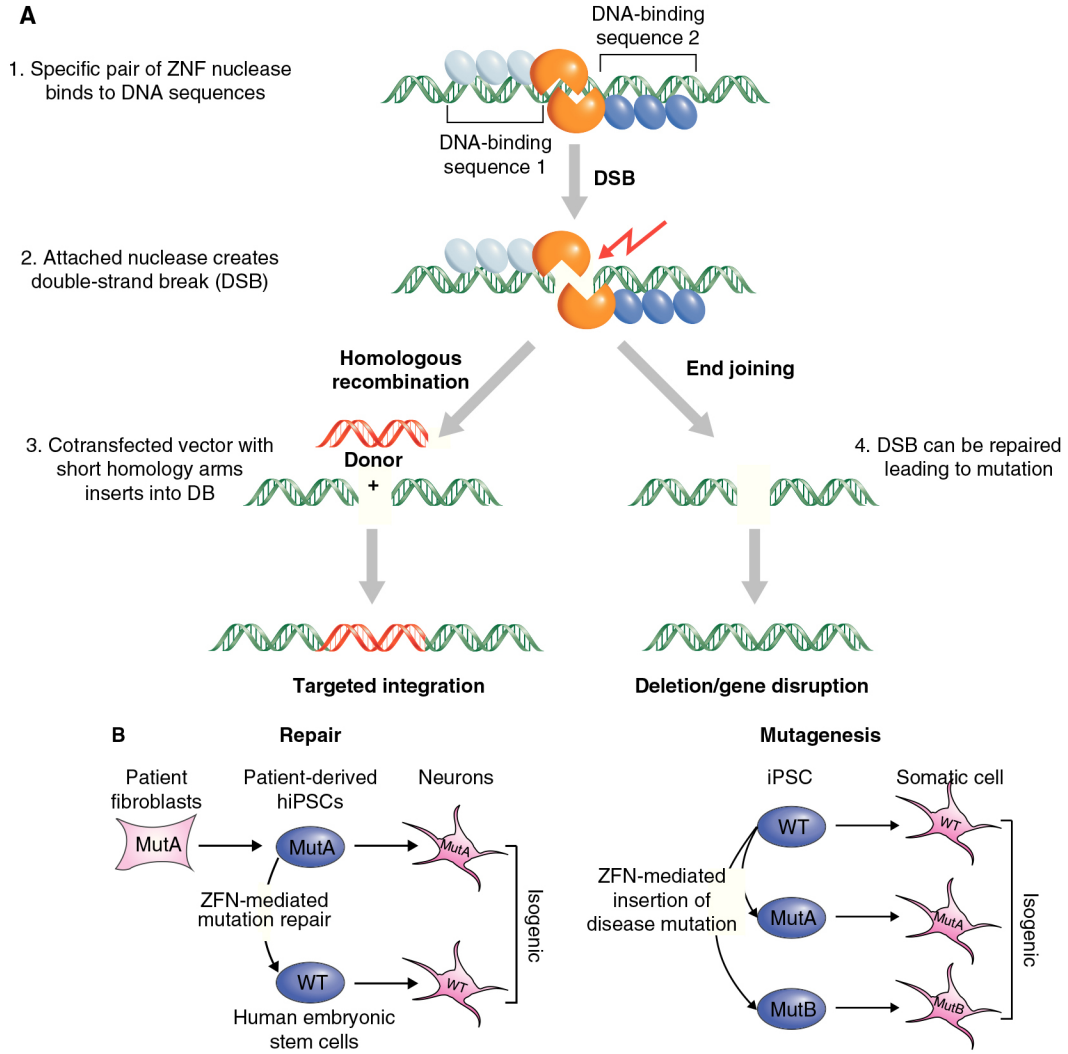

Figure 9. ZFN and TALE nuclease-mediated gene targeting. (*A*) (*1*) DNA-binding proteins—either zinc-finger or TALE proteins in blue fused to a Fok1 restriction nuclease in orange—are designed to specifically recognize two adjacent DNA-binding sequences with a defined spacing. (*2*) On binding of the zinc fingers, the FOK1 nuclease domains dimerize, become active, and cut the DNA. (*3*) If a donor plasmid carrying DNA (red, DNA) homologous to the DSB is ectopically provided to the cell, this can be used to repair the DNA lesion. A donor plasmid can be designed so that it carries additional sequence in between the homology arms. On repair of the DSB with such a donor, the genomic locus will be altered to carry this additional sequence as an insertion at the site of the DSB. (*4*) Alternatively, the DSB is repaired, incurring deletion or sequence alteration that disrupts gene function. (*B*) Using ZFN (or TALEN)-mediated gene targeting, a disease causing mutation is either corrected in a patient-derived iPSC (*left* illustration), or disease-causing mutations are introduced into wild-type (WT) ES cells (*right* illustration). The result of either manipulation will be the generation of isogenic sets of iPSCs, providing a genetically matched control for functional studies. (*B*, Adapted, with permission, from Soldner et al. 2011.)