

Fig. S1. Characterization of two enhancers for *Foxd3* in CNS and PNS cells. (A) *In situ* hybridization of *Foxd3* in a chick embryo at E3.5. Dorsolaterally migrating HNK-1+ melanocytes do not express *Foxd3* (arrowheads). (B) Cross-species alignment of the genomic region containing enhancers #168 and #169 from the Vista Enhancer browser, located 300-350 kb upstream of the *Foxd3*-coding region. The mouse sequence was aligned against orthologs in Rhesus, dog, chicken, *Xenopus tropicalis* and zebrafish showing an evolutionary conservation of the enhancers in the vertebrata genomes (top). Enlargement of the #168 enhancer (bottom). (C) E11.5 mouse embryos expressing β -galactosidase reporter activity under the control of either #168 or #169 enhancer elements. *lacZ* signal in the PNS is evident only with the #168. Images were taken from the Vista enhancer browser (<http://enhancer.lbl.gov>). (D) Electroporation of the #169 reporter (169::Cre/CAGG-LoxP-STOP-LoxP-GFP) along with control RFP showing 24 hours after transfection GFP+ cells only in CNS, while control RFP signal is evident in both CNS and PNS. (E) The #168 reporter construct (168::Cre/CAGG-LoxP-STOP-LoxP-GFP) was electroporated at E3 into hemi-NTs. One day later, progenitors expressing the #168 reporter (green) co-express *Foxd3* (blue), which is transcribed at this stage in specific CNS subpopulations. Scale bars: 150 μ m in A; 100 μ m in D; 80 μ m in E.

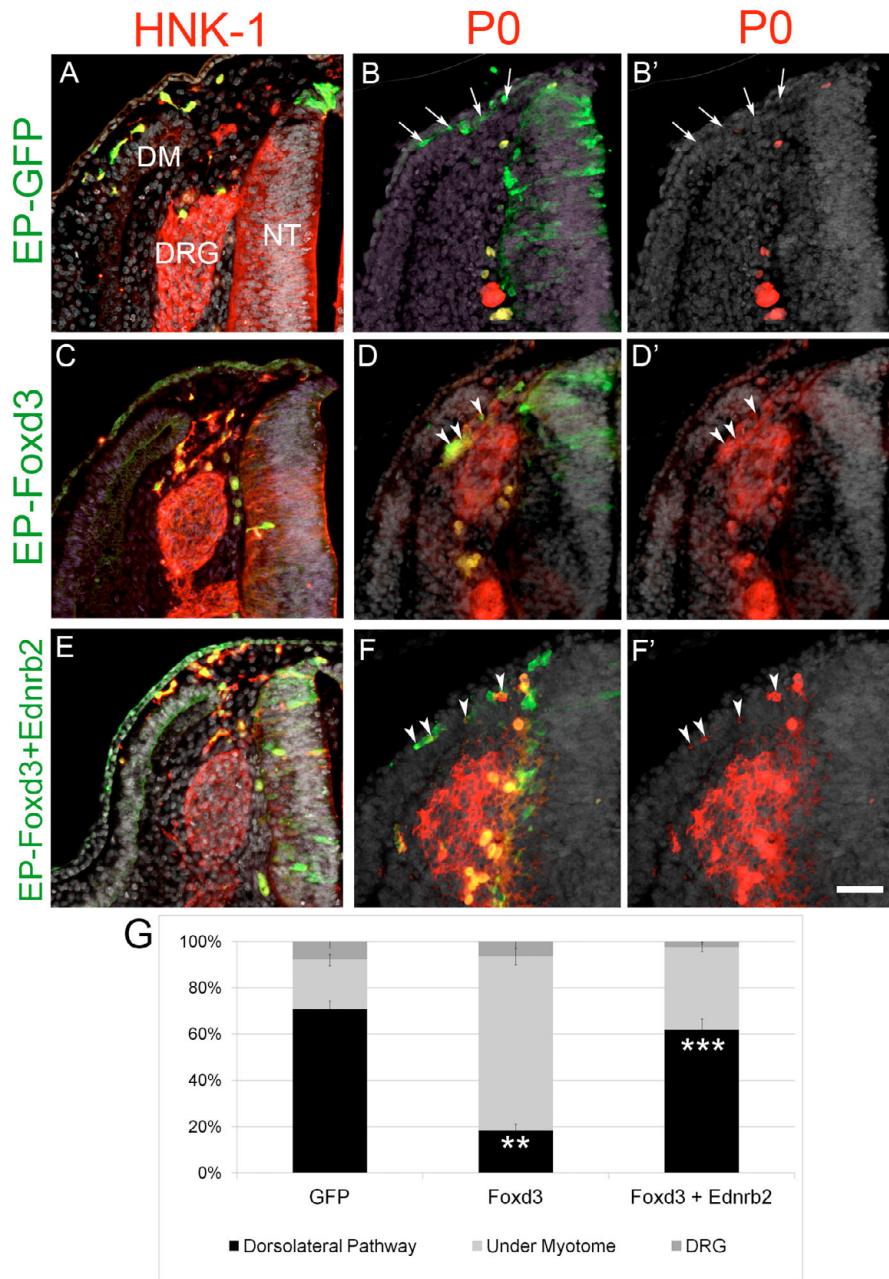


Fig. S2. Ednr2 rescues dorsolateral migration but not fate of late Foxd3-transfected cells. (A-B') Electroporation of control GFP into the late NT (35 ss, flank level) yields NC cells that migrate along the subectodermal path between ectoderm and dermomyotome (DM) that are P0 negative (arrows). (C-D') Transfection of Foxd3 shifts migration towards the ventral pathway underneath the DM and cells upregulate P0 (arrowheads). (E-F') Co-electroporation of Foxd3 and Ednr2 results in invasion of the subectodermal pathway by cells expressing P0 (arrowheads in F,F'). (G) Quantification of the percentage of NC cells in different locations. Very few NC cells colonize the DRG in the late-transfected embryos. Foxd3 reduces the percentage of cells in the dorsolateral path when compared with control GFP (** $P < 0.01$). Ednr2, in conjunction with Foxd3, restores dorsolateral migration compared with Foxd3 alone (** $P < 0.005$). Scale bars: 75 μm in A,B,E; 80 μm in C,D; 70 μm in F.

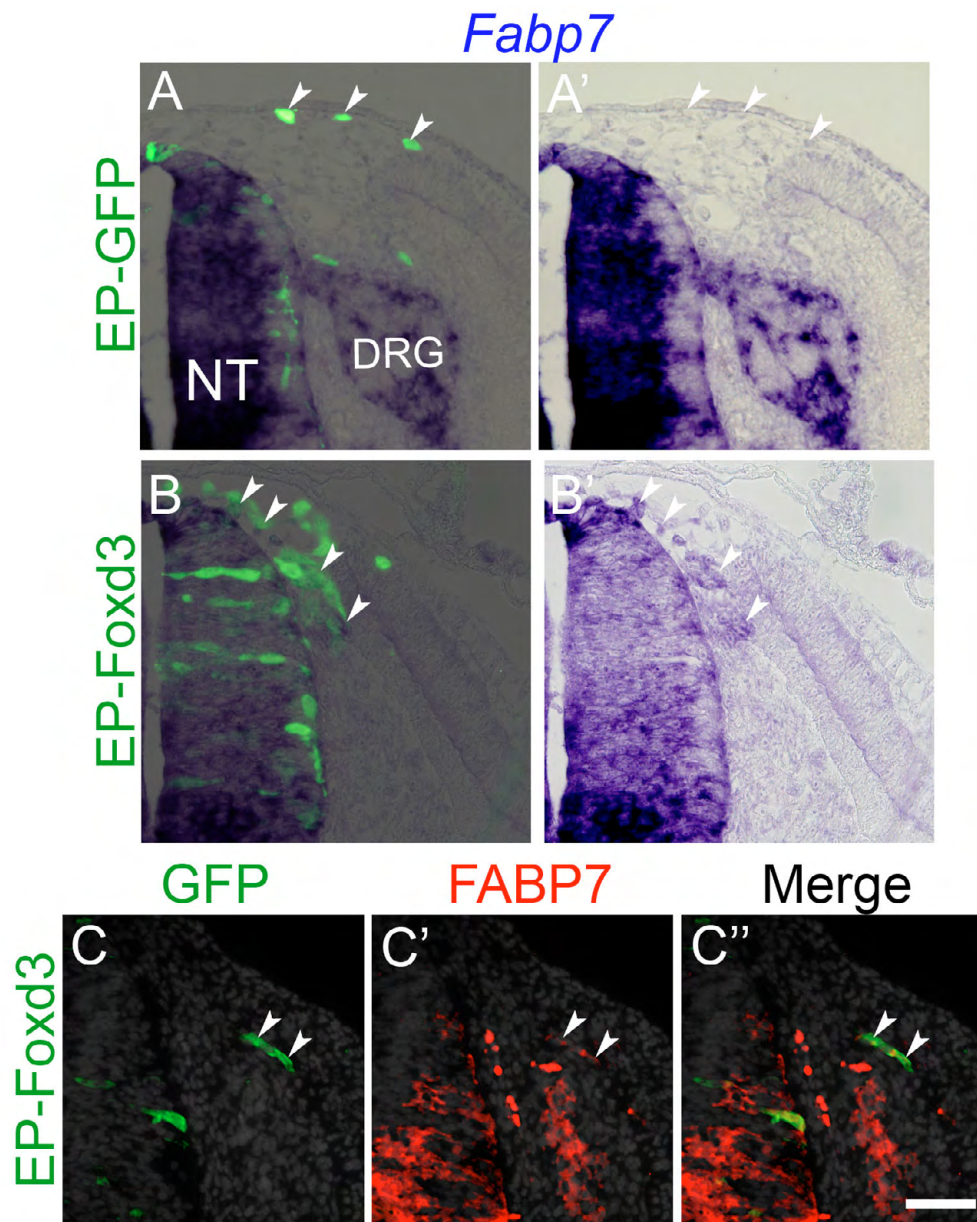


Fig. S3. Foxd3 induces FABP7 expression in prospective melanocytes. (A,A') Twenty-four hours after electroporation of control GFP into the NT of 35 ss embryos, GFP+ cells are found in the dorsolateral pathway and are negative for *Fabp7* mRNA. (B-C'') Similar electroporation of Foxd3 results in upregulation of Fabp7 at both mRNA (B,B') and protein (C-C'') levels in migrating NC cells forced by Foxd3 misexpression to migrate through the ventral path. Scale bars: 75 μ m in A-B'; 85 μ m in C-C''.

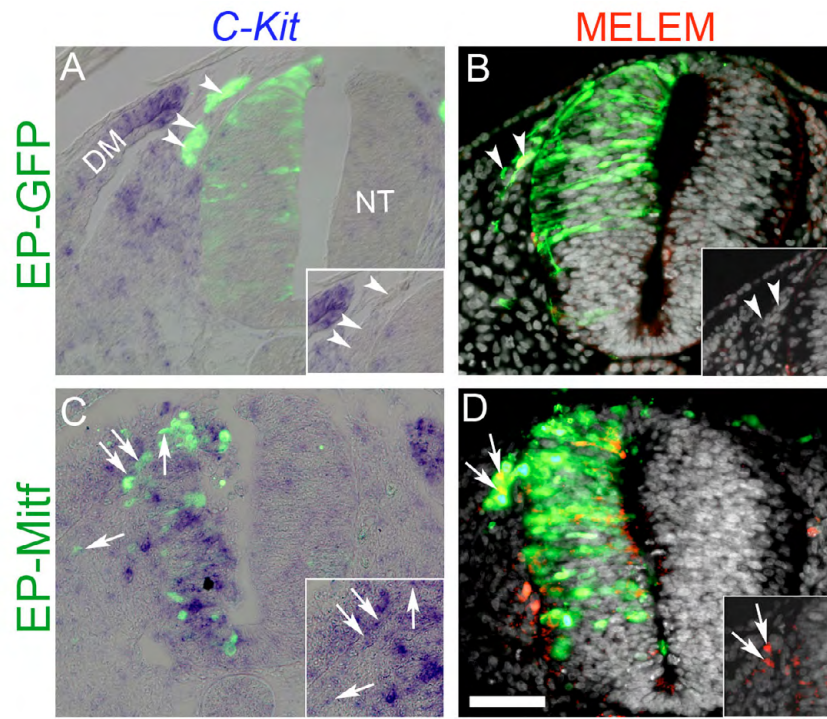


Fig. S4. Mitf reprograms neural progenitors into melanocytes without altering their original dorsoventral migration. (A,B) Electroporation of control-GFP into the early NT yields NC cells that migrate dorsoventrally medial to the dermomyotome (DM) and are negative for the melanocytic markers *Kit* and MELEM (arrowheads). **(C,D)** Electroporation of Mitf-GFP upregulated both *Kit* and MELEM (arrows) without affecting migration. Scale bars: 50 μ m in A,C; 55 μ m in D; 60 μ m in B.

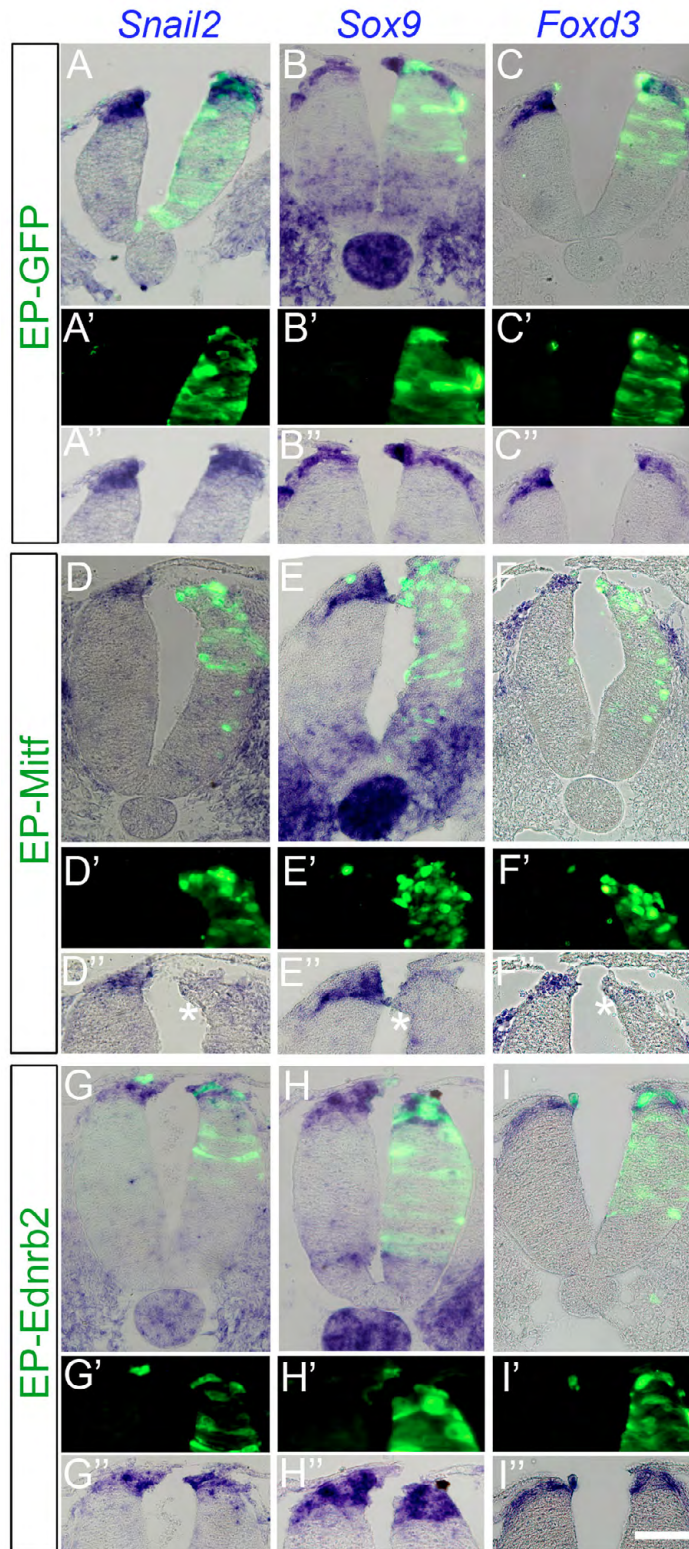


Fig. S5. Mitf, but not Ednrb2, inhibits transcription of *Snail2*, *Sox9* and *Foxd3* in the dorsal NT. (A-C'') Co-expression of electroporated control-GFP and *Snail2*, *Sox9* or *Foxd3* in the dorsal NT. There are symmetric bilateral expression patterns. (D-I'') Mitf inhibits transcription of the three genes in the transfected sides (D-F'', asterisks), whereas Ednrb2 has no effect (G-I''). Scale bars: 50 μ m in A-A'', D-D'', F-F'', I-I''; 45 μ m in B-C'', E-E'', G-G''; 40 μ m in H-H''.

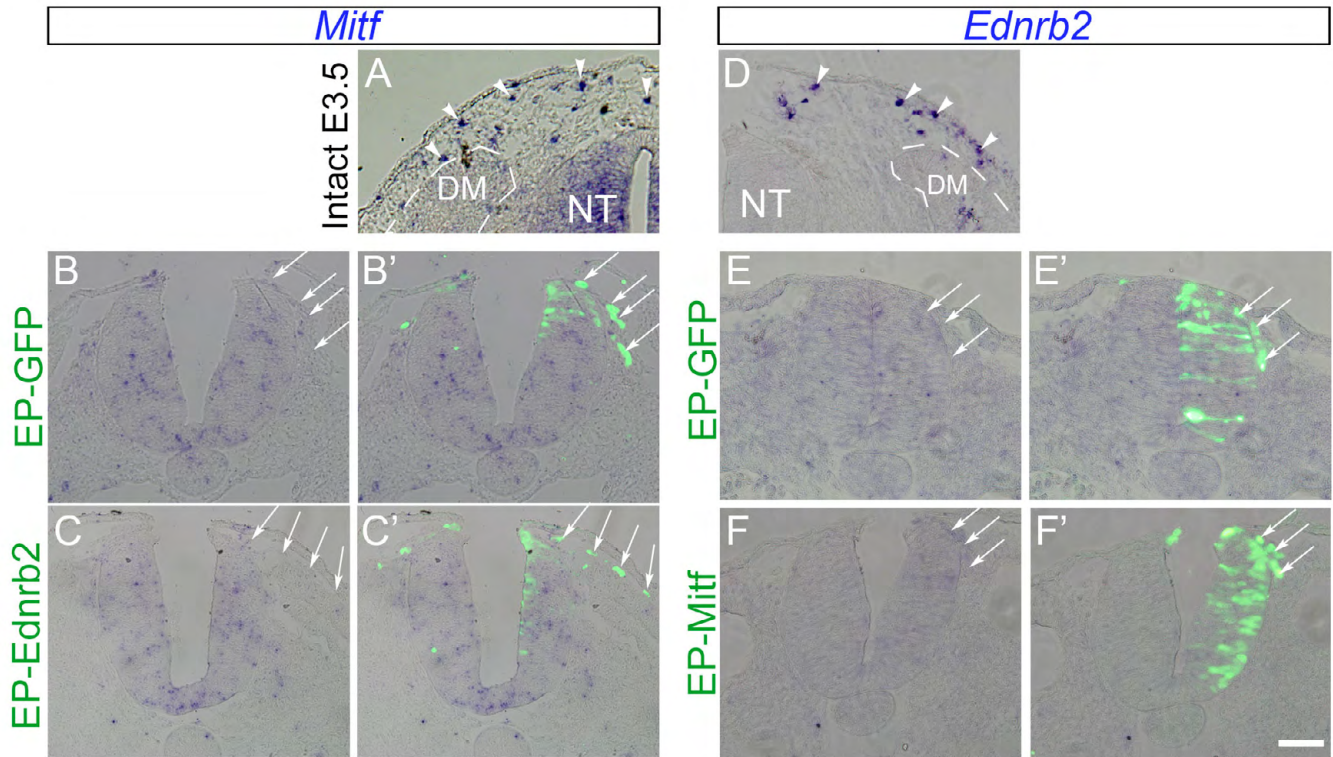


Fig. S6. Ednrb2 does not upregulate *Mitf* transcription and vice versa. (A,D) Expression of *Mitf* and *Ednrb2* mRNAs in normal melanocytes (arrowheads) at E3.5. (B-C') Early electroporations of either control GFP or *Ednrb2* do not upregulate *Mitf* transcription in either hemi-NT or emigrating NC. Control labeled cells migrate dorsoventrally, whereas *Ednrb2*-misexpressing cells were diverted into the subectodermal path (C,C', arrows). (E-F') Early electroporation of control GFP does not affect *Ednrb2* mRNA, whereas *Mitf* marginally enhances its transcription (F,F', arrows) compared with the normal levels of expression (D). Scale bars: 50 μm in A,C,C',D,E,E'; 60 μm in F,F'; 75 μm in B,B'.