





Figure S2: No significant difference in Neutrophils or Macrophages between tumors treated with SB431542 or vehicle and between skin treated with chronic UV, SB431542 or vehicle

Leukocytes were isolated from tumors arising in UVB irradiated mice treated with vehicle or SB431542 at 30 weeks. Multicolor flow cytometry was used to immunophenotype myeloid cells by gating for CD11b⁺ ly6G⁺ and F4/80⁺. (A) Percentage of Neutrophils (CD11b⁺ Ly6G⁺) and (B) Percentage of Macrophages (F4/80⁺ CD11b⁺ Ly6G⁺) in total viable CD45⁺ leukocytes. N= at least 7 tumors per group. SKH-1 mice were treated with 10 μ M SB43152 or acetone (V) everyday with UVB treatment on alternative days for 1 week. Skin was harvested 40 hr post last UVB treatment. (C) Percentage of Neutrophils (CD11b⁺ Ly6G⁺) and (D) Percentage of Macrophages (F4/80⁺ CD11b⁺ Ly6G⁺) in total viable CD45⁺ leukocytes. Error bars = ±SEM, ns= not significant.



CD45





Figure S3. SB431542 suppresses UVB-induced skin inflammation.

SKH1 mice were irradiated with 1 MED UVB 3X on alternate days with or without topical acetone (vehicle) or SB431542 pretreatment, and skin was harvested after 1 week. (A) anti-CD45 immunohistochemistry on vehicle or SB431542 treated skin. (B) Anti-CD3 immunohistochemistry on vehicle or SB431542 treated skin.





SKH-1 mice were treated with 10 μ M SB43152 or acetone (V) everyday with UVB treatment on alternative days for 2 weeks (6 total UVB exposures). Lymph nodes and skin were harvested 40 hr post last UVB treatment and IFN γ expression in CD4 and CD8 T cells (A, B) or skin CD4 T cells (C) was analyzed by flow cytometry.



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Figure S5: UVB and SB treatment do not change the phospho-Smad2 status of Langerhans cells SKH-1 mice were pretreated with 10 μ M SB43152 or acetone (V) for 1 hour followed by UVB treatment at MED dose. LCs were isolated from the epidermis 2 hrs after UVB and stained for p-Smad2 and analyzed by FACs. (A)Percentages of phospho-Smad2⁺ LCs from UV treated or untreated epidermis gated on MHCII^{high} CD11c⁺ cells. (B) Mean Fluorescence Intensities (MFI) of phospho-Smad2 staining in LCs from UV treated or untreated epidermis.



Figure S6: Topical ALK5 inhibition suppresses chronic UVB-induced migration of dermal DC subsets

SKH-1 mice were treated with 10 μ M SB43152 or acetone (V) everyday with UVB treatment on alternative days for 1 week. Lymph nodes were harvested 40 hr post last UVB treatment and multicolor flow cytometry was used to immunophenotype dendritic cells by gating for MHCII^{high} CD11c⁺ cells, B220⁻ and CD8⁻. (A) Cell numbers of the CD207⁻ dDC subset in SDLN. (B) Cell numbers of the CD207⁺ CD103⁺ dDC subset in the SDLN. ns= not significantly different.



Figure S7: Effect of SB431542 on UVB induced migration of Langerhans cells

(A) Numbers of CD207⁺ CD103⁻ DC subset in the SDLN of SKH-1 mice 72 hrs after 1 MED UVB IR dose with 10 μ M SB43152 or acetone (V) pretreatment. (B) Numbers of CD207⁺ CD103⁻ DC subset in media after 72 hrs of ear explant culture. Ears were harvested immediately after a single MED UVB with 10 μ M SB43152 or acetone (V) pretreatment split and floated in complete RPMI media and cultured for 72 hrs. (C) LC numbers in SDLN of mice exposed to UVB IR for 1 week. SKH-1 mice were treated with 10 μ M SB43152 or acetone (V) everyday with UVB treatment on alternative days for 1 week. Lymph nodes were harvested 40 hr post UVB IR and multicolor flow cytometry was used to immunophenotype the dendritic cells.



Figure S8: UV-induced migration of CD207subset is suppressed in ear explant culture of CD11c-DNR mice.

Ears of CD11c-DNR (DNR) and wildtype (WT) mice were harvested immediately after a single MED UVB and split and floated in complete RPMI media and cultured for 72 hrs. Cell numbers of the CD207-subset in media of ear explant culture are shown.



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Figure S9 Langerhans cell migration in UVB irradiated wildtype and CD11c-DNR mice (A) Lymph node cell numbers of CD207⁺ CD103⁻ DC subset in CD11c-DNR transgenic mice (DNR) or wildtype (WT) 72 hrs post UVB. (B) Cell numbers of CD207⁺ CD103⁻ DC subset in media after 72 hrs of ear explant culture with and without UVB irradiation.

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