VPS33B and VIPAR are essential for epidermal lamellar body biogenesis and function

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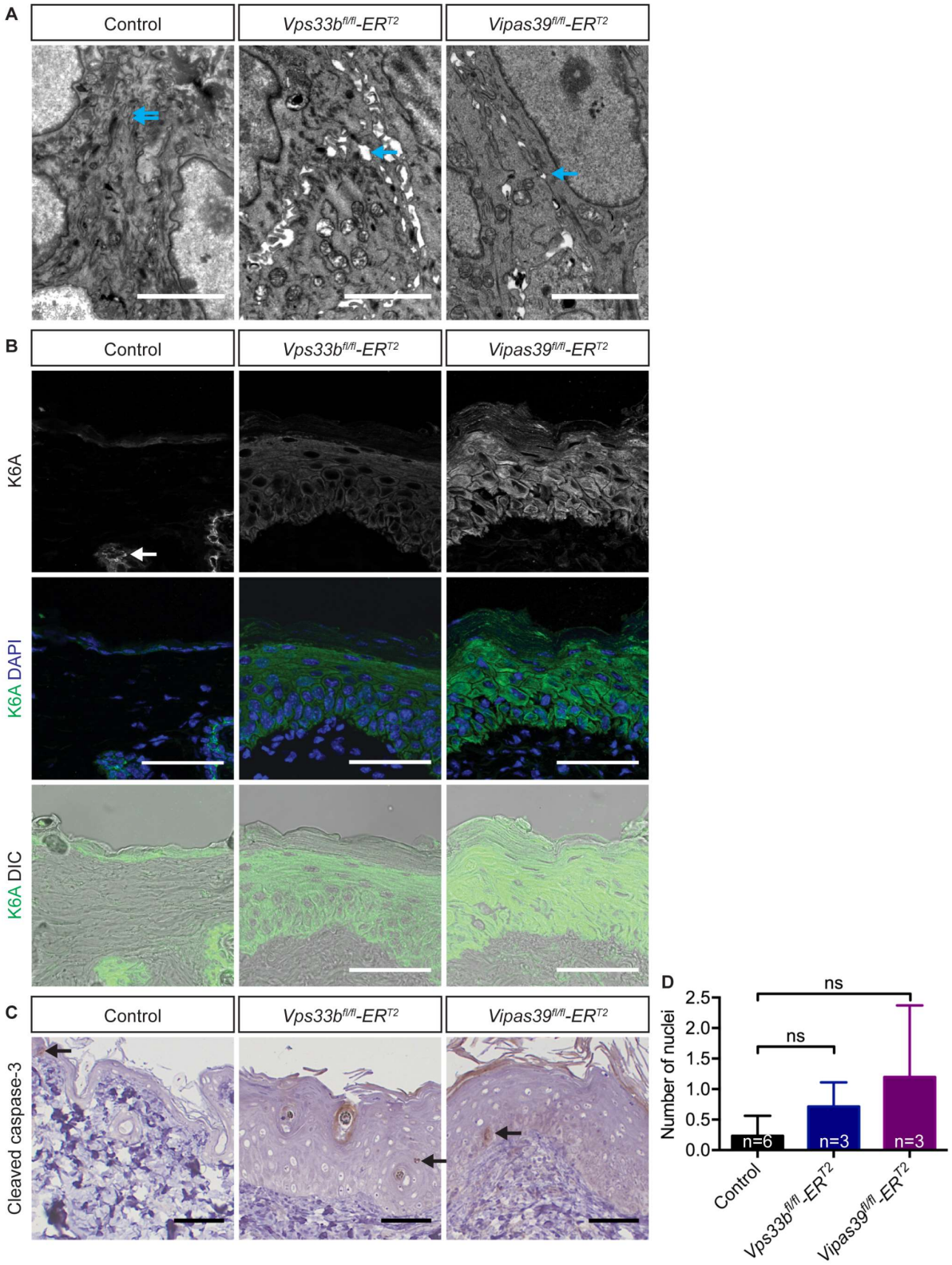
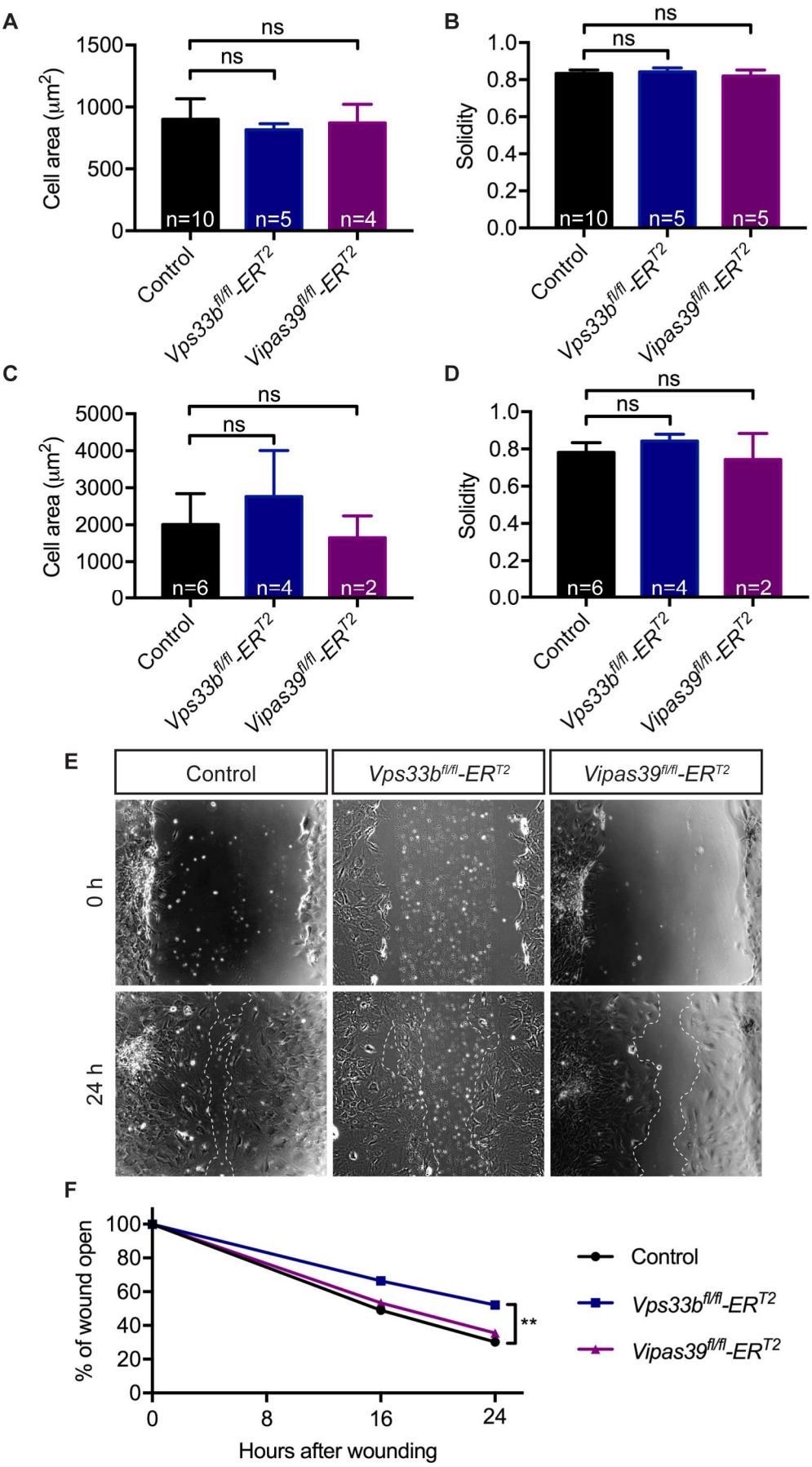


Supplementary Figure 1– Vps33b and Vipar deficient murine epidermis shows spongiosis, increased expression of hyperproliferative keratin K6A and no change in the expression of cleaved caspase-3 apoptotic marker. A - TEM images of control, *Vps33b*$^{fl/fl}$*ER*^{T2} and *Vipas39*$^{fl/fl}$*ER*^{T2} murine biopsies. Normal cell-cell contact (double arrow), wider gaps (single arrow). Scale bars = 5 µm. Images are representative of two independent mouse biopsies. B - K6A staining of control, *Vps33b*$^{fl/fl}$*ER*^{T2} and *Vipas39*$^{fl/fl}$*ER*^{T2} skin sections, counterstained with DAPI. Hair follicles (white arrow). Scale bars = 50 µm. Images are a maximum projection of a z-stack. DIC images are a single z-slice image. Images are representative of results from at least six control and three *Vps33b*$^{fl/fl}$*ER*^{T2} and *Vipas39*$^{fl/fl}$*ER*^{T2} independent murine biopsies.C – Immunohistochemical staining for cleaved caspase-3 in control, *Vps33b*$^{fl/fl}$*ER*^{T2} and *Vipas39*$^{fl/fl}$*ER*^{T2} epidermis. Cleaved caspase-3 positive cells (black arrow). Scale bars = 100 µm. Images are representative of results from at least six control and three *Vps33b*$^{fl/fl}$*ER*^{T2} and *Vipas39*$^{fl/fl}$*ER*^{T2} independent murine biopsies. D - Number of cleaved caspase-3 stained nuclei per mm of epidermis. Three fields of view analysed per mouse, number of mice analysed are indicated. Data are mean ± standard deviation, ordinary one-way ANOVA, non-significant (ns) p>0.05.



Supplementary Figure 2 - Cell spreading and wound healing are not substantially affected in Vps33b and Vipar deficient mice. A - Keratinocyte area after 1 h on fibronectin. B - Keratinocyte solidity after 1 h on fibronectin. C - Fibroblast area after 1 h. D - Fibroblast solidity after 1 h. For cell spreading at least three fields of view were measured per isolation, number of isolations analysed (n) are indicated in three independent keratinocyte and two independent fibroblast experiments. E - Representative images of wound healing experiments of control, *Vps33b$^{fl/fl}$ER^{T2}* and *Vipas39$^{fl/fl}$ER^{T2}* fibroblasts. F - Percentage wound closure in control, *Vps33b$^{fl/fl}$ER^{T2}* and *Vipas39$^{fl/fl}$ER^{T2}* wound healing assays. Results are representative of experiments from at least three independent fibroblast isolations. Data are mean ± standard deviation, ordinary one-way ANOVA, * p≤0.05, ** p≤0.01, all other comparisons were non-significant (ns) p>0.05.