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SUPPLEMENTARY MATERIAL

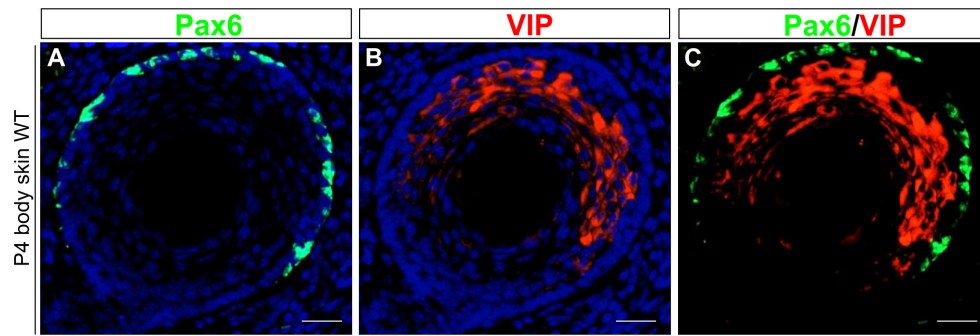
corresponding to:

Regulation of Merkel cell development by Pax6

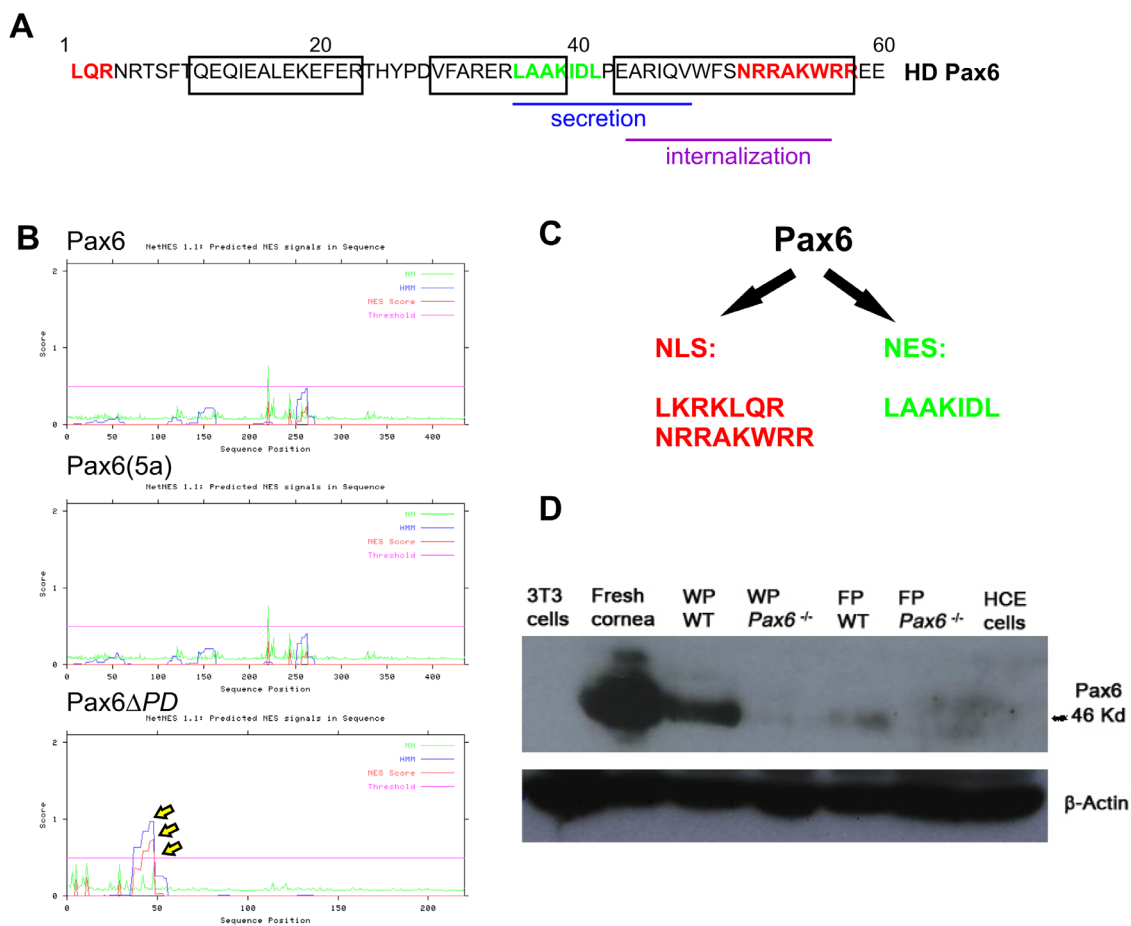
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Supplementary Fig. S1. VIP expression was not detected in embryonic and neonatal Merkel cells in mouse. (A-C) *Vasoactive intestinal peptide (VIP)* expression was analyzed within postnatal (P4) Merkel cells and was detected within the external root sheath cell of hair follicles from the body skin (B,C). However it did not overlap Pax6 (A,C) thus indicating that VIP is not expressed within the P4 Merkel cells in mouse. Scale bar: 20 μm .



Supplementary Fig. S2. In silico prediction of nucleo-cytoplasmic shuttling of Pax6 protein. (A) Previous studies suggested the presence of internalization and secretion signals within the homeodomain of Pax6 (La Saffre et al., 2007). **(B)** To extend the data from the present study suggesting that Pax6 is an active nucleo-cytoplasmic shuttling protein within the Merkel cells, in silico analysis of Pax6 isoforms sequence was carried out. The NES prediction server (<http://www.cbs.dtu.dk/services/NetNES/>) was used for analyzing putative residues participating to the formation of a NES in the three isoforms of Pax6. The NES is only predicted in the Pax6 Δ PD isoform (indicated by the yellow arrows), although the same amino acid sequence is present in all the three isoforms. **(C)** The predicted NES and NLS signals within the homeodomain of Pax6. **(D)** Western Blot analysis of the whisker pad (WP WT) and foot pad (FP WT) from E18.5 wild-type embryos, suggested that the Merkel cells express the major isoform of Pax6 containing both homeodomain and paired domains. Pax6 expression was also detected within the foot pad (FP WT), and thus within the glabrous skin, although at lower levels compared to the whisker pad (WP WT). This finding suggested that Merkel cells distributed in different body locations (whisker follicles, body skin and foot pad) share Pax6 expression. As expected, Pax6 protein was undetectable in both the whisker pad (WP Pax6^{-/-}) and foot pad (FP Pax6^{-/-}) from Pax6^{-/-} mutant embryos. Fresh corneas were used as positive control for Pax6 expression, while 3T3 fibroblasts and HCE (human corneal epithelial cells) cell lines, which do not express Pax6, were used as negative controls.