

Supplemental Fig. 2. Engineered L1 retrotransposition in ectodermal cells using the Ad-L1 virus. (A) Scheme and rationale of the Ad-L1 retrotransposition assay. See Figure 1 for further details. Below the scheme, representative data obtained in Ad-L1 infected HeLa cells is shown. The left panel shows a microscopy image of HeLa cells expressing L1-EGFP after retrotransposition; the middle panel shows a representative FACS histogram acquired 7 days post-infection in HeLa cells (SSC vs EGFP); the right panel shows a representative image of a βeta gal (blue signal) staining assay conducted on Ad-L1 infected HeLa cells. (B) Representative retrotransposition assays using the Ad-L1 in HeLa cells. Shown is a merged image of Ad-L1 infected HeLa cells analyzed 3 days after infection and stained with an antibody against BrdU (red) and EGFP (green); nuclear DNA was stained with DAPI (blue). The panel contains the individual captured images used in the merged picture. White bars, 20 mm. (C) Efficient Ad-L1 infection in hESCderived NPCs. A representative merged image is shown of Ad-L1 infected hESC-derived NPCs analyzed 5 days after infection and stained with an antibody against βeta gal (light blue) and EGFP (green); nuclear DNA was stained with DAPI (dark blue). The panel contains the individual captured images used in the merged picture. White bars, 20 mm. (**D**) Ad-L1 mediated retrotransposition in infected HFFs. An image of rare L1-EGFP expressing cells in Ad-L1 infected HFFs is shown. (E) Representative results from the PCR-intron assay conducted on gDNA isolated from Ad-L1 infected keratinocytes 7 days post-infection. In the PCR assay, pk87 cells were used as a positive control (see Methods). Genomic DNAs were restricted with Swa I prior to PCR. The amplification of a portion of the βeta gal expression cassette (present in the Ad-L1) served as a control to demonstrate efficient infection of keratinocytes.