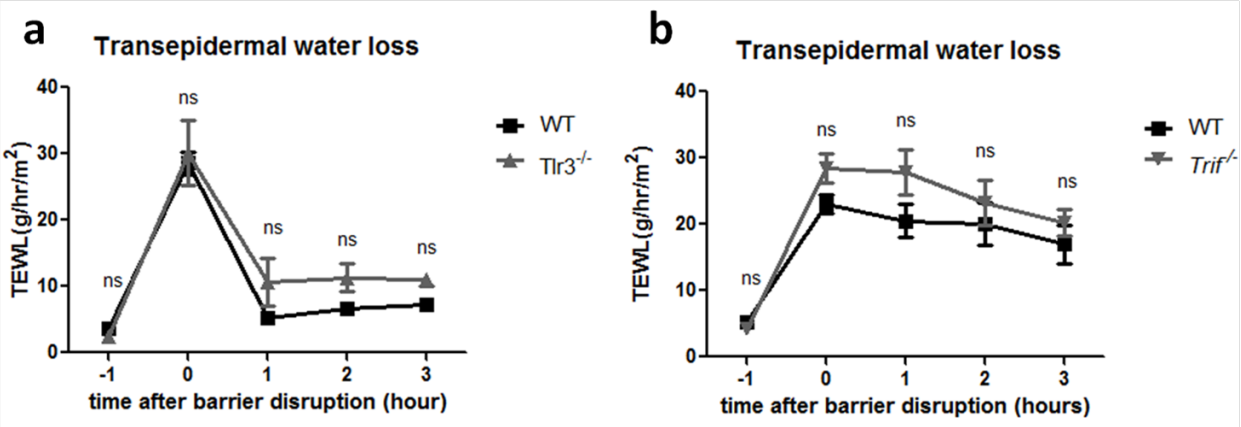
Supplementary Figure 1:



Supplementary Figure 1: *Tlr3*$^{-/-}$ and *Trif*$^{-/-}$ mice show no barrier defect after chemical depilation or tape stripping barrier disruption TEWL was measured in mice at hourly intervals after barrier disruption using a chemical depilatory reagent (a). n = 3. Two-way ANOVA. TEWL was measured in mice at hourly intervals after barrier disruption by tape stripping (b). n = 6. Two-way ANOVA. Data are mean +/- SEM and are representative of at least three independent experiments. ns = $P > 0.05$.

Supplementary Figure 2:

Supplementary Figure 2: *Tlr3*$^{-/-}$ mice exhibit chronic non-healing wounds after UVB damage

WT and *Tlr3*$^{-/-}$ mice were photographed at 8 and 16 weeks after exposure to a single 5 kJ/m^2 dose of UVB. Photographs are from two separate experiments.

SUPPLEMENTARY MATERIALS & METHODS

Cell culture and stimuli

NHEKs were obtained from Life Technologies (catalog number: C-001-5C; Carlsbad, CA), and grown in serum-free EpiLife cell culture media (Life Technologies, Carlsbad, CA) containing 0.06 mM Ca^{2+} and 1 × EpiLife Defined Growth Supplement (EDGS, Life Technologies, Carlsbad, CA) at 37 °C under standard tissue culture conditions. All cultures were maintained for up to eight passages in this medium with the addition of 100 U ml^{-1} penicillin, 100 µg ml^{-1} streptomycin, and 250 ng ml^{-1} amphotericin B. Cells at 60–80% confluence were treated with Poly (I:C) (1 µg/ml; Invivogen, San Diego, CA) or U1 RNA (1 µg/ml; kindly donated from Dr. Eric L. Greidinger (University of Miami and the Miami VA Medical Center; Miami, Florida)) in 12-well flat-bottom plates (Corning Incorporated Life Sciences, Lowell, MA) for up to 24 hours. Cells treated with *in-vitro* transcribed snRNA (U2, U4, U6, U12, scaRNA9, and scaRNA18) were also treated with 2.5 µl/ml Dharmafect 1 (Thermo Scientific, Pittsburg, PA) during treatments per manufacturer's instructions. After cell stimulation, RNA was extracted using TRIzol reagent (Invitrogen, Carlsbad, CA). RNA was stored at −80 °C.

Quantitative real-time PCR

Total RNA was extracted from cultured keratinocytes using TRIzol Reagent (Invitrogen) and 1 µg RNA was reverse-transcribed using iScript cDNA Synthesis Kit (Bio-Rad, Hercules, CA). Pre-developed TaqMan Gene Expression Assays (Applied Biosystems, Foster City, CA) were used to evaluate mRNA transcript levels of ABCA12, GBA, SMPD1, TGM1, TNF, IL-6, CDSN, TJP1, OCLN, CLDN1, DSG1, DSG3, PKP1, DSP, JUP, DSC1, DSC2, CLDN4, CLDN5, CLDN7, CLDN11, CLDN23, and TLR3. Glyceraldehyde-3-phosphate dehydrogenase mRNA transcript levels were evaluated using a VIC-CATCCATGACAACTTTGGTA-MGB probe with primers 5′-CTTAGCACCCCTGGCCAAG-3′ and 5′-TGGTCATGAGTCCTTCCACG-3′. All analyses

were performed in triplicate and were representative of three to five independent cell stimulation experiments that were analyzed in an ABI Prism 7000 Sequence Detection System (Life Technologies, Carlsbad, CA). Fold induction relative to glyceraldehyde-3-phosphate dehydrogenase was calculated using the $\Delta\Delta C_t$ method. Results were considered to be significant if $P<0.05$.

Transepithelial Electric Resistance (TEER)

Primary human keratinocytes (PHK) were isolated from discarded neonatal foreskins. PHK were plated in Keratinocyte-SFM (Life Technologies, Carlsbad, CA) in 24-well *Costar® Transwell inserts* (polyester membranes, *0.4*-µm pore size; *Corning Life Sciences, Corning, NY).* After cells were confluent, media was switched to Dulbecco modified Eagle medium (Life Technologies, Carlsbad, CA) with 10% heat-inactivated FBS (Life Technologies, Carlsbad, CA), 1% Pen/Strep and 0.2% Amphotericin B (Life Technologies, Carlsbad, CA) allowing PHK differentiation and TJ formation. At the same time, TLR3 ligand, Poly (I:C) *(Amersham/*GE Healthcare, Piscataway, NJ) was placed in upper wells for 8 days. Culture media was changed every other day. TEER was measured with an EVOMX voltohmmeter (World Precision Instruments, Sarasota, Fla) which has probes that measure the resistance from the upper chamber in the transwell insert through differentiated keratinocytes into the lower chamber of the cell culture plate. As tight junction function increases, so do resistance measurements. The resistance of cell-free filters was subtracted from each experimental value. The study was approved by the Research Subject Review Board at the University of Rochester Medical Center and was conducted according to Declaration of Helsinki Principles (De Benedetto et al. 2011).

Paracellular flux assay

PHK were seeded in Transwell inserts and treated as described above. After 48 h, 0.02% fluorescein sodium (Sigma-Aldrich) in PBS was added to the upper well, while PBS alone was added to the lower

well. Samples were collected from the lower well after 30 minutes. The amount of fluorescein sodium that diffused from across the filter was measured with the iQ5 Multicolor real-time PCR detection system (Bio-Rad). Paracellular flux was presented as follows: Paracellular flux (fold of control)= fluorescein intensity of treatment groups/ fluorescein intensity of control group (Kuo et al. 2013).

siRNA knockdown of gene expression

TLR3 and control siRNA were purchased from Dharmacon (Chicago, IL). One nanomole of each siRNA was electroporated into 3 x 10^6 keratinocytes using Amaxa nucleofection reagents as previously described (Borkowski et al. 2013) (VPD-1002) (Lonza AG, Walkersville, MD), 48 hours prior to cell culture treatments.

snRNA and Primer Sequences

U1 RNA Sequence (164 bp): atacttacct ggcaggggag ataccatgat cacgaaggtg gttttcccag ggcgaggctt atccattgca ctccggatgt gctgacccct gcgatttccc caaatgtggg aaactcgact gcataatttg tggtagtggg ggactgcgtt cgcgctttcc cctg

U1 Forward Primer (*T7*):

*TAATACGACTCACTATAGGG*ATACTTACCTGGCAGGGGAGA

U1 Reverse Primer:

CAGGGGAAAGCGCGA

U2 RNA Sequence (188 bp): atcgcttctc ggccttttgg ctaagatcaa gtgtagtatc tgttcttatc agtttaatat ctgatacgtc ctctatccga ggacaatata ttaaatggat ttttggagca gggagatgga ataggagctt gctccgtcca ctccacgcat cgacctggta ttgcagtacc tccaggaacg gtgcaccc

U2 Forward Primer (T7):

*TAATACGACTCACTATAGGG*ATCGCTTCTCGGCCTTTT

U2 Reverse Primer:

GGGTGCACCGTTCCTG

U4 RNA Sequence (144 bp): agctttgcgc agtggcagta tcgtagccaa tgaggtctat ccgaggcgcg attattgcta attgaaaact tttcccaata ccccgccgtg acgacttgca atatagtcgg cactggcaat ttttgacagt ctctacggag actg

U4 Forward Primer (*T7*):

*TAATACGACTCACTATAGGG*AGCTTTGCGCAGTGGC

U4 Reverse Primer:

TCTCCGTAGAGACTGTCAAAAATTG

U6 RNA Sequence (106 bp): gtgctcgctt cggcagcaca tatactaaaa ttggaacgat acagagaaga ttagcatggc ccctgcgcaa ggatgacacg caaattcgtg aagcgttcca tatttt

U6 Forward Primer (T7):

*TAATACGACTCACTATAGGG*GTGCTCGCTTCGGCAG

U6 Reverse Primer:

AAAAATATGGAACGCTTCACG

U12 RNA Sequence (149 bp): tgccttaaac ttatgagtaa ggaaaataac gattcggggt gacgcccgaa tcctcactgc taatgtgaga cgaatttttg agcgggtaaa ggtcgccctc aaggtgaccc gcctactttg cgggatgcct gggagttgcg atctgcccg

U12 Forward Primer (T7):

*TAATACGACTCACTATAGGG*TGCCTTAAACTTATGAGTAAGGAAAAT

U12 Reverse Primer:

CGGGCAGATCGCAACT

SCARNA9 Sequence (353 bp): ctttctgaga tctgctttta gtgaagtgga tcaatgatga aactagccaa atctgagcat cagaagtctt tccagtctac ctgatgcatg atctctacag ttctgagaag caaaactata aaacaatgta aaacaataag ggcatatgtc tggtgtgtgt gtgtgtgtgt gtgtgtgtgt gtgtgtgtac gcacatgtgt ttataaagat aacagctgta ggaatgaatg agattgaggg tgggggggtg cgtatgtatg tctatgaaag cctaatcatt tctgggcaat gatgaaaagg ttttactact gatctttgta actatgatgg tttctacact tgacctgagc tca

SCARNA9 Forward Primer (T7):

*TAATACGACTCACTATAGGG*CTTTCTGAGATCTGCTTTTAGTGAAGT

SCARNA9 Reverse Primer:

TGAGCTCAGGTCAAGTGTAGAAAC

SCARNA18 Sequence (134 bp): ttgcatgtgg aaatgtctgc ttctcattcc ttgggagcag gaatatgttc ataacatgct acattaacaa aggagttctc agggctgcca accttctagt aaaggttgag tggtagtata tttctccaac ataa

SCARNA18 Forward Primer (T7):

*TAATACGACTCACTATAGGG*TTGCATGTGGAAATGTCTGC

SCARNA18 Reverse Primer:

TTATGTTGGAGAAATATACTACCACTCAAC

Secondary RNA structure generation

In order to depict the secondary structure of the snRNAs in this manuscript, sequences were taken from Pubmed or UCSC genome browser. These sequences were then analyzed for complementary base pairing using RNAfold (http://rna.tbi.univie.ac.at/cgi-bin/RNAfold.cgi). The following filters were selected to obtain the secondary structure information:

(1) minimum free energy (MFE) and partition function

(2) avoid isolated base pairs

The secondary structure and sequence information was then used with the VARNA applet (http://varna.lri.fr/) to generate the secondary structure diagrams (Gruber et al. 2008; Blin et al. 2009)

Ultrastructural Analysis

Mouse dorsal skin was excised 24 hours after UVB exposure and then immersed in modified Karnovsky's fixative (2.5% glutaraldehyde and 2% paraformaldehyde in 0.15 M sodium cacodylate buffer, pH 7.4) for at least 4 hours, post fixed in 1% osmium tetroxide in 0.15 M cacodylate buffer for 1 hour and stained en bloc in 3% uranyl acetate for 1 hour. Samples were dehydrated in ethanol, embedded in Durcupan epoxy resin (Sigma-Aldrich), sectioned at 50 to 60 nm on a Leica UCT ultramicrotome, and picked up on Formvar and carbon-coated copper grids. Sections were stained with 3% uranyl acetate for 5 minutes and Sato's lead stain for 1 minute. Grids were viewed using a JEOL 1200EX II (JEOL, Peabody, MA) transmission electron microscope and photographed using a Gatan digital camera (Gatan, Pleasanton, CA), or viewed using a Tecnai G2 Spirit BioTWIN transmission electron microscope equipped with an Eagle 4k HS digital camera (FEI, Hilsboro, OR). Pictures taken at 2000X represent 73 μm^2. Samples were also sectioned and stained with Toluidine Blue.

Cytokine/Chemokine Analysis

8 mm biopsy punches were taken from the dorsal skin of UVB exposed mice 24 hours after UVB exposure (5 kJ/m^2) and homogenized in 700 ul RIPA buffer (50mM Tris, 150 mM NaCl, 0.1% SDS, 0.5% sodium deoxycholate, 1% triton X-100) with 1X cOmplete Protease Inhibitor Cocktail (11697498001, Roche, Basel, Switzerland) using a MiniBeadBeater-16, Model 607 (BioSpec Products Inc., Bartlesville, OK). BCA Protein Assay Kit (Thermo Scientific Pierce, San Diego, CA) was used to measure protein concentration. 10 ug of total protein was used for analysis using MILLIPLEX MAP Mouse

Cytokine/Chemokine Magnetic Bead Panel - Immunology Multiplex Assay (MCYTOMAG-70K, Millipore, Billerica, MA).