Fig. 5. Otx2, Sox2 and Sox3 expression remains unchanged upon DNMT3A knock down. Embryos were injected with 3A MO on the embryonic right side. Morpholinos were FITC-labeled (green, small insets). DNMT3A knock down leaves (A) Otx2 (n=13/13), (B) Sox2 (n=7/7) and (C) Sox3 (n=7/8) expression unaltered. Dashed circles indicate position of otic domain. (D) Model of function of DNMT3A during otic development. In presence of DNMT3A, CpG islands in the putative repressor binding sites are methylated, preventing repressor binding to the target gene. When DNMT3A is decreased, methylation of repressor binding sites is reduced or not present. This allows repressors to bind and prevents target gene expression.

Fig. S1. Morpholino knock down of DNMT3A reduces otic expression of Sox10 and Soho1. DNMT3A MO (FITC-labeled, green, small insets) was electroporated into the right half of the embryo at HH4. (A) Sox10 RNA expression was reduced in the otic placode at HH11 (arrowhead) upon electroporation of DNMT3A MO1 (n=6/6) and DNMT3A MO2 (n=4/5). (B) Soho1 RNA expression was reduced on the DNMT3A injected side MO (n=3/3; 1.0 mM concentration).

Fig. S2. Tcf3 and Erm expression are unaltered upon DNMT3A knock down. DNMT3A MO or control MO were electroporated into the right or left half of the embryo, respectively, at HH4. Morpholinos were FITC-labeled (green, small insets). RNA levels of (A) Tcf3 (n=6/6) and (B) Erm (n=5/5) were not reduced on the DNMT3A MO injected side. Black line indicates level of section (A', B').

Arrowheads mark the position of the otic placode/vesicle on the 3AMO electroporated side.