

Tristetraprolin expression by keratinocytes protects against skin carcinogenesis

Assabban *et al*, Supplementary figures

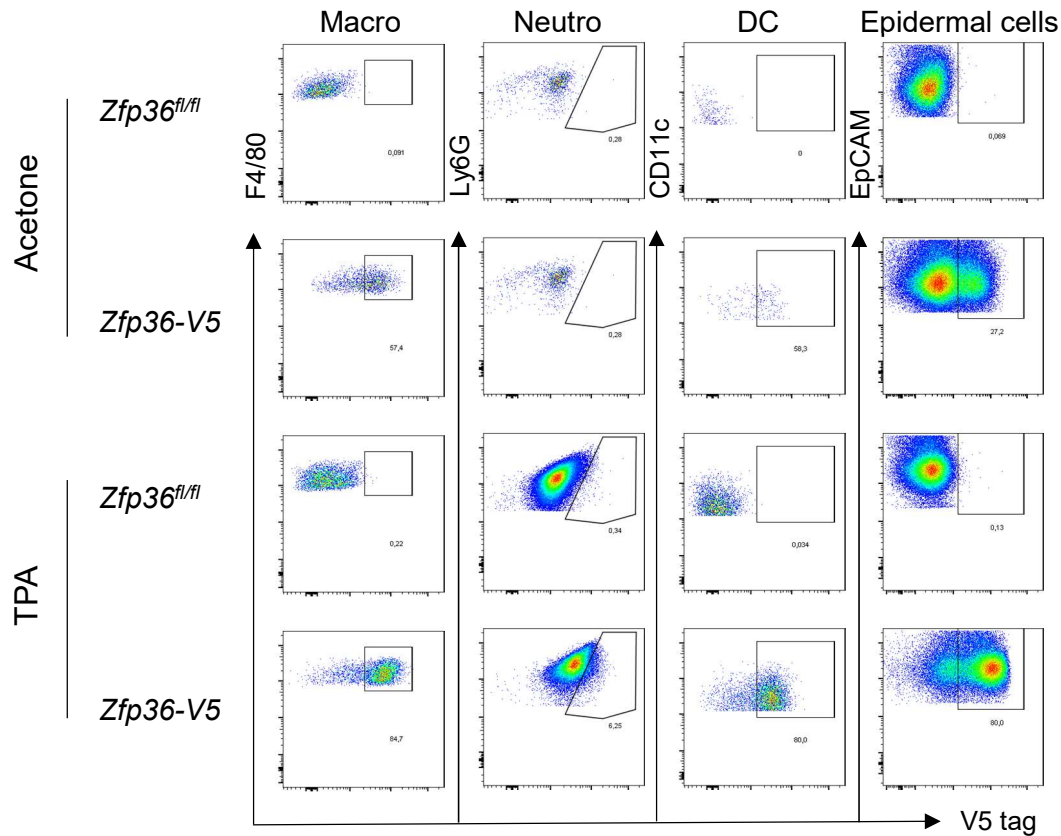
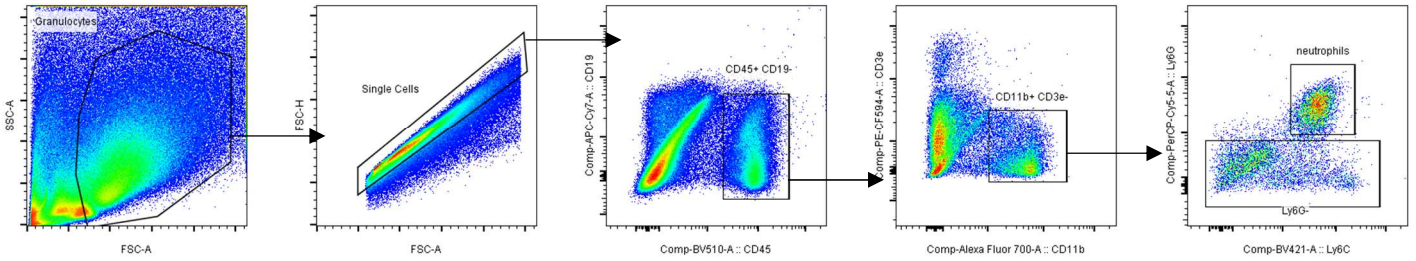


Figure S1 (related to Figure 2A): Gating strategies for skin cell populations after acute inflammatory stimulation. *Zfp36-V5* and *Zfp36^{fl/fl}* mice were treated for 3 days with TPA or acetone. Representative skin immune cells were gated based on the following gating strategy: MHCII^{high} macrophages (CD45⁺CD19⁻CD3 ϵ ⁻CD11b⁺Ly6G⁻F4/80⁺MHCII^{high}), neutrophils (CD45⁺CD19⁻CD3 ϵ ⁻CD11b⁺Ly6G⁺) and DCs (CD45⁺CD19⁻CD3 ϵ ⁻CD11b⁺Ly6G⁻F4/80⁻CD11c⁺MHCII⁺). Keratinocytes were gated based on CD45⁻CD140a⁻CD31⁻EpCAM⁺ staining.

Neutrophils



$\gamma\delta$ T cells

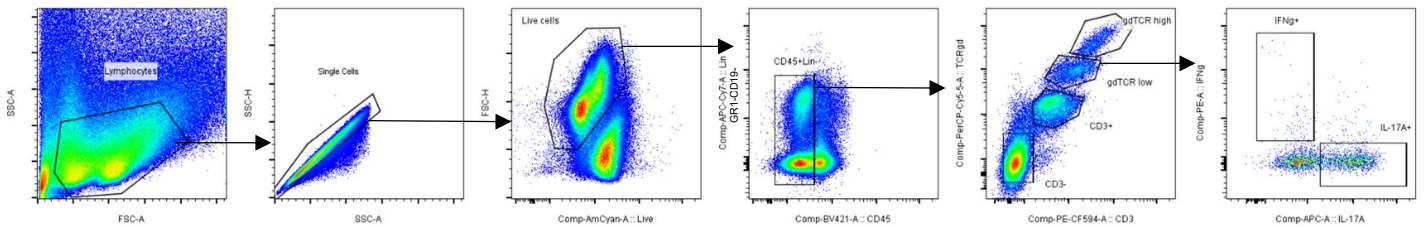


Figure S2 (related to Figure 2D): Gating strategies for skin immune cell populations after DMBA/TPA treatment. *Zfp36^{ΔEP}* mice and their littermate controls (*Zfp36^{fl/fl}*) were topically treated on back skin with DMBA/TPA for 12 to 20 weeks. After exclusion of doublets, neutrophils were characterized based on CD19-CD3 ϵ -CD45⁺CD11b⁺Ly6C⁺Ly6G^{hi}. After exclusion of dead cells, IL17A-producing dermal $\gamma\delta$ T cells were selected by GR1-CD19-CD45⁺CD3 ϵ ⁺ $\gamma\delta$ TCR⁺IL17A⁺ staining.

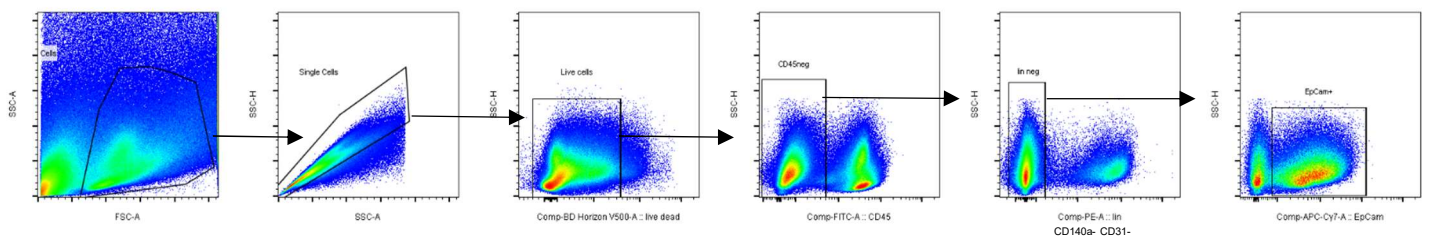


Figure S3 (related to Figure 4): Gating strategy to isolate EpCAM-positive cells from skin and tumors after DMBA/TPA treatment. After up to 40 weeks of carcinogenic treatment on *Zfp36^{ΔEP}* and control mice, epithelial cells from papillomas (~3 mm size), adjacent non-tumoral skin and mock skin, were sorted by exclusion of dead cells and CD45-CD140a-CD31-EpCAM⁺ selection.

Supplementary table

Table S1: List of RT-PCR primers

Target	Sequence FW 5' to 3'	Sequence RV 5' to 3'	Taqman probe 5' to 3'
<i>Actb</i>	TCC-TGA-GCG-CAA-GTA-CTC-TGT	CTG-ATC-CAC-ATC-TGC-TGG-AAG	ATC-GGT-GGC-TCC-ATC-CTG-GC
<i>Areg</i>	GGA-CAA-TGC-AGG-GTA-AAA-GTT-	TGA-AAG-AAG-GAC-CAA-TGT-CAT-	/
<i>Cxcl1</i>	CCG-AAG-TCA-TAG-CCA-CAC-TC	TTT-CTG-AAC-CAA-GGG-AGC-TT	AAG-GCA-AGC-CTC-GCG-ACC-AT
<i>Cxcl2</i>	ACA-TCC-AGA-GCT-TGA-GTG-TGA	GCC-CTT-GAG-AGT-GGC-TAT-G	CCC-ACT-GCG-CCC-AGA-CAG-AA
<i>Il17f</i>	ATT-CCA-GAA-CCG-CTC-CAG-TT	CTG-GGC-CTC-AGC-GAT-CTC-T	TGG-GAT-TAC-AAC-ATC-ACT-CGA-GAC-CC
<i>Inhba</i>	GGA-GAA-CGG-GTA-TGT-GGA-GA	TGG-TCC-TGG-TTC-TGT-TAG-CC	/
<i>Lcn2</i>	GAC-TTC-CGG-AGC-GAT-CAG-T	ACA-TCG-TAA-AGC-TGC-CTT-CTG	TGG-ACC-GCA-TTG-CCT-GCCAGG-CCC-A
<i>Lif</i>	AGA-ATC-AAC-TGG-CAC-AGC	TGG-AAA-GAT-GGG-AAG-TCT-G	/
<i>Pgf</i>	CCG-GCC-CTG-GCT-GCA-TTG-AA	CAG-GCA-AAG-CCC-ACA-GGC-GA	/
<i>S100a8</i>	CCT-TTG-TCA-GCT-CCG-TCT-TC	CAA-GGC-CTT-CTC-CAG-TTC-AG	AAG-GAA-ATC-TTT-CGT-GACAAT-GCC-G
<i>S100a9</i>	AGC-CTT-GAG-CAA-GAA-GAT-GG	TTG-ATG-GAA-GGT-GTC-GAT-GA	TGG-AGC-GCA-GCA-TAA-CCA-CCA
<i>Sema4d</i>	CCG-ACC-TGG-AGC-GTG-TAT-C	AGA-CAA-ACT-TCC-TCC-CCT-TC	/
<i>Sema6d</i>	CAG-AAG-CAT-GGG-AGA-TGG-AT	GCC-ACC-CAT-GTC-GTT-TTT-AC	/
<i>Tnf</i>	CAG-ACC-CTC-ACA-CTC-AGA-TCA	CAC-TTG-GTG-GTT-TGC-TAC-GA	TCG-AGT-GAC-AAG-CCT-GTA-GCC-CA
<i>Zfp36</i>	GAC-CAC-CGG-ACA-CTG-AAC-T	TCC-TGG-AAT-CTT-AAG-TGCTGT-GA	/