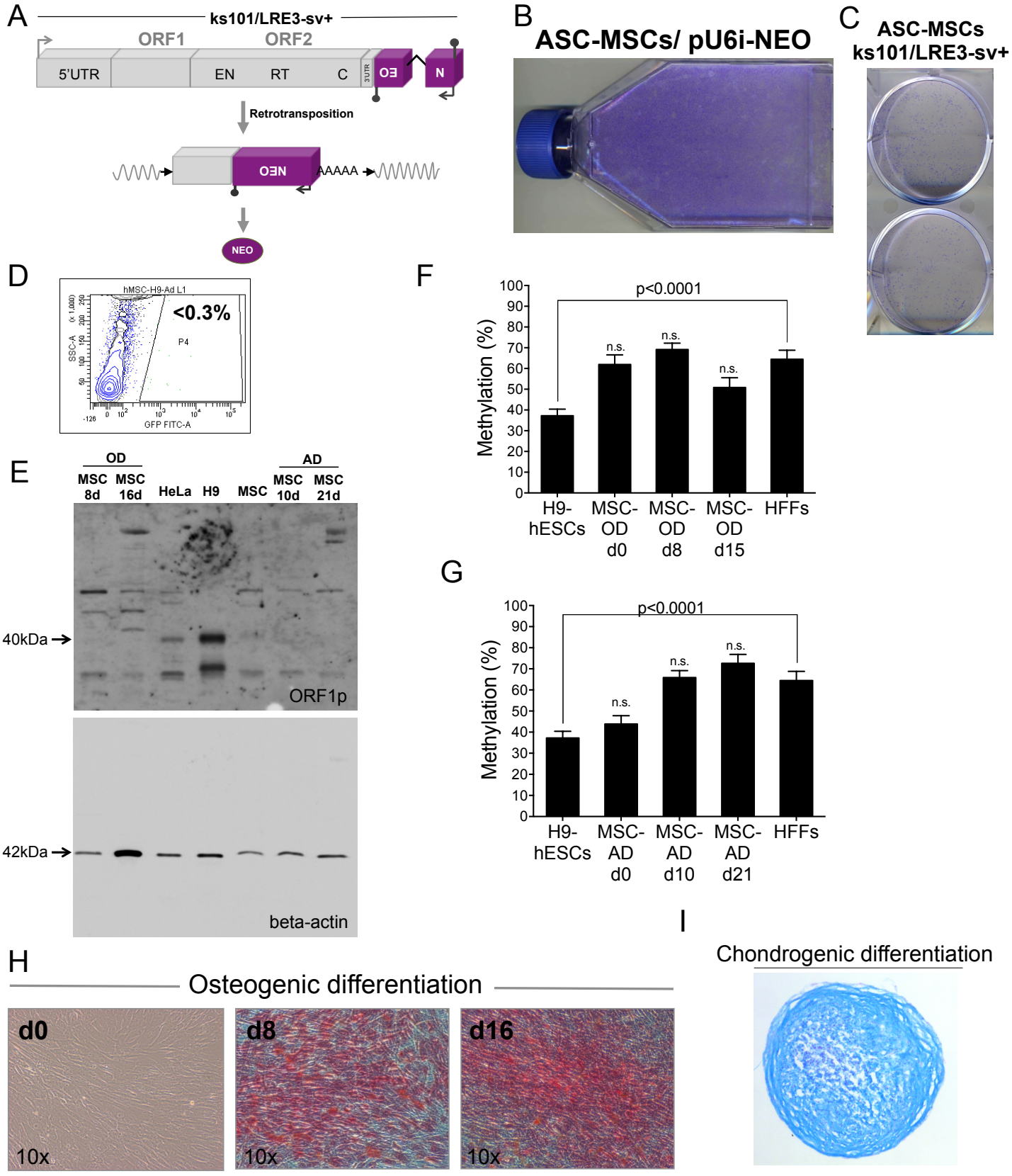

Supplemental Fig. 4. Engineered L1 retrotransposition in MSCs and differentiating MSCs. (**A**) Rationale of the retrotransposition assay using plasmids containing the *mneoI* retrotransposition indicator cassette. Briefly, cells transfected with plasmid ks101/LRE3-sv+ can activate expression of the *neomycin phosphotransferase* gene (*NEO*, purple oval) only after a round of L1 retrotransposition. (**B**) Transfection efficiency and clonability controls on MSCs. Cultured MSCs were transfected with a plasmid containing an expression cassette for the *neomycin phosphotransferase gene* (*NEO*, plasmid pU6i-NEO) and cells then were selected with G418. (**C**) Representative results of an L1-retrotranspositon assay using plasmid ks101/LRE3-sv+ in ASC-derived MSCs. (**D**) Representative

results of an L1-retrotranspositon assay using plasmid ks101/LRE3-sv+ in ASC-derived MSCs. (**D**) Representative FACS histogram analysis of hESC-derived MSCs infected with Ad-L1 and analyzed 7 days after infection. The percentage of EGFP-expressing cells is indicated within the histogram (SSC vs EGFP) as determined in triplicate. (**E**) Full-size Western blot scans of those shown in Figure 3F. (**F&G**) DNA methylation analyses in differentiating MSCs. The graph indicates the percentage of methylated CpG residues within L1Hs 5´UTR sequences as measured by bisulfite-PCR assays in the indicated sample during MSC differentiation. One-way ANOVA with Tukley was used as a statistical method. Also indicated is the p value of the comparisons (n.s., not significant). (**H**) A representative image of Alizarin Red staining at the indicated time (days) since the initiation of differentiation. (**I**) A representative image of Alcian blue staining of MSCs differentiated to chondrocytes after 21 days (see Methods). Note that a stained section is shown.