Supporting Information

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SI Materials and Methods

Mice and Reagents. All mice were maintained under specific pathogen-free conditions in the animal facility of the Institute of Cellular and Organismic Biology at Academia Sinica, and the experimental protocol was approved by Academia Sinica's Institutional Animal Care and Utilization Committee. Mice were genotyped with genomic PCR using primers described previously (1). A primer set specific for P-selectin (*Selp*; 5′-TTGTAAATCAGAAGGAAGTGG-3′ and 5′-CGAGTTACTCTTGATGTAGATCTCC-3′) was used for normalization. C57BL/6 mice were purchased from the Laboratory Animal Center in Taiwan and used at 6–10 wk of age. 293T and NIH 3T3 cells were cultured in DMEM (Invitrogen) supplemented with 10% (vol/vol) FBS and 100 U/mL penicillin/streptomycin.

Antibodies for Flow Cytometry Analysis. The following antibodies were used for flow cytometry analysis in this study: FITC- or allophycocyanin (APC)-conjugated anti-mouse Gr-1 (clone RB6-8C5; eBioscience), phycoerythrin (PE)-conjugated anti-mouse Mac-1 (clone M1/70; BD Pharmingen), FITC-conjugated anti-mouse CD4 (clone RM4-5; BD Pharmingen), APC-Cy7–conjugated anti-mouse CD8 (clone 53-6.7; BD Pharmingen), APC-conjugated anti-mouse CD44 (clone IM7; BD Pharmingen), PE-conjugated anti-mouse CD62L (clone MEL-14; BD Pharmingen), APC-conjugated anti-mouse CD138 (clone 281-2; BD Pharmingen), PE-conjugated anti-mouse B220 (clone RA3-6B2; BD Pharmingen), and APC-conjugated CD3 (clone 145-2C11; BD Pharmingen).

Isolation of Immune Cells. Spleen and bone marrow cells were isolated from control (Ctrl) and conditional knockout (CKO) mice. Cells were stained with antibodies specific for CD3, B220, Gr-1, and Mac-1 as described above and separated using a FACSAria II cell sorter (BD Biosciences).

Isolation and Culture of Primary Keratinocytes. For culture of mouse primary keratinocytes, mice were killed, and tail skin was peeled off of tail cartilage, followed by overnight incubation in 2.5 mg/mL dispase II (Roche) at 4 °C. The epidermis was then separated from the dermis using forceps and further digested into single suspended keratinocytes with TrypLE (Invitrogen). The keratinocytes were washed with PBS and passed through a 70-μm mesh to remove debris before use. Freshly isolated keratinocytes were cultured in Progenitor Cell Targeted Medium (CELLnTEC) in collagen-coated 24-well plates at a density of 0.4×10^6 cells in 0.4 mL/well. In some cases, cultured keratinocytes were treated with LPS (5 μg/mL; Sigma-Aldrich) or TNF-α (50 ng/mL; PeproTech) for 24 h. In some cases, PR domain containing 1, with ZNF domain (*Prdm1*) was deleted in vitro by adding 50 nM 4-hydroxytamoxifen (4OHT) or the solvent control, ethanol (EtOH). Human keratinocytes were cultured as described previously (2). For stimulation of human keratinocytes, cells were cultured without feeder cells in collagen-coated plates in Progenitor Cell Targeted Medium.

Plasmids. The Blimp-1 expression vector has been described previously (3). HA-tagged FBJ osteosarcoma oncogene (c-Fos) and fos-related antigen 1 (Fra-1) cDNA was cloned into the pcDNA expression vector. For construction of the luciferase reporter, the colony stimulating factor 3 (*Csf3*) promoter region from −3,886 to +20 relative to the transcriptional start site was amplified from tail genomic DNA of C57BL/6 mice by PCR and then cloned into the pGL3B vector (Promega). The activator protein 1 (AP-1)–binding site located in the −652 to −646 region of the *Csf3* promoter was mutated by site-directed mutagenesis. Details of cloning procedures are available on request. The *Renilla* luciferase reporter driven by the *thymidine kinase* (*tk*) promoter (pRL-TK) has been described previously (3).

RNA Isolation and Quantitative RT-PCR. Total RNAs were isolated on an RNeasy spin column (Qiagen), and 250 ng of total RNAs were used for cDNA synthesis with a High-Capacity cDNA Reverse-Transcriptase Kit (Applied Biosystems) according to the manufacturer's instructions. Quantitative RT-PCR (qRT-PCR) analysis of the cDNA was carried out using the TaqMan primer sets or primers for the SYBR Green method on an ABI Prism 7300 sequence detection system (Applied Biosystems) as reported previously (3). The deletion efficiency of *Prdm1* transcripts was determined using the Taqman primer set (ID_MA00197285_A1; Applied Biosystems) and the internal control Mrp32 primer set (ID_MA00777741_SH; Applied Biosystems). Mouse *Prdm1* transcript levels were measured using the Taqman primer set as described previously (4). The following primer sets were used in the SYBR Green method: *Prdm1*, 5′-CGAAATGCCCTTCTACCCTG-3′ and 5′-GCGTTCAAGTAAGCGTAGGAGT-3′; thymic stromal lymphopoietin (*Tslp*), 5′-GACTGTGAGAGCAAGCCAGCT-3′ and 5′-CTCCGGGCAAATGTTTTGTC-3′; S100 calcium binding protein A8 (*S100a8*), 5′-GAAATCACCATGCCCTCTACAAG-3′ and 5′-TTTATCACCATCGCAAGGAACTC-3′; S100 calcium binding protein A9 (*S100a9*), 5′-GGAGCGCAGCATAACCACCATC-3′ and 5′-GCCATCAGCATCATACACTCCTCA-3′; *Csf3*, 5′-GAGCAGTTGTGTGCCACCTACA-3′ and 5′-CCAGAGAGTGGCCCAGCA-3′; C-C chemokine ligand 2 (*Ccl2*), 5′-GGCTCAGCCAGATGCAGTTAAC-3′ and 5′-CCTACTCATTGGGATCATCTTGCT-3′; C-X-C chemokine ligand 1 (*Cxcl1*), 5′-GCAGACCATGGCTGGGATT-3′ and 5′-TGTCAGAAGCCAGCGTTCAC-3′; vascular endothelial growth factor a (*Vegfa*), 5′-GAGCAGAAGTCCCATGAAGTGAT-3′ and 5′-CTGCTGTGCTGTAGGAAGCTCAT-3′; *Il1a*, 5′-TGGCCAAAGTTCCTGACTTGT-3′ and 5′-ATGAAGTGAGCCATAGCTTGCA-3′; *Il1b*, 5′-AGTTGACGGACCCCAAAAGAT-3′ and 5′-GTGCAGTTGTCTAATGGGAACGT-3′; *Tnf*, 5′-GACCCTCACACTCAGATCATCTTCT-3′ and 5′-CCTCCACTTGGTGGTTTGCT-3′ *Cxcl2*, 5′-ACTGCGCCCAGACAGAAGTC-3′ and 5′-CAGTTAGCCTTGCCTTTGTTCAG-3′; *Cxcl5*, 5′-ATCTAGCTGAAGCTGCCCCTTC-3′ and 5′-GGGATCACCTCCAAATTAGCG-3′; *Il24*, 5′-CAGCCCAGTAAGGACAATTCCA-3′ and 5′-GCGACTTCTGTATCCAACTGTTTG-3′; FBJ osteosarcoma oncogene (*Fos*), 5′-ATCGGCAGAAGGGGCAAAGTAG-3′ and 5′-GCAACGCAGACTTCTCATCTTCAAG-3′; FBJ osteosarcoma oncogene b (*Fosb*), 5′-CTCCTCCTGGACGAATTGA-3′ and 5′-CAGTCAGATAGGGGTTCACATT-3′; *Jun*, 5′-CAGAGAGGAAGCGCATGAGG-3′ and 5′-TTCCTTTTCCGGCACTTGG-3′; *Junb*, 5′-TCACCGAGGAGCAGGAGG-3′ and 5′-GGTCGTCCAGGGCTTTGA-3′; fos-like antigen 1 (*Fosl1*), 5′-GATGGTGCAGCCTCATTTCC-3′ and 5′-CCCGATTTCTCATCCTCCAAT-3′; *Fosl2*, 5′-CCCACAATCAACGCCATCA-3′ and 5′-CCTCAGGAGACAGCTGCTCAT-3′; activating transcription factor 2 (*Atf2*), 5′-CTGGCAGGACCATGAATTAGTG-3′ and 5′-CCTCTGTTTCATGGCAAAGACA-3′; *Jund*, 5′-CACACATCACGCCACAGAAGT-3′ and 5′-TGTCCCTACCCTGCTGTTTCTT-3′, and actin, beta (*Actb*), 5′-GCTGTATTCCCCTCCATCGTG-3′ and 5′-CACGGTTGGCCTTAGGGTTCAG -3′.

Immunoblot Analysis. Keratinocytes from Ctrl and CKO mice were isolated and cultured in CnT-07 Medium (CELLnTEC). Cells were washed before lysing with lysis buffer [0.5% (wt/vol) Triton X-100, 20 mM Hepes (pH 7.9), 300 mM NaCl, 1 mM EDTA, 1 mM EGTA, and protease inhibitor mixture (Roche)]. Immunoblotting was performed essentially as described previously (4), using anti–Blimp-1 (1:250) (5), anti–c-Fos (1:200; Abcam), and anti–Fra-1 (1:400; Santa Cruz Biotechnology). Antibodies specific to β-actin (Sigma-Aldrich) or tubulin (Thermo Scientific) were used as loading controls.

Histology, Immunofluorescence, and Immunocytochemical and Immunohistochemistry Staining. To prepare frozen sections, freshly isolated skin samples were embedded in optimal cutting temperature compound and immediately immersed in liquid nitrogen. Cryosections (5 μm) were cut and fixed with acetone for further immunofluorescence staining. Adjacent sections were stained with primary antibodies, including anti–Blimp-1 (5), anti–Gr-1 (clone RB6-8C5; eBioscience), anti-K1 (clone PRB149P; Covance), anti-K5 (clone PRB-160P; Abcam), anti-loricrin (rabbit polyclone; Abcam) and anti-Ki67 (clone TEC-3; Dako) for immunohistochemistry (IHC) staining and Alexa Fluor 488-conjugated anti-mouse CD4 (clone RM4-5; Biolegend), anti-mouse F4/80 (clone BM8; eBioscience), and PE-conjugated anti-rat IgG (Invitrogen) for immunofluorescence staining. IHC staining was done using secondary antibodies conjugated to HRP or alkaline phosphatase, and the color was developed using tetramethylbenzidine or nitro blue tetrazolium/5-bromo-4-chloro-3-indolyl phosphate substrates. Some sections were further counterstained with hematoxylin. For immunofluorescence staining, images were acquired under a fluorescence microscope (Leica DM6000B).

2,4-Dinitrofluorobenzen–Induced Contact Dermatitis (Contact Hypersensitivity) and Tape-Stripping Test. Mice injected with 4OHT for 2 mo were used for induction of contact dermatitis. In brief, abdominal skin of Ctrl and CKO mice was shaved and sensitized with 20 μL 0.5% (wt/vol) 2,4-dinitrofluorobenzene (DNFB) in acetone/olive oil (4:1). After 5 d, each side of the ear was challenged with either 0.2% DNFB or solvent for 24 h. Ear inflammation was determined by increases in ear thickness after challenge and H&E staining. Ear thickness was measured with a micrometer (Mitutoyo) before and after challenge. The ears were then removed for pathological examination with H&E staining. For tape-stripping test, skin of Ctrl and CKO mice in dorsal area was shaved and stripped for 10 strokes using transparent tape as described previously (6). Two days later, mice were killed, and skin was removed for pathological examination with H&E staining.

Blood Leukocyte Count, Multiplex Array, and ELISA. Peripheral blood samples were collected from Ctrl and CKO mice using EDTA as an anticoagulant. Blood samples were diluted in cell pack buffer, and the numbers of leukocytes were determined with an automated hematology analyzer (XT-1800i; Sysmex). Luminex assays were performed following the manufacturer's procedures using the Mouse Ig Isotyping Kit and MILLIPLEX MAP Mouse Cytokine/Chemokine panel (Millipore). Luminex samples were analyzed with a Luminex 200 (Millipore). Serum levels of granulocyte colony-stimulating factor (G-CSF) were determined with an ELISA system (R&D Systems).

Lentiviral Vector Preparation and Generation of Lentiviral Vector-Producing shRNA for *Fos* or *Fosl1* Lentiviral vectors were prepared as described previously (7). Viral supernatants were concentrated, and the viral titers were determined as described previously (7). Keratinocytes were transduced by lentiviral vectors at a multiplicity of infection of 5. For overexpression of Blimp-1–GFP, GFP cDNA was fused in C termini of Blimp-1 cDNA and expressed by a lentiviral vector as described previously (8). For knockdown of c-Fos and Fra-1, shRNAs specific for *Fos* and *Fosl1* were cloned into a lentiviral vector essentially following a cloning strategy described in a previous report (9), with the following sequences: shFos#1, 5′-GGGAGCTGACAGATACACTCC-3′; shFos#2, 5′-GGGCTGCACTACTTACACGTC-3′; shFosl1#1, 5′-GGGATCCCCAAAGCTGCTCAC-3′; shFosl1#2, 5′- GGGTCCTTTGACACCCTTACC-3′.

Microarrays. Fully confluent primary keratinocytes in 24-well plates were harvested for microarray study according to the protocol provided by the manufacturer (3′ IVT Express Kit; Affymetrix). The biotinylated cRNAs were hybridized to the GeneChip Mouse Expression Array 430A (Affymetrix). After staining, the array was scanned, and images were acquired with a GeneChIP Scanner 3000 (Affymetrix). Data were analyzed with GeneSpring GX11 software (Agilent). The 12 most up-regulated and most down-regulated genes in CKO + EtOH mice were illustrated on a heat map using MultiExperiment Viewer, version 4.6.1, of the TM4 Microarray Software Suite. The microarray data from this study have been submitted to the National Center for Biotechnology Information's Gene Expression Omnibus database (identifier GSE34586).

ChIP. ChIP was performed essentially as described previously (3, 10). In brief, 1×10^7 primary keratinocytes lentivirally transduced with Blimp-1–GFP or GFP-encoding vector were used per reaction, and 5 μg of anti-Blimp-1 (3, 11) was used to precipitate the Blimp-1–chromatin complex. Immunoprecipitated chromatin samples were quantified by qRT-PCR using primers that specifically amplified fragments encompassing the Blimp-1–binding site or within 100 bp of the Blimp-1–binding site at each individual gene locus using the SYBR Green method on an ABI Prism 7300 system (Applied Biosystems). Data were normalized to GFP vector-transduced cells. The following primers were used in this study: *Csf3* site 1, 5′-ATGCACACAGACACACCTCACC-3′ and 5′-AACAAAGGCCAAAACCTGTGG-3′; *Csf3* site 2, 5′-GAGCTTTGCTTCCCTCTGTCTC-3′ and 5′-AGAACCTTGAGCTGGAGGCTG-3′; *Csf3* site 3, 5′-CCAGGGCTATCGAGACAGAGAA-3′ and 5′-GAGGACAGGCCAAGGGATTC-3′; *Csf3* sites 4, 5, and 6, 5′-AGGACCTGAGCTGGACAAAGAG-3′ and 5′-AGAGAGTGGCCCAGCAACAC-3′; *Csf3* sites 7 and 8, 5′-TTCTCTCCACTTCCGAGTTTTGT-3′ and 5′-CAGGGCTCACTGATTTTTTGG-3′; *Cxcl1* site 1, 5′-CTCTTATCGGGCTGCAAACCT-3′ and 5′-ACAGCCATCCATTGGATGAAG-3′; *Cxcl1* site 2, 5′-TCTGGAGCACACAGCTCTTTCA-3′ and 5′-CGGATACAGGGAGGTGAGCTTA-3′; *Cxcl1* sites 3 and 4, 5′-TTGCAGGGAAACACCCTGTAC-3′ and 5′-CTGAGTCCTGTTGTGGAGCTCTAG-3′; *Cxcl1* site 5, 5′-CTCAAAACCCCTATGCAAATGG-3′ and 5′-AGGGAAATCTCACTGGCAAAAG-3′; *Cxcl5* site 1, 5′-CGGCTGACCTGTCCTTTTCTC-3′ and 5′-ACTGGTCTTTGAGGATCTGATGTG-3′; *Cxcl5* site 2, 5′-AGGCAGTGGTTAGCCAGACTATG-3′and 5′-AGCGGCTTAGCCAATCTGTTAC-3′; *Cxcl5* site 3, 5′-CAATGCAAGGCCAGCTTAGG-3′ and 5′-CGGCTGGCACTCTGGTAATAG-3′; *Cxcl5* site 4, 5′-TGGATCCAGAAGCTCCTGTGA-3′ and 5′-GCCCGCTGCAGAGACTAAGATA-3′; *Cxcl5* site 5, 5′-GAAACCATTGTCCCTGAAGCTT-3′ and 5′-TCACAATTTCCGATAGTGTGACAG-3′; *Il24* sites 1, 2, and 3, 5′-CCCTGGGTATCCCACACCTATT-3′ and 5′-CTCCCAAATGCTGGGATTAAAG-3′; *Il24* site 4, 5′-CAGCAGTACCCACCAGCTAGGT-3′ and 5′-GCCGAGTCAAGGGAAAAAGAA-3′; *Il24* sites 5 and 6, 5′-ACCAGCCCTGTGTGTCAAGTC-3′ and 5′-CCATAGCAGGAAGCTAAGAAGCA-3′; *Il24* sites 7 and 8, 5′-TGAAGTATTTCTCCAGGGAAGCA-3′ and 5′-TAAGCAAGGCTGGGAGAACTGT-3′; *Il24* sites 9 and 10, 5′-CAGGTGTGAGGAGATTGTGTGAA-3′ and 5′-ACCTCGCTTTGTGCTTTTTGTC-3′; *Il24* sites 11 and 12, 5′-ATCACAAGCATCCGGCTGTT-3′ and 5′-GAAGGGAACGACTCCCATCAG-3′; *Il24* site 13, 5′-GCT-

TCAGTGGTCCGCTTCTTC-3′ and 5′-TTGGCCAGAGTGGAGAATGAC-3′; *Il24* site 14, 5′-AGTTTTACAGGTCCCCGATGTC-3′ and 5′-CATAAACGAGGCCACTTGAACAC-3′; *Il1b* site 1, 5′-AAGGCTCCCTTCTCTCTGATGA-3′ and 5′-ACGACTTAGCACAGGCCTTAGG-3′; *Il1b* site 2, 5′-GGCAAAAGAAGGCTGCCTAGTA-3′ and 5′-TCTGCAGCACTGAAGGGAAATA-3′; *Il1b* sites 3 and 4, 5′-CCACCCTTCAGTTTTGTTGTGA-3′ and 5′-GATGAGCCTATTAGGCCTCGAA-3′; *Il1b* site 5, 5′-CCGTTCCTTCATTCCTCAGAGA-3′ and 5′-GTTGCCATAGCTGCTTCAGACA-3′; *Il1b* sites 6 and 7, 5′-ACTCAGGAGGCAGCTGTTTCTC-3′ and 5′-CGTCACCCAAGGCTGAACTAAC-3′; *Il1b* sites 8, 9, and 10, 5′-AGCCTACTGGGTCCCTGTGTT-3′ and 5′-GGCAGTTATTGCATGTCCATCA-3′; *Il1b* site 11, 5′-ATCACCCCCAACAGAAGTCGTA-3′ and 5′-ACCCTTTCACTGCATTCTCCAA-3′, *Fosl1* site 1, 5′-CCTGGAGGCTCCTGAGAAAGAT-3′ and 5′-GTGCCTGGATTGGTGTTGCT-3′; *Fosl1* site 2, 5′-CTTCGGTCCTCCTGCCTTTATT-3′ and 5′-ACCTCCTGTTTCCGCATTTGT-3′; *Fosl1* site 3, 5′-ACCCTGGTTTGTCAATAACCTTTC-3′ and 5′-GTTGGACTGTGCCACCATCAC-3′; *Fosl1* site 4, 5′-TCTGCAAGAAAAGGAAGGAAAGG-3′ and 5′-CCTGAATGCTGGGATTAAATGC-3′; *Fosl1* site 5, 5′-GGGAGGTAGAGGCAGAGGAATC-3′ and 5′-CCTCACAGATGGGCATACTTTCA-3′; *Fosl1* sites 6 and 7, 5′-TGCGTATGGAAGCTGAAAAGAA-3′ and 5′-GTCACCCCGAGGTACAAAGC-3′.

Transfection and Luciferase Reporter Assay. Keratinocytes isolated from skin of neonatal mice were used in luciferase reporter assay, and transfection was performed using the nucleofection technique (Lonza AG) according to the manufacturer's suggestions. A total of 3×10^6 primary keratinocytes were used in each transfection, and cultured in each well of 12-well plates. *Renilla* luciferase reporter driven by the *tk* promoter (pRL-TK) was used as the internal control in all reporter assays for normalization. After 24 or 48 h, cells were washed and subjected to measurement of luciferase activity with the Promega Dual-Luciferase Reporter Assay System. Fold repression was calculated as described previously (3).

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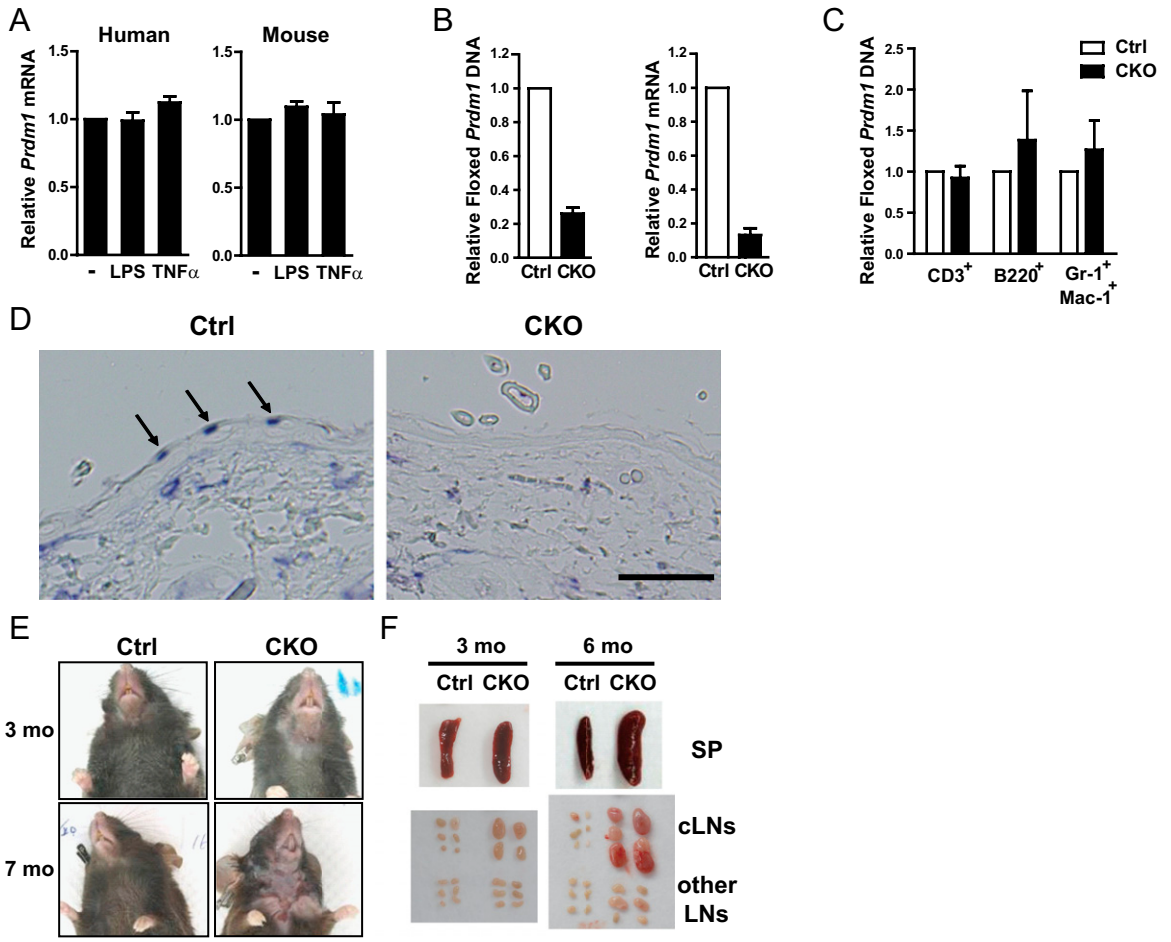

Fig. S1. Blimp-1 mRNA in human and mouse keratinocytes and deletion of epidermal *Prdm1* in adult mice leads to spontaneous development of skin ulcers and enlarged lymphoid organs. (*A*) qRT-PCR showing the level of Blimp-1 mRNA in human (*Left*) and mouse (*Right*) primary keratinocytes treated with LPS (5 μg/mL) or TNF-α (50 ng/mL) or untreated (–) for 24 h. Data are mean ± SEM ($n = 3$). (*B*) Deletion of *Prdm1* alleles at 2 mo after i.p. injection of 4OHT, followed by isolation of genomic DNA or RNA samples from Ctrl and CKO tail skin keratinocytes for qPCR (*Left*) or qRT-PCR (*Right*) analysis, respectively. Primers were designed to amplify the floxed *Prdm1* alleles, and primers for *Selp* were used for normalization of genomic DNA. *Actb* was used for normalization of RNA. Data are mean ± SEM (*Left*, $n = 3$; *Right*, $n = 4$). (*C*) Deletion of *Prdm1* alleles at 1–2 mo after injection of 4OHT, followed by isolation of genomic DNA from indicated immune cells for qPCR with primers described in *B*. (*D*) IHC staining with anti–Blimp-1 of skin samples collected from Ctrl and CKO mice at 2 mo after injection with 4OHT. The Blimp-1 signals were present in the granular layer of Ctrl epidermis (*Left*, arrows) but were absent in CKO epidermis (*Right*). (Scale bar: 50 μm.) (*E* and *F*) At 3 and 7 mo after 4OHT injection, CKO mice, but not Ctrl mice, spontaneously developed alopecia and ulcers in the neck skin (*E*), along spleen (SP) and cervical lymph node (cLN) enlargement (*F*).

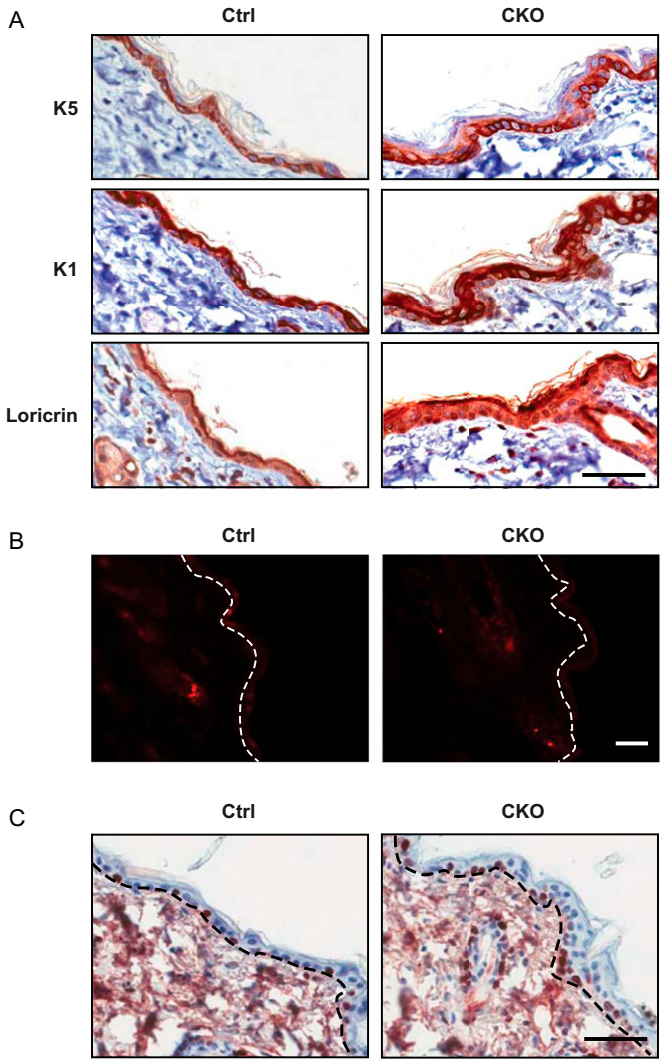

Fig. S2. Differentiation, apoptosis, and proliferation of epidermis in Ctrl and CKO mice. (*A*) IHC staining of skin samples collected from Ctrl and CKO mice with basal layer-specific anti-K5, suprabasal-specific anti-K1, or terminally differentiated keratinocyte-specific anti-loricrin at 2 mo after injection with 4OHT. (*B*) TUNEL staining of skin sections from Ctrl and CKO mice. (*C*) IHC staining for Ki67 in Ctrl and CKO mice. The dotted line in *B* and *C* denotes the epidermis–dermis boundary. (Scale bars: 100 μm in *A* and *C*; 50 μm in *B*.)

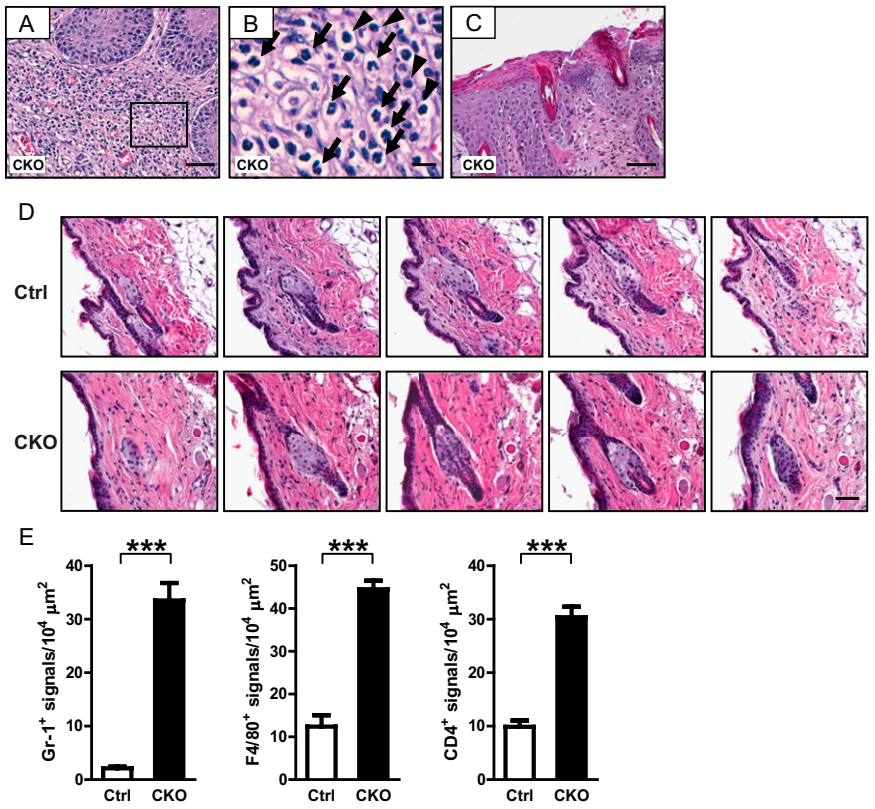

Fig. S3. Skin inflammation with parakeratosis and enlarged sebaceous glands in CKO mice. (*A* and *B*) H&E staining of neck skin sections from CKO mice at 6 mo after 4OHT injection, showing inflamed skin and enlarged vessels (*A*) and infiltration of granulocytes (arrows) and lymphocytes (arrowheads) into the dermis (*B*). (*C*) H&E staining showing parakeratosis in CKO mice at 6 mo after injection with 4OHT. (*D*) H&E staining of serial sections of Ctrl (*Upper*) and CKO skin (*Lower*) showing enlarged sebaceous glands in CKO mice. Both Ctrl and CKO mice were injected with 4OHT for 2 mo. (*E*) Numbers of granulocytes (*Left*), macrophages (*Center*), and $CD4^+$ T cells (*Right*) calculated from the stained skin sections of Ctrl and CKO mice. ***$P < 0.005$. (Scale bars: 50 μm in *A*, *C*, and *D*; 10 μm in *B*.)

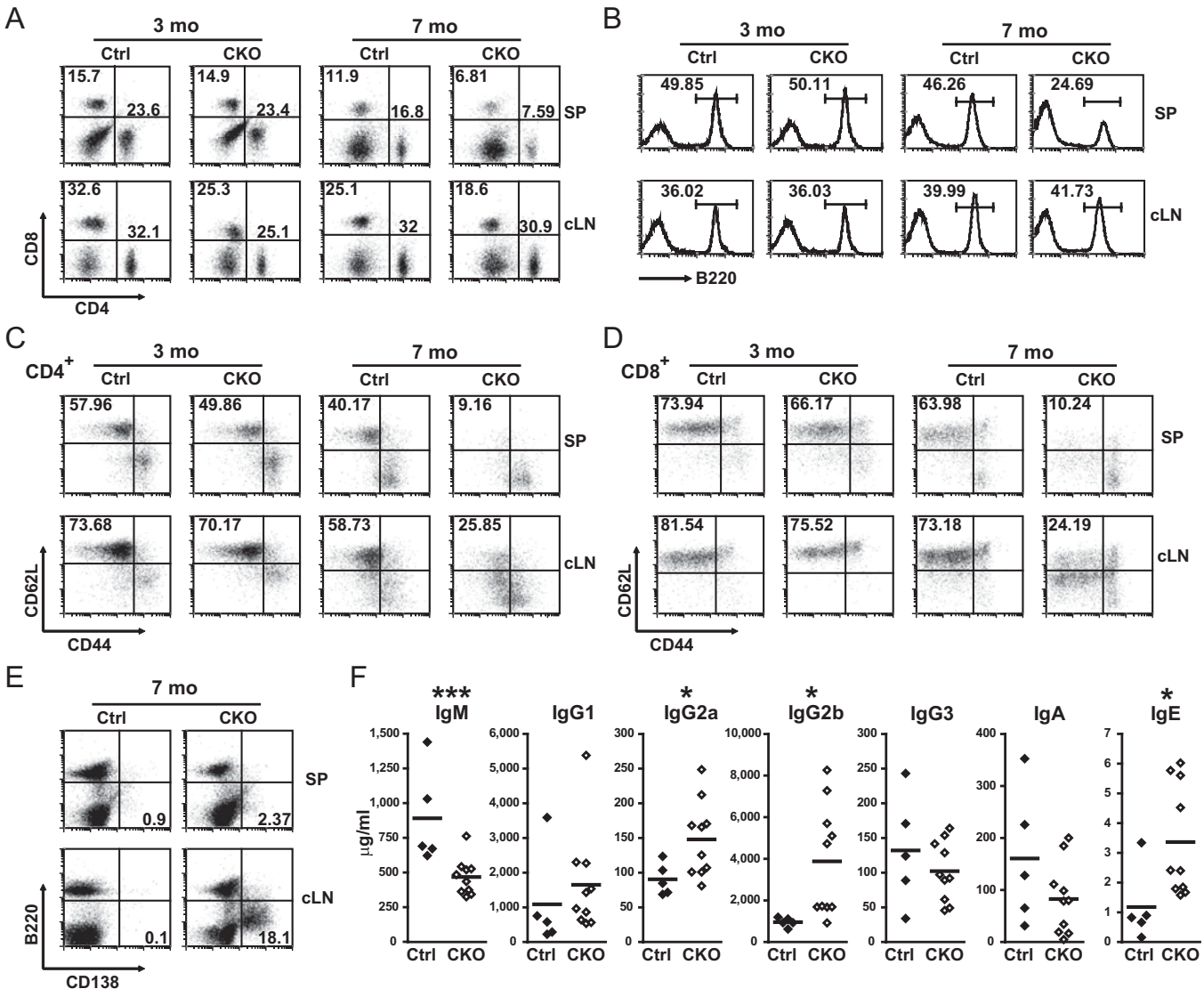

Fig. S4. CKO mice exhibit aberrant adaptive immune responses. (*A–D*) At 3 and 7 mo after 4OHT injection, splenocytes (SP) and cells of cervical lymph nodes (cLN) from Ctrl and CKO mice were isolated for flow cytometry analysis of the frequencies of the indicated immune cell markers. The percentages of $CD4^+$ and $CD8^+$ T cells (*A*) and $B220^+$ B cells (*B*) are shown in the quadrants. Dot plots show the frequencies of naïve and effector/memory $CD4^+$ (*C*) and $CD8^+$ (*D*) T cells. The percentage of naïve $CD4^+$ or $CD8^+$ T cells is indicated in each upper-left quadrant. (*E*) Dot plots showing the frequency of $B220^{low/-}CD138^+$ plasma cells from SPs and cLNs from Ctrl and CKO mice. Data are representative of at least three independent experiments from paired mice. (*F*) Levels of Ig subtypes in sera of Ctrl or CKO mice at 6 mo after 4OHT injection as analyzed with the Luminex assay. The mean in each group is designated by a horizontal line (Ctrl, $n = 5$; CKO, $n = 6$). $^*P < 0.05$; $^{***}P < 0.005$, comparing Ctrl and CKO groups.

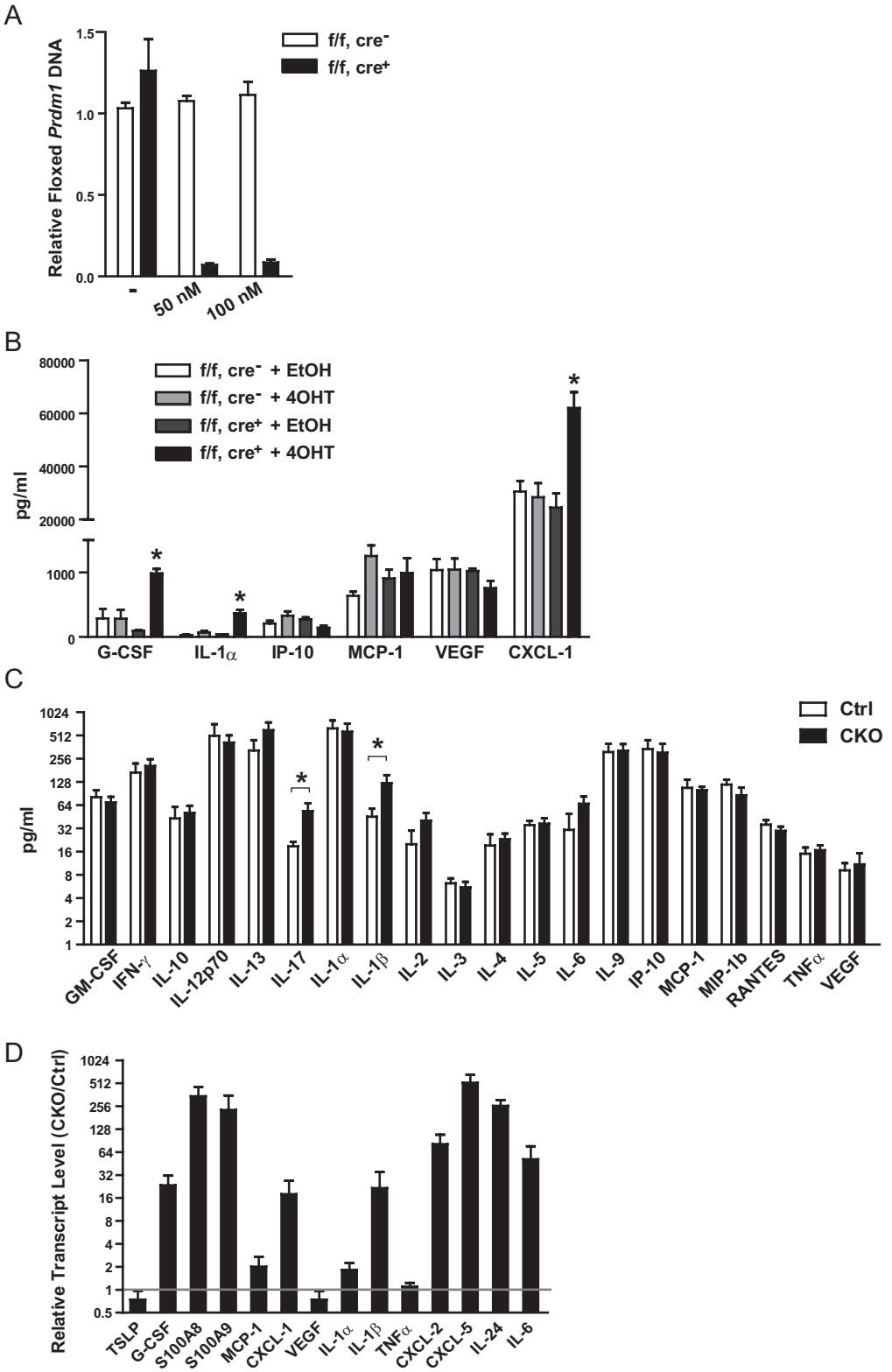

Fig. S5. Elevated production of G-CSF and IL-1α after deletion of *Prdm1* in keratinocytes. (*A*) Keratinocytes isolated from *Prdm1*$^{f/f}$, K5-CreER^{+} (f/f, Cre^{+}) and *Prdm1*$^{f/f}$, K5-CreER^{-} (f/f, Cre^{-}) mice were treated with the indicated dosages of 4OHT or EtOH for 3 d, followed by qPCR of genomic DNA to determine the in vitro deletion efficiency of *Prdm1* alleles. Here, "−" indicates untreated cells. (*B*) Culture medium from *A* was refreshed, and the supernatants in the culture were collected 2 d later for the Luminex assay. Levels of certain cytokines/chemokines, including GM-CSF, IFN-γ, IL-10, IL-12p70, IL-13, IL-17, IL-1β, IL-2, IL-3, IL-4, IL-5, IL-6, IL-9, macrophage inflammatory protein 1 beta (MIP-1β), regulated upon activation, normal T-cell expressed and secreted (RANTES), and TNF-α, were below the level of detection (<20 pg/mL) in this assay. Data are mean ± SEM ($n = 4$). *$P < 0.05$ compared with the other three control groups. (*C*) Luminex assay showing levels of the indicated cytokines/chemokines in serum of CKO mice at 6 mo after 4OHT injection. Data are mean ± SEM ($n = 9$). *$P < 0.05$. (*D*) qRT-PCR analysis of expression of the indicated cytokines/chemokines in neck skin tissue from Ctrl and CKO mice at 6 mo after 4OHT injection. Data are mean ± SEM ($n = 4$).

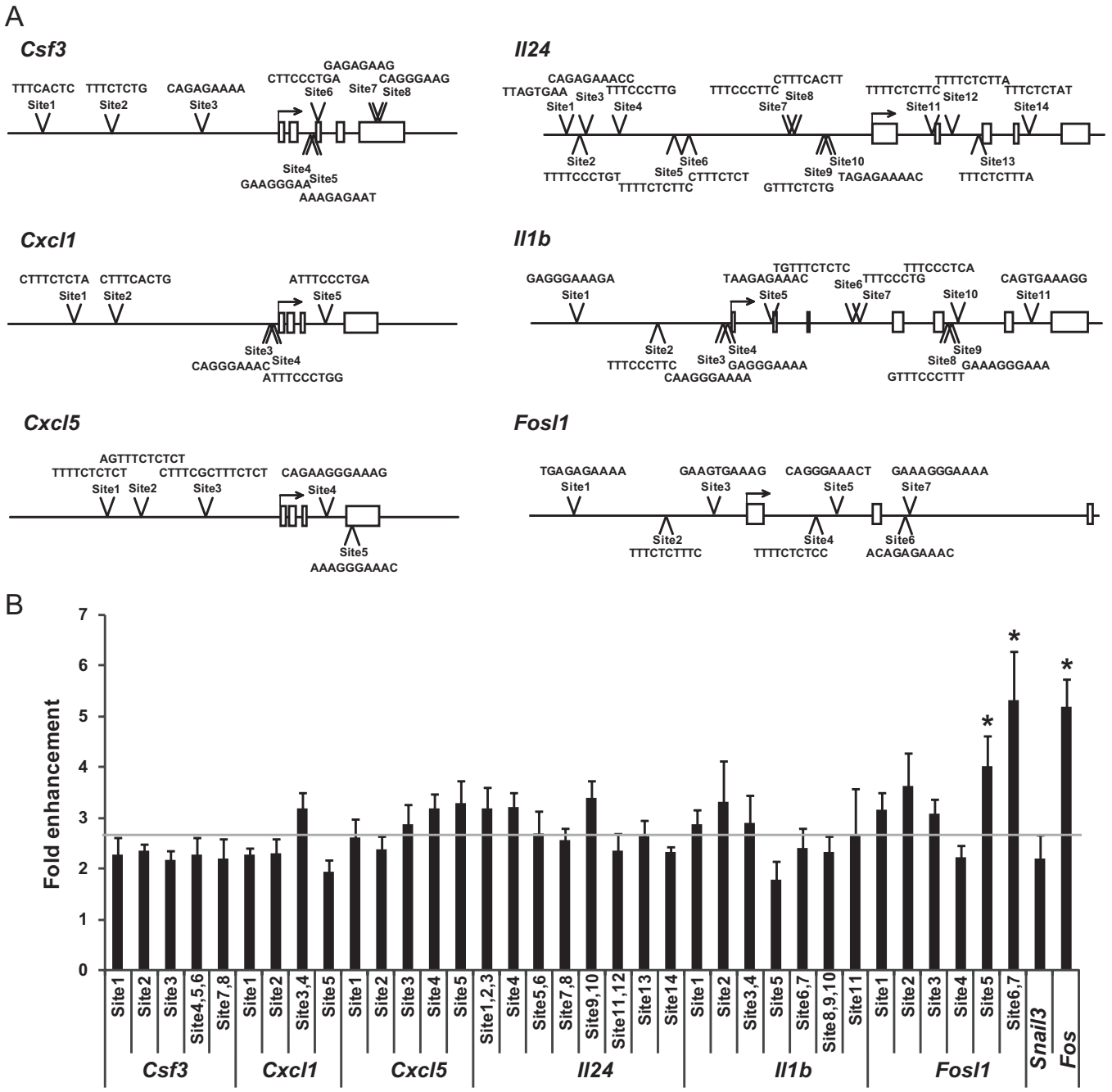

Fig. S6. ChIP of Blimp-1 binding at putative sites. *(A)* Putative Blimp-1–binding sites and sequences located between −5,000 bp and +5,000 bp of the transcriptional start site of the indicated genes. The arrow indicates the transcriptional start site, and open rectangles indicate exons. (*B*) Keratinocytes from neonatal mice were transduced with lentiviral vectors producing Blimp-1 fused with GFP (Blimp-1–GFP) or GFP alone. Binding of Blimp-1 to the putative Blimp-1–binding sites was analyzed with ChIP, followed by qPCR of immunoprecipitated DNA. Data represent the fold enhancement of gene loci in Blimp-1–GFP–expressing cells relative to that in GFP-expressing cells. *Snail3* served as the negative control locus. Data are mean ± SEM ($n = 5$). *$P < 0.05$.

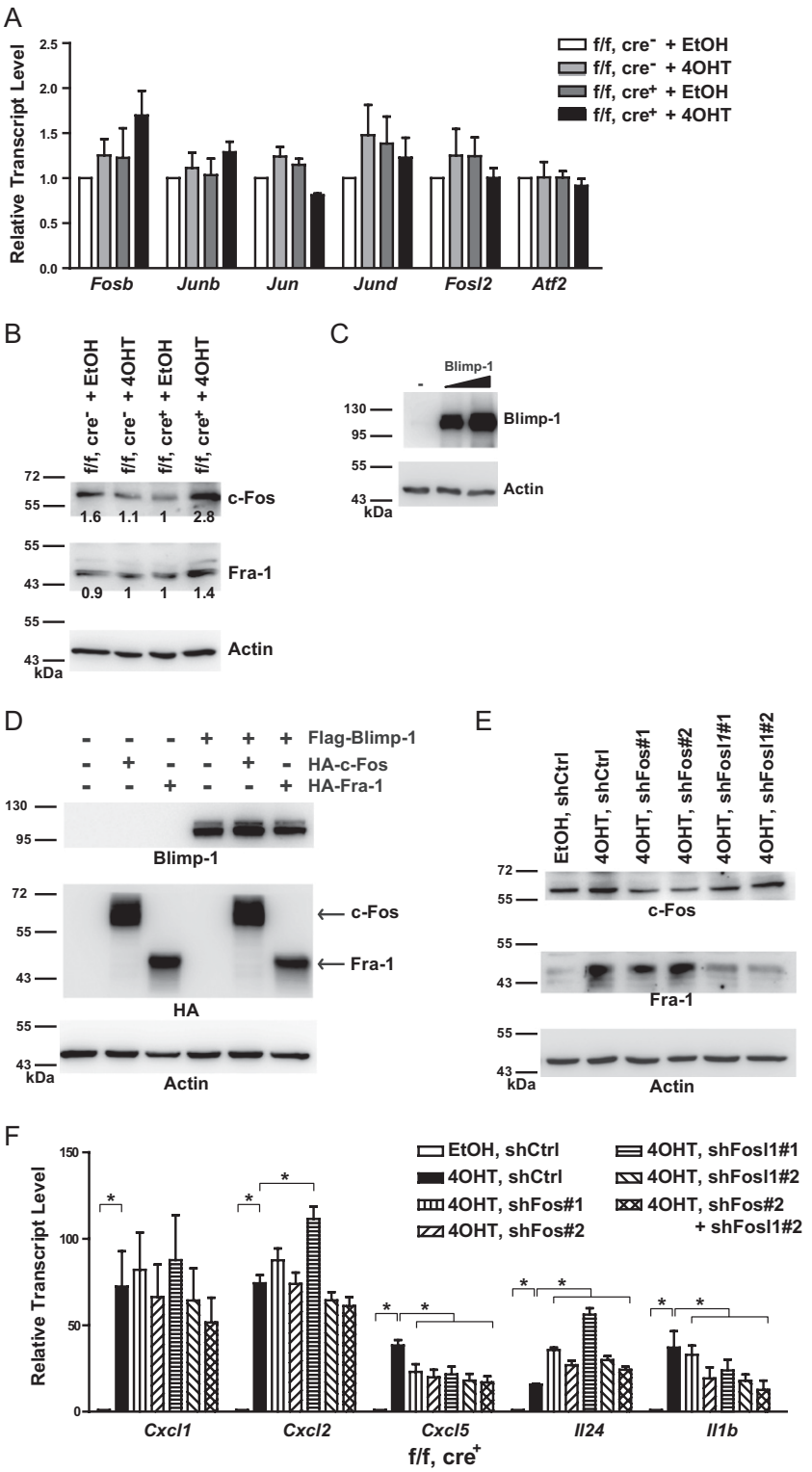

Fig. S7. Blimp-1 regulates *Fos* and *Fosl1*. *(A)* qRT-PCR analysis of the indicated AP-1 family members using RNA prepared from the indicated keratinocytes treated with 50 nM 4OHT or EtOH. (*B*) Immunoblot analysis of c-Fos and Fra-1 using lysates prepared from the indicated keratinocytes treated with 4OHT or EtOH. Protein band intensity was quantified and normalized to the level of the internal control (actin) in each corresponding lane first and then further compared with the ratio obtained from the f/f, Cre^+ + 4OHT group. (*C* and *D*) Immunoblot analysis of the expression of Blimp-1 (*C* and *D*) and indicated HA-tagged AP-1 family proteins (*D*) in primary keratinocytes transfected with the indicated expression vectors for the luciferase reporter assay. Actin served as the loading control. (*E*) Immunoblot analysis of c-Fos and Fra-1 expression at 5 d after transduction of the indicated primary keratinocytes with lentiviral vectors producing shCtrl, two different shRNAs against Fos (shFos#1 and shFos#2), or two different shRNAs against Fosl1 (shFosl1#1 and shFosl#2). Actin served as the loading control. (*F*) qRT-PCR analysis of mRNA levels of the indicated cytokines/chemokines from *Prdm1*-intact or *Prdm1*-deleted keratinocytes transduced with lentiviral vectors producing shCtrl, shFos#1, shFos#2, shFosl1#1, shFosl#2, or both shFos#2 and shFosl1#2 (shF+F). Results are representative of two independent experiments performed in triplicate and represent mean ± SEM.

Table S1. Genes with more than twofold up-regulation in microarray analyses in comparing the *Prdm1*$^{f/f}$, *K5-CreER*$^{+}$ (f/f, Cre^{+}) + 4OHT with *Prdm1*$^{f/f}$, *K5-CreER*$^{-}$ (f/f, Cre^{-}) + EtOH groups

Up-regulated genes			Fold (compared with f/f, Cre^{-} + EtOH)			
Gene symbol	Gene name	Unigene (Avadis)	f/f, Cre^{-} + EtOH	f/f, Cre^{-} + 4OHT	f/f, Cre^{+} + EtOH	f/f, Cre^{+} + 4OHT
Cxcl2	chemokine (C-X-C motif) ligand 2	Mm.4979	1	1.097	1.420	24.313
Il24	interleukin 24	Mm.196691	1	0.998	0.785	17.079
Cxcl1	chemokine (C-X-C motif) ligand 1	Mm.21013	1	1.129	1.485	16.355
Hmga2	high mobility group AT-hook 2	Mm.157190	1	0.774	0.947	15.988
Cxcl5	chemokine (C-X-C motif) ligand 5	Mm.4660	1	1.633	1.270	12.242
Fst	follistatin	Mm.4913	1	0.581	0.548	11.168
Mal	myelin and lymphocyte protein, T-cell differentiation protein	Mm.39040	1	0.761	0.615	10.872
Dusp6	dual specificity phosphatase 6	Mm.1791	1	0.973	1.245	10.800
Areg	amphiregulin	Mm.8039	1	1.914	1.830	9.138
Phlda1	pleckstrin homology-like domain, family A, member 1	Mm.3117	1	1.080	1.014	9.086
Cst6	cystatin E/M	Mm.36816	1	2.583	1.099	8.669
Fos	FBJ osteosarcoma oncogene	Mm.246513	1	1.189	1.119	6.334
Klk6	kallikrein related-peptidase 6	Mm.3944	1	2.277	1.647	6.201
Ptges	prostaglandin E synthase	Mm.28768	1	1.273	1.054	5.080
Odc1	ornithine decarboxylase, structural 1	Mm.34102	1	0.792	0.807	4.309
Nppb	natriuretic peptide precursor type B	Mm.2740	1	1.111	1.302	4.044
Ctgf	connective tissue growth factor	Mm.393058	1	0.963	0.830	4.036
Gyk	glycerol kinase	Mm.246682	1	1.174	1.072	3.975
Akr1c18	aldo-keto reductase family 1, member C18	Mm.41337	1	1.544	1.257	3.962
Tnfaip2	tumor necrosis factor, alpha-induced protein 2	Mm.255332	1	1.343	0.772	3.795
Cryab	crystallin, alpha B	Mm.178	1	1.681	1.063	3.639
Hbegf	heparin-binding EGF-like growth factor	Mm.289681	1	1.086	0.901	3.600
Fosl1	fos-like antigen 1	Mm.6215	1	1.139	1.177	3.595
Trim2	tripartite motif-containing 2	Mm.44876	1	1.371	1.152	3.511
Tsc22d1	TSC22 domain family, member 1	Mm.153272	1	1.223	1.028	3.501
Prom1	prominin 1	Mm.6250	1	0.840	0.719	3.460
Cldn3	claudin 3	Mm.158662	1	1.208	1.139	3.337
Upp1	uridine phosphorylase 1	Mm.4610	1	1.251	1.087	3.214
Lif	leukemia inhibitory factor	Mm.4964	1	1.119	1.132	3.198
Itga3	integrin alpha 3	Mm.57035	1	1.193	1.209	3.133
Plod2	procollagen lysine, 2-oxoglutarate 5-dioxygenase 2	Mm.79983	1	0.604	0.892	3.102
Procr	protein C receptor, endothelial	Mm.3243	1	0.563	0.908	3.079
Cd44	CD44 antigen	Mm.423621	1	1.030	0.990	3.044
Depdc6	DEP domain containing 6	Mm.295397	1	0.871	0.984	3.012
Tyrp1	tyrosinase-related protein 1	Mm.30438	1	1.124	1.385	2.956
Akap12	A kinase (PRKA) anchor protein (gravin) 12	Mm.27481	1	0.959	0.992	2.947
Homer2	homer homolog 2 (Drosophila)	Mm.228	1	1.038	0.890	2.922
Il1a	interleukin 1 alpha	Mm.15534	1	1.145	1.043	2.906
Angptl2	angiopoietin-like 2	Mm.208919	1	1.243	1.311	2.899
Tubb2a	tubulin, beta 2A	Mm.469917	1	1.098	1.269	2.836
Zfand2a	zinc finger, AN1-type domain 2A	Mm.24521	1	1.320	1.189	2.819
Mid1ip1	Mid1 interacting protein 1 (gastrulation specific G12-like (zebrafish))	Mm.29429	1	1.319	1.043	2.767
Mdm2	transformed mouse 3T3 cell double minute 2	Mm.22670	1	1.027	0.933	2.719
Etv4	ets variant gene 4 (E1A enhancer binding protein, E1AF)	Mm.5025	1	1.138	1.083	2.701
Tm4sf1	transmembrane 4 superfamily member 1	Mm.856	1	0.794	0.763	2.668
Dusp7	dual specificity phosphatase 7	Mm.275584	1	1.152	1.047	2.624
Epha2	Eph receptor A2	Mm.2581	1	0.998	0.907	2.576
Igfbp7	insulin-like growth factor binding protein 7	Mm.233470	1	0.991	0.954	2.576
Bid	BH3 interacting domain death agonist	Mm.235081	1	1.175	1.039	2.537
Psca	prostate stem cell antigen	Mm.46395	1	0.780	0.783	2.524
Ikbke	inhibitor of kappaB kinase epsilon	Mm.386783	1	0.965	1.041	2.524
Gjb3	gap junction protein, beta 3	Mm.90003	1	0.917	0.962	2.488
Bcl2l15	BCLI2-like 15	Mm.297245	1	1.100	0.900	2.473
Lipg	lipase, endothelial	Mm.299647	1	1.196	0.888	2.402
Ddit4l	DNA-damage-inducible transcript 4-like	Mm.250841	1	0.821	0.955	2.399

Table S1. Cont.

Up-regulated genes			Fold (compared with f/f, Cre$^-$ + EtOH)			
Gene symbol	Gene name	Unigene (Avadis)	f/f, Cre$^-$ + EtOH	f/f, Cre$^-$ + 4OHT	f/f, Cre$^+$ + EtOH	f/f, Cre$^+$ + 4OHT
Prl8a9	prolactin family8, subfamily a, member 9	Mm.46091	1	0.786	1.146	2.359
Cd55	CD55 antigen	Mm.101591	1	0.814	0.837	2.347
Cgref1	cell growth regulator with EF hand domain 1	Mm.45127	1	0.675	0.708	2.320
Rin1	Ras and Rab interactor 1	Mm.271922	1	0.862	0.996	2.253
Krt18	keratin 18	Mm.22479	1	0.805	0.797	2.237
Gapdhs	glyceraldehyde-3-phosphate dehydrogenase, spermatogenic	Mm.436562	1	0.953	0.895	2.228
Pdpn	podoplanin	Mm.2976	1	0.630	0.809	2.202
Tfrc	transferrin receptor	Mm.28683	1	1.007	0.978	2.170
Arhgef3	Rho guanine nucleotide exchange factor (GEF) 3	Mm.248606	1	0.899	0.899	2.159
Acaa1b	acetyl-Coenzyme A acyltransferase 1B	Mm.379402	1	0.724	0.866	2.148
Has3	hyaluronan synthase 3	Mm.56986	1	0.925	0.782	2.137
Dhh	desert hedgehog	Mm.384073	1	0.573	0.697	2.127
Ccnd1	cyclin D1	Mm.273049	1	0.618	0.859	2.084
Rapgef3	Rap guanine nucleotide exchange factor (GEF) 3	Mm.24028	1	0.878	0.750	2.064
Pigf	phosphatidylinositol glycan anchor biosynthesis, class F	Mm.219685	1	0.671	0.983	2.031
Pdpn	podoplanin	Mm.2976	1	0.630	0.809	2.202
Tfrc	transferrin receptor	Mm.28683	1	1.007	0.978	2.170
Arhgef3	Rho guanine nucleotide exchange factor (GEF) 3	Mm.248606	1	0.899	0.899	2.159
Acaa1b	acetyl-Coenzyme A acyltransferase 1B	Mm.379402	1	0.724	0.866	2.148
Has3	hyaluronan synthase 3	Mm.56986	1	0.925	0.782	2.137
Dhh	desert hedgehog	Mm.384073	1	0.573	0.697	2.127
Ccnd1	cyclin D1	Mm.273049	1	0.618	0.859	2.084
Rapgef3	Rap guanine nucleotide exchange factor (GEF) 3	Mm.24028	1	0.878	0.750	2.064
Pigf	phosphatidylinositol glycan anchor biosynthesis, class F	Mm.219685	1	0.671	0.983	2.031

Table S2. Genes with more than twofold down-regulation in microarray analyses in comparing f/f, Cre$^+$ + 4OHT with f/f, Cre$^-$ + EtOH groups

Down-regulated genes			Fold (compared with f/f, Cre$^-$ + EtOH)			
Gene symbol	Gene name	Unigene (Avadis)	f/f, Cre$^-$ + EtOH	f/f, Cre$^-$ + 4OHT	f/f, Cre$^+$ + EtOH	f/f, Cre$^+$ + 4OHT
Rptn	repetin	Mm.1417	1	0.300	0.980	13.976
Chdh	choline dehydrogenase	Mm.259916	1	1.204	0.929	7.545
Matn2	matrilin 2	Mm.396856	1	1.157	0.981	7.464
Figf	c-fos induced growth factor	Mm.297978	1	1.346	1.035	6.688
Lce1f	late cornified envelope 1F	Mm.158176	1	0.517	1.230	6.478
Krt1	keratin 1	Mm.183137	1	0.553	0.665	6.243
Lce1d	late cornified envelope 1D	Mm.176243	1	0.346	1	5.978
Fgf13	fibroblast growth factor 13	Mm.7995	1	1.259	1.031	4.860
Col4a1	collagen, type IV, alpha 1	Mm.738	1	1.337	1.267	4.724
Pcdh21	protocadherin 21	Mm.156506	1	1.588	1.100	4.693
Lgals9	lectin, galactose binding, soluble 9	Mm.341434	1	0.972	0.888	4.583
Tgfbi	transforming growth factor, beta induced	Mm.14455	1	1.471	0.994	4.552
Vav3	vav 3 oncogene	Mm.282257	1	1.024	0.901	4.423
Rbbp8	retinoblastoma binding protein 8	Mm.154275	1	1.415	1.230	4.115
Igfbp3	insulin-like growth factor binding protein 3	Mm.29254	1	0.500	0.593	3.989
Igfbp4	insulin-like growth factor binding protein 4	Mm.233799	1	1.617	1.002	3.858
Dbp	D site albumin promoter binding protein	Mm.378235	1	1.277	1.215	3.745
Amot	angiomotin	Mm.100068	1	1.184	1.044	3.586
Lce1a2	late cornified envelope 1A2	Mm.279773	1	0.414	1.280	3.577
Irx4	Iroquois related homeobox 4 (Drosophila)	Mm.103784	1	1.374	0.775	3.482
Fxyd6	FXYD domain-containing ion transport regulator 6	Mm.208287	1	0.940	0.949	3.478
Pthlh	parathyroid hormone-like peptide	Mm.28440	1	0.660	0.859	3.417
Lce1i	late cornified envelope 1I	Mm.291782	1	0.461	1.093	3.270
Vldlr	very low density lipoprotein receptor	Mm.4141	1	1.508	1.332	3.227

Table S2. Cont.

Down-regulated genes			Fold (compared with f/f, Cre$^-$ + EtOH)			
Gene symbol	Gene name	Unigene (Avadis)	f/f, Cre$^-$ + EtOH	f/f, Cre$^-$ + 4OHT	f/f, Cre$^+$ + EtOH	f/f, Cre$^+$ + 4OHT
Prdm1	PR domain containing 1, with ZNF domain	Mm.4800	1	0.395	1.219	3.218
Fhl1	four and a half LIM domains 1	Mm.3126	1	1.210	1.237	3.188
Ear5	eosinophil-associated, ribonuclease A family, member 5	Mm.377125	1	0.170	1.328	3.179
Stbd1	starch binding domain 1	Mm.21965	1	1.428	1.088	3.090
Sostdc1	sclerostin domain containing 1	Mm.43375	1	1.039	1.135	3.060
Lce3a	late cornified envelope 3A	Mm.387664	1	0.551	1.468	3.036
Hrnr	hornerin	Mm.208047	1	0.350	1.113	3.003
Antxr1	anthrax toxin receptor 1	Mm.232525	1	1.137	1.054	2.955
Adh7	alcohol dehydrogenase 7 (class IV), mu or sigma polypeptide	Mm.8473	1	1.189	1.164	2.906
Cyp2s1	cytochrome P450, family 2, subfamily s, polypeptide 1	Mm.275188	1	0.804	0.861	2.886
Fgfr3	fibroblast growth factor receptor 3	Mm.6904	1	1.210	1.075	2.853
Serpinb1a	serine (or cysteine) peptidase inhibitor, clade B, member 1a	Mm.20144	1	0.686	0.767	2.844
Eppk1	epiplakin 1	Mm.259929	1	1.021	1.313	2.839
Lce1b	late cornified envelope 1B	Mm.291769	1	0.384	1.154	2.838
Psmb8	proteasome (prosome, macropain) subunit, beta type 8 (large multifunctional peptidase 7)	Mm.180191	1	0.788	0.954	2.832
Pygl	liver glycogen phosphorylase	Mm.256926	1	0.991	0.882	2.810
Col5a2	collagen, type V, alpha 2	Mm.10299	1	1.303	1.288	2.804
Acss2	acyl-CoA synthetase short-chain family member 2	Mm.255026	1	1.200	1.096	2.790
Mmp2	matrix metallopeptidase 2	Mm.29564	1	1.176	1.113	2.743
Steap4	STEAP family member 4	Mm.31403	1	0.857	0.671	2.729
Oplah	5-oxoprolinase (ATP-hydrolysing)	Mm.322738	1	0.785	0.816	2.729
Cyp39a1	cytochrome P450, family 39, subfamily a, polypeptide 1	Mm.376968	1	1.133	0.929	2.722
Phyh	phytanoyl-CoA hydroxylase	Mm.391704	1	0.787	0.742	2.700
Agrp	agouti related protein	Mm.441696	1	0.655	1.008	2.625
Ctnnal1	catenin (cadherin associated protein), alpha-like 1	Mm.218891	1	1.044	0.765	2.621
Ahcyl2	S-adenosylhomocysteine hydrolase-like 2	Mm.210899	1	1.165	0.931	2.596
Rgn	regucalcin	Mm.2118	1	0.950	1.062	2.591
Osmr	oncostatin M receptor	Mm.10760	1	1.070	1.099	2.548
Il13ra1	interleukin 13 receptor, alpha 1	Mm.24208	1	0.854	0.795	2.537
Ctsc	cathepsin C	Mm.322945	1	0.980	0.943	2.533
Socs3	suppressor of cytokine signaling 3	Mm.3468	1	1.251	1.229	2.514
Cyp1b1	cytochrome P450, family 1, subfamily b, polypeptide 1	Mm.214016	1	0.630	0.928	2.478
Gas1	growth arrest specific 1	Mm.22701	1	1.171	0.861	2.470
Il6st	interleukin 6 signal transducer	Mm.4364	1	0.911	0.865	2.438
Hfe	hemochromatosis	Mm.2681	1	1.092	1.083	2.433
Prkcdbp	protein kinase C, delta binding protein	Mm.3124	1	0.792	0.841	2.430
Flot1	flotillin 1	Mm.2931	1	0.897	0.950	2.418
Lce1h	late cornified envelope 1H	Mm.23784	1	0.485	1.061	2.410
Saa3	serum amyloid A 3	Mm.14277	1	0.305	0.369	2.398
Sord	sorbitol dehydrogenase	Mm.371580	1	0.945	0.920	2.396
Atp1a1	ATPase, Na+/K+ transporting, alpha 1 polypeptide	Mm.280103	1	1.009	0.881	2.388
Zfp238	zinc finger protein 238	Mm.330700	1	1.104	0.937	2.380
Mgst1	microsomal glutathione S-transferase 1	Mm.14796	1	0.921	0.805	2.371
Gata3	GATA binding protein 3	Mm.313866	1	1.175	1.127	2.369
Clybl	citrate lyase beta like	Mm.34608	1	1.141	1.070	2.326
Tbx1	T-box 1	Mm.295194	1	0.753	0.912	2.325
Lpo	lactoperoxidase	Mm.41236	1	0.488	0.951	2.313
Idh1	isocitrate dehydrogenase 1 (NADP+), soluble	Mm.9925	1	0.895	0.918	2.306
Fzd6	frizzled homolog 6 (Drosophila)	Mm.4769	1	1.062	0.883	2.292
Gstm1	glutathione S-transferase, mu 1	Mm.37199	1	1.060	1.083	2.245
Glul	glutamate-ammonia ligase (glutamine synthetase)	Mm.210745	1	1.016	0.747	2.211
Cyp4b1	cytochrome P450, family 4, subfamily b, polypeptide 1	Mm.1840	1	0.715	0.746	2.202
Adamts1	a disintegrin-like and metallopeptidase (reprolysin type) with thrombospondin type 1 motif, 1	Mm.1421	1	1.071	1.028	2.188
Maoa	monoamine oxidase A	Mm.21108	1	0.958	1.050	2.183
Plxdc1	plexin domain containing 1	Mm.39617	1	0.947	0.907	2.173