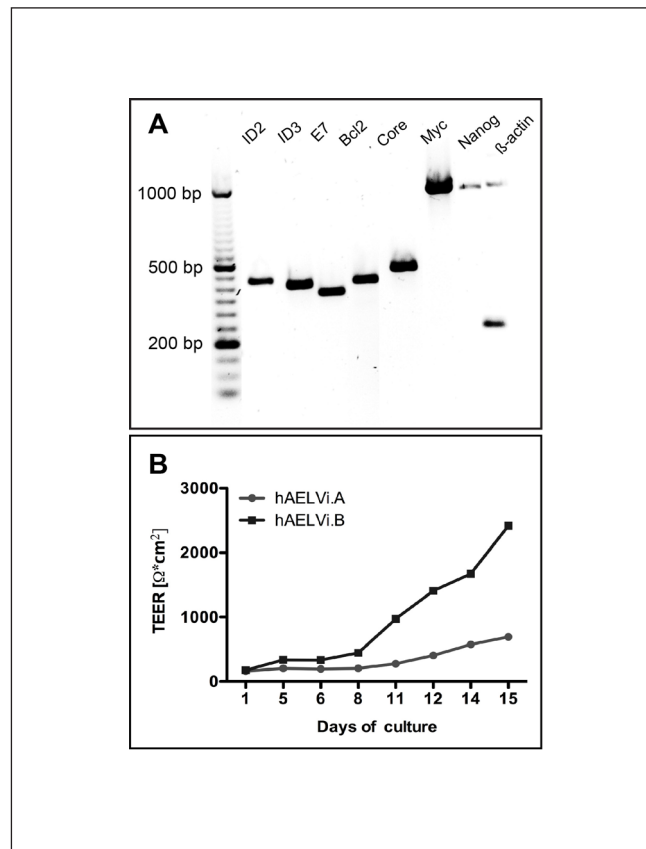


Kuehn et al.:

# Human Alveolar Epithelial Cells Expressing Tight Junctions to Model the Air-Blood Barrier

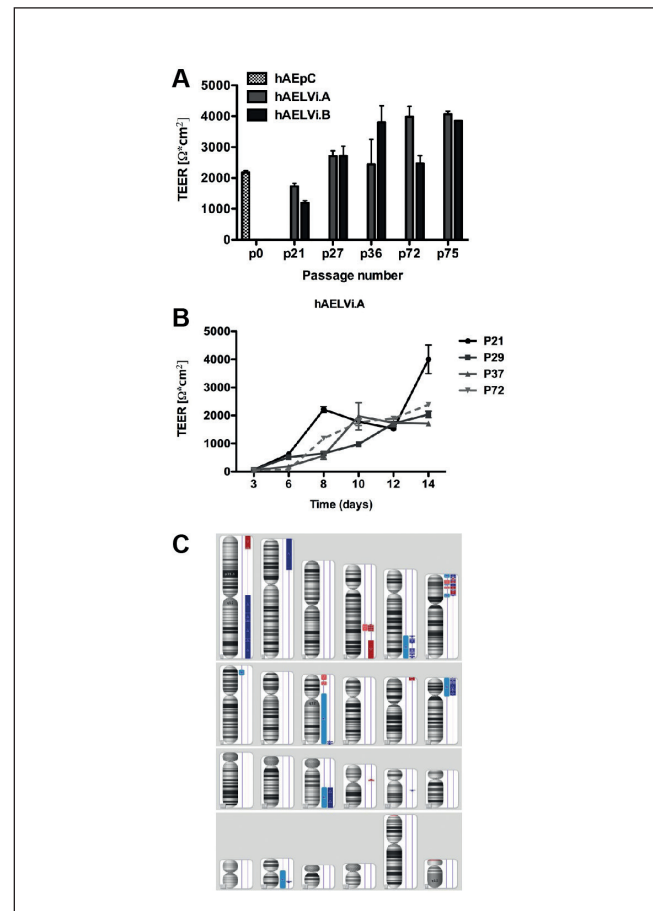
## Supplementary Data



**Fig. S1: Immortalization of hAEPc**

(A) PCR of recombinant genes on genomic DNA after lentivirus mediated transfection with immortalizing genes after 29 passages: 7 genes were identified: Id2, Id3, E7, Bcl2, Core, Myc and Nanog.

(B) TEER measurement of immortalized cells (hAELVi.A and hAELVi.B) during 15 days growing on Transwell® membranes.



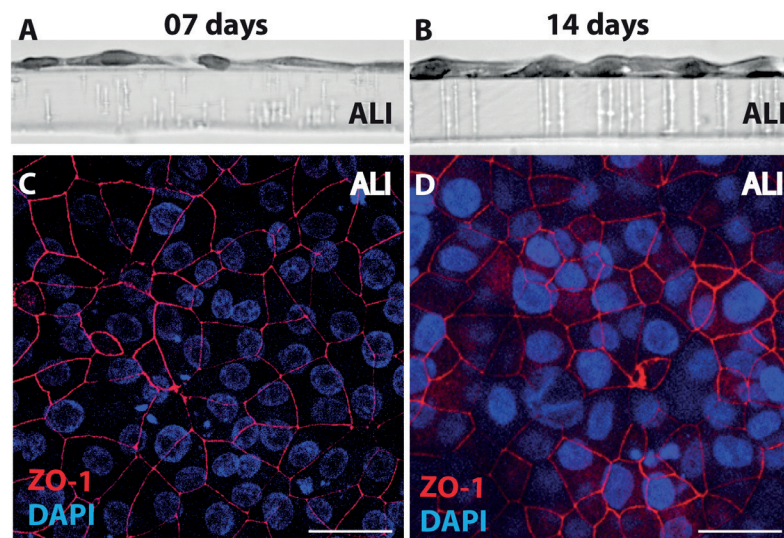
**Fig. S2: Barrier properties and chromosome profiling of growing hAELVi cell passages**

(A) TEER measurement of immortalized cells in different passages, after 15 to 30 days in culture, compared to primary ATI cells cultivated for 15 days in culture. (B) TEER curves of different passages of hAELVi.A cells during 14 days in culture. Data shown as mean  $\pm$  SEM (n = 3). (C) Chromosome profiling of hAELVi.A passage 43 was performed with Affymetrix Cytoscan HD array. Loss of a chromosome region is indicated as a red sign and duplication of a chromosome region is indicated as a blue sign.



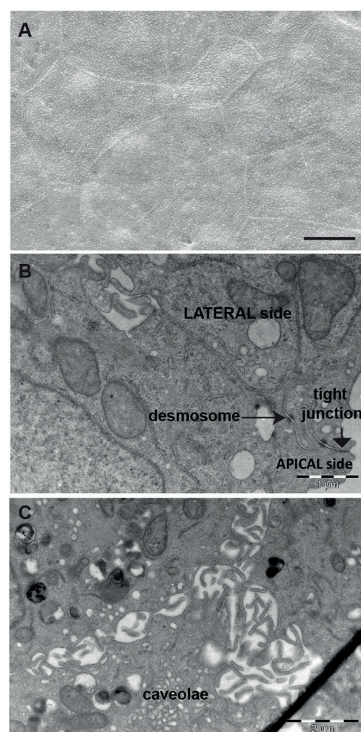
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<http://dx.doi.org/10.14573/altex.1511131s>



**Fig. S3: Morphology of hAELVi.B grown under air-liquid interface (ALI) conditions**

Samples were fixed after 7 days (A,C) or 14 days (B,D). (A,B) Histological cross-sections (100X). (C,D) Z-stacks of images from confocal laser scanning microscopy of hAELVi.B immunolabeled with anti-ZO-1 antibody (red). Cell nuclei are stained with DAPI (blue). Bar: 20  $\mu$ M.



**Fig. S4: Ultrastructure of hAELVi cells grown under air-liquid interface (ALI) conditions**

Scanning electron microscopy (A) and transmission electron microscopy (B,C) of hAELVi.B on Transwell® filters after 12 days; desmosomes (arrow), tight junctions (arrowhead) and caveolae are indicated in the TEM images. Bars (A: 5  $\mu$ M; B: 1  $\mu$ M; C: 2  $\mu$ M).