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Appendix S1

SUPPLEMENTARY MATERIALS AND METHODS

Skin biopsies

Written informed consent, according to the Declaration of Helsinki principles, was obtained from all patients involved in the study. The National Medical Research Council approved the study. Skin biopsies (8-mm punch biopsies) were collected from the skin lesions of 12 patients with psoriasis, including 6 samples from scalp psoriasis and 6 from psoriasis vulgaris of SGP skin (Table SI[1]). Part of each biopsy was paraffin-embedded and used for immunohistochemistry (IHC) and part of each biopsy was stored in RNAlater (Qiagen, Hilden, Germany) at –70°C until RNA isolation for RT-PCR.

Immunohistochemistry staining

For IHC analyses, freshly prepared paraffin-embedded sections from scalp psoriasis and from psoriasis vulgaris of SGP skin were used. After samples were deparaffinized and rehydrated, samples were incubated in 3% H_2O_2 for 15 min to eliminate endogenous peroxidase activity, followed by heat-induced antigen retrieval. After blocking in 1% bovine serum albumin (BSA) solution, sections were incubated overnight at 4°C with primary antibodies against human CD4 (rabbit monoclonal IgG [ab133616]: Abcam, Cambridge, UK), human CD11c (rabbit monoclonal IgG [ab52632]: Abcam), human CD83 (mouse monoclonal IgG [ab123494]: Abcam), human CD1a (mouse monoclonal IgG [AM33361PU-T]: Acris, Rockville, MD, USA), human IL-17 (rabbit polyclonal IgG [bs-2140R]: Bioss Antibodies, Woburn, MA, USA), human IL-23 (rabbit polyclonal [PA5-20239]: Thermo Fisher, Rockford, IL, USA), human IFN-gamma (rabbit polyclonal [NBP1-19761]: Novus Biologicals, Littleton, CO, USA), human TNF-alfa (mouse monoclonal IgG [SAB1404480-100UG]: Sigma-Aldrich, St. Louis, MO , USA), human CCL2/MCP1 (mouse monoclonal IgG1 [NBP2-22115]: Novus Biologicals), human CCL20/MIP-3-α (mouse monoclonal IgG [LS-B7409]: LifeSpan Biosciences, Seattle WA, USA), human lipocalin/NGAL (rabbit polyclonal IgG [PA5-32476]: Invitrogen, Life Technologies, San Francisco, CA, USA), human S100A8 (rabbit polyclonal IgG [HPA024372]: Sigma-Aldrich), human loricrin (rabbit monoclonal IgG [NBP1-33610]: Novus Biologicals), human filaggrin (mouse monoclonal IgG: [ab17808]: Abcam), and human KRT17 (rabbit polyclonal IgG [ab53707]: Abcam). After primary antibody incubation, anti-mouse/rabbit HRP-conjugated secondary antibodies (Dako, Agilent Technologies, Santa Clara, CA, USA) were applied. Washing steps before and after incubating with antibodies were performed with TBST for 5 min, 3 times per step. Vector® VIP and ImmPACT™ NovaRED™ Kits (VECTOR Laboratories, Burlingame, CA, USA) were used to detect signals. Sections were background stained with methylene green. The detection of each protein was carried out on all sections in parallel at the same time, so that we could evaluate comparable protein levels.

RNA isolation, reverse transcription, and real-time quantitative PCR

Total RNA was isolated using TriReagent solution (Sigma-Aldrich) with Tissue Lyser (QIAGEN, Hilden, Germany) and previously autoclaved metal beads (QIAGEN). RNA concentration and purity were assessed with a NanoDrop spectrophotometer (Thermo Scientific, Bioscience, Budapest, Hungary) and an Agilent 2100 Bioanalyser (Agilent Technologies, Santa Clara, CA, USA), respectively. Samples were treated with DNase I (Applied Biosystems, Foster City, CA, USA). The reverse transcription step (1 µg RNA) was performed using a high capacity cDNA Archive Kit (Invitrogen) according to the manufacturer's instructions and the indicated thermal protocol. The qRT-PCR measurements were conducted in triplicate using pre-designed FAM-MGB assays and TaqMan® Gene Expression Master Mix from Applied Biosystems (Life Technologies).

The following primers were used: PPIA (Hs99999904_m1), IL-17A (Hs00174383_m1), IL-1β (Hs00174097_m1), IL-12B (Hs01011518_m1), IL-23 (Hs00900829_g1), IFNγ (Hs00174143_m1), TNFα (Hs00174128_m1), CCL2 (Hs00234140_m1), CCL20 (Hs00355476_m1), S100A7 (Hs00161488_m1), S100A8 (Hs00374264_g1), S100A9 (Hs00610058_m1), DEFB4B (hBD-2) (Hs00175474_m1), LCN2 (Hs01008571_m1), FLG (Hs00856927_g1), KRT17 (Hs00356958_m1), and KRT6A (Hs01699178_g1).

Reactions were performed with a LightCycler® 480 System (Roche, Basel, Switzerland). Using either the comparative Ct method or standard curves, the relative mRNA levels were calculated and normalized to the expression of PPIA mRNA.

Statistical analysis

The Kolmogorov–Smirnov test was used to assess the distribution of data. In the case of normal distribution, independent *t*-tests were used. In the absence of normal distribution, Mann–Whitney *U* tests were used for statistical comparison of 2 experimental data. *p*-values < 0.05 were considered statistically significant (*$p<0.05$; **$p<0.01$; ***$p<0.001$). Statistical analyses were performed using GraphPad Prism software version 7 (GraphPad Software Inc., San Diego, CA, USA) and SPSS 25 (SPSS package for Windows, Release 25; SPSS Inc., Chicago, IL, USA).