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Grp1-associated scaffold protein regulates skin homeostasis after ultraviolet irradiation

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Supplementary Information

Table S1: Sequences of primers and probes used in southern analysis of homologous recombination (HR) at the *Grasp* locus (Figure 1) using long-template PCR

	Sequence (5' → 3')
A1	CAGACAGCCATGCAGAAAG
A2	TITATATGCTACCTGGAAGCAGT
P1	AGCTACTAGTATAACTTCGTATAGCATACATTATACGAAGTTATTCTAGAC
P2	GACTTCTGTCTGAAGTCAGTGGCA
B1	TGGAGCAGGGATAGAGAAGGAAC
B2	GGCTACATCAAGGGCAGTCAG
P3	CAAAGAACGGAGCCGGTTG
P4	GGCAGAATCTGGTAGAGAAG

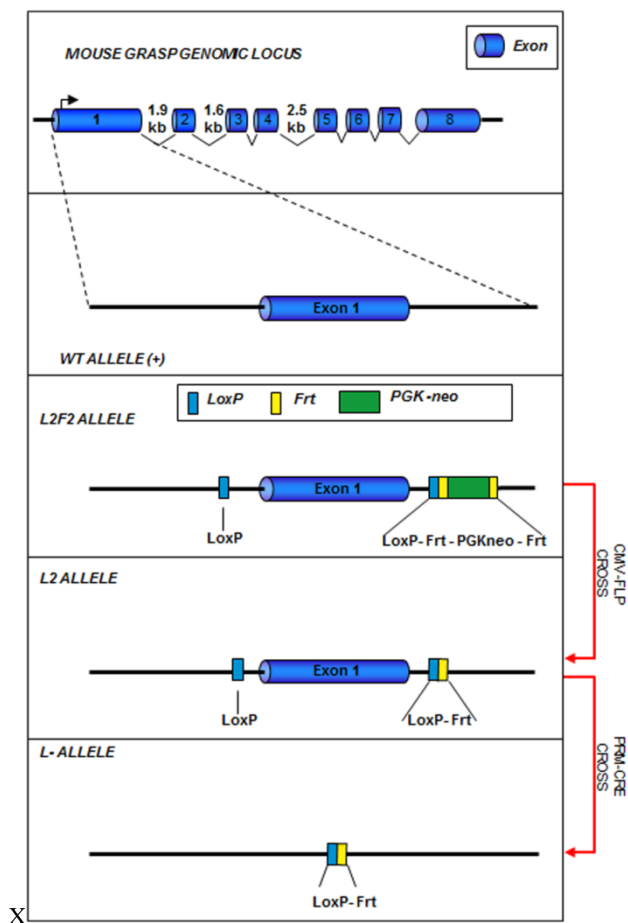


Fig. S1: Schematic representation of *Grasp* locus, targeting vector, and L2F2, L2 and L- *Grasp* alleles. LoxP and Frt sites are as indicated and the appropriate mouse lines used to breed L2F2 mice to L2 and eventually into the L- *Grasp* are also indicated.

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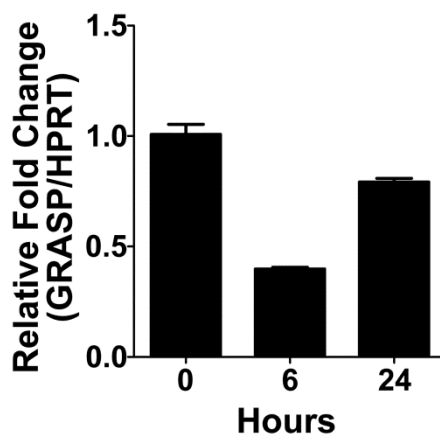


Fig. S2: *Grasp* is not induced in the keratinocyte culture following UVB exposure. RT-qPCR analysis of *Grasp* transcripts in cultured skin keratinocytes prepared from wt mice and collected at indicated times after UVB exposure. The reduction in *Grasp* expression at both 6 and 24 hrs was statistically different from time zero ($p < 0.001$).

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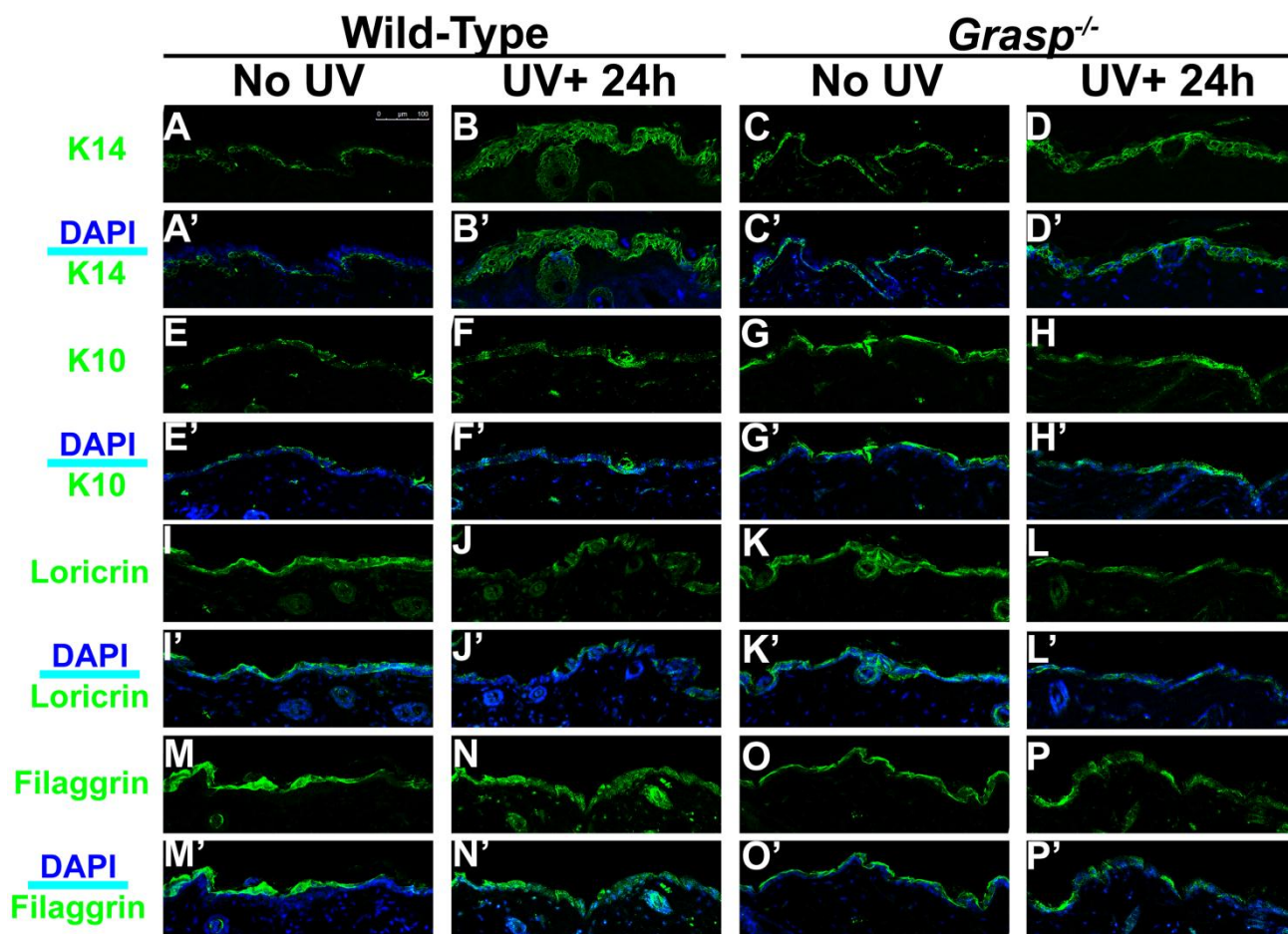


Fig. S3: Expression characteristics of epidermal markers in *Grasp*^{-/-} mice are comparable to control wt mice at the indicated times of UVB exposure. (A-P) Immunohistochemical analysis comparing expression of the proliferation K14 (green) and differentiation markers K10, Loricrin, and Filaggrin (green) with Hoechst 3342 DNA counterstain (blue; A'-P'). We observed an increased staining for K14 in wt mice but not the *Grasp*^{-/-} after 24hrs of UVB exposure. Note the presence of differentiation proteins distributed normally to suprabasal epidermis in *Grasp*^{-/-} tissue. These images were captured on a Leica TCS SP8 Confocal with a using a 20x objective. Scale bars, 100 μm.

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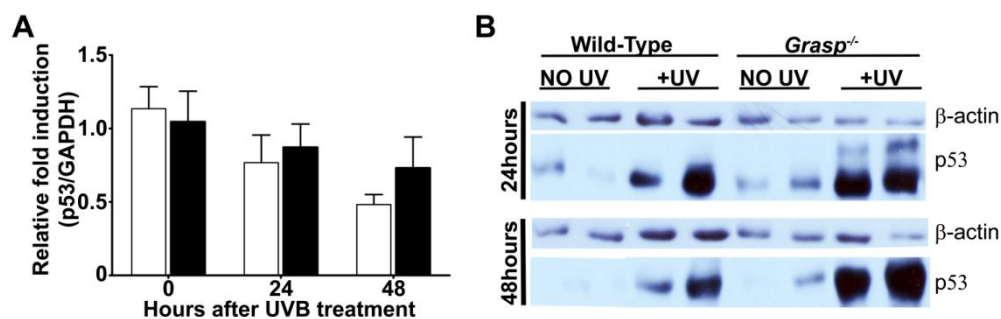


Fig.S4 The steady-state levels of the p53 transcript and the induced levels of the p53 protein after UVB exposure are comparable between the wt and *Grasp*^{-/-} mice. (A) RT-qPCR was performed to assess p53 transcript levels in skin samples obtained from wild-type (white bars) and *Grasp*^{-/-} (solid bars) mice at indicated times after UVB exposure. Values in (A) represent the mean \pm S.E.M. of three mice of both genotypes at each time-point. (B) Immunoblot analysis of p53 protein was performed in skin lysates obtained from wt and *Grasp*^{-/-} mice at indicated times after UVB exposure.