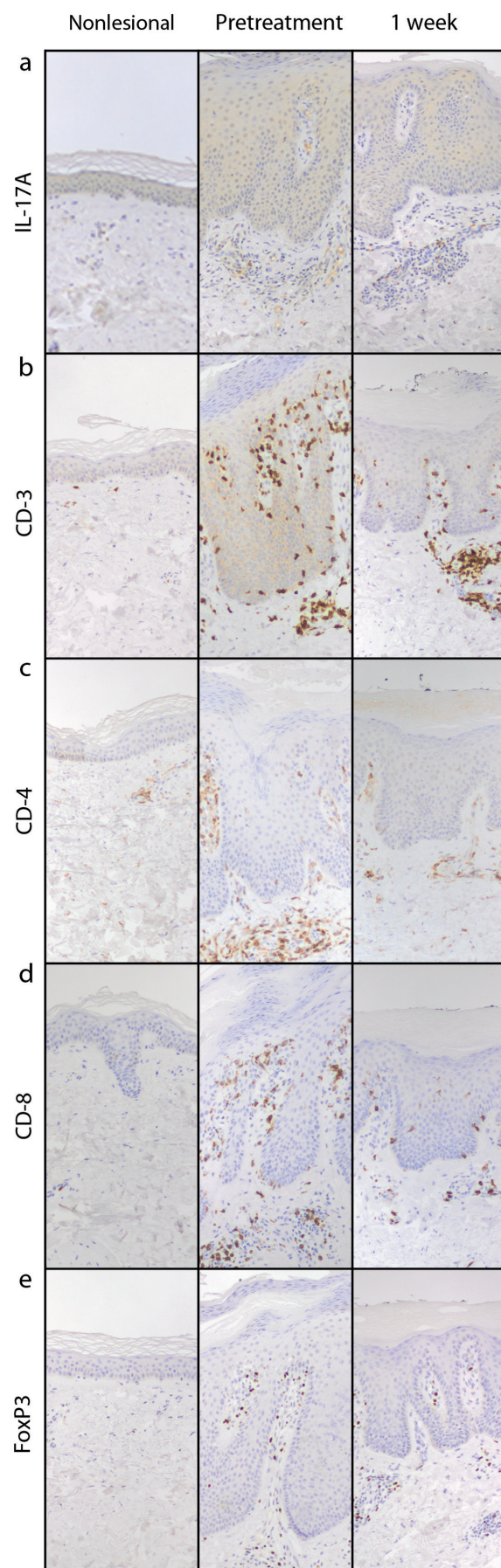
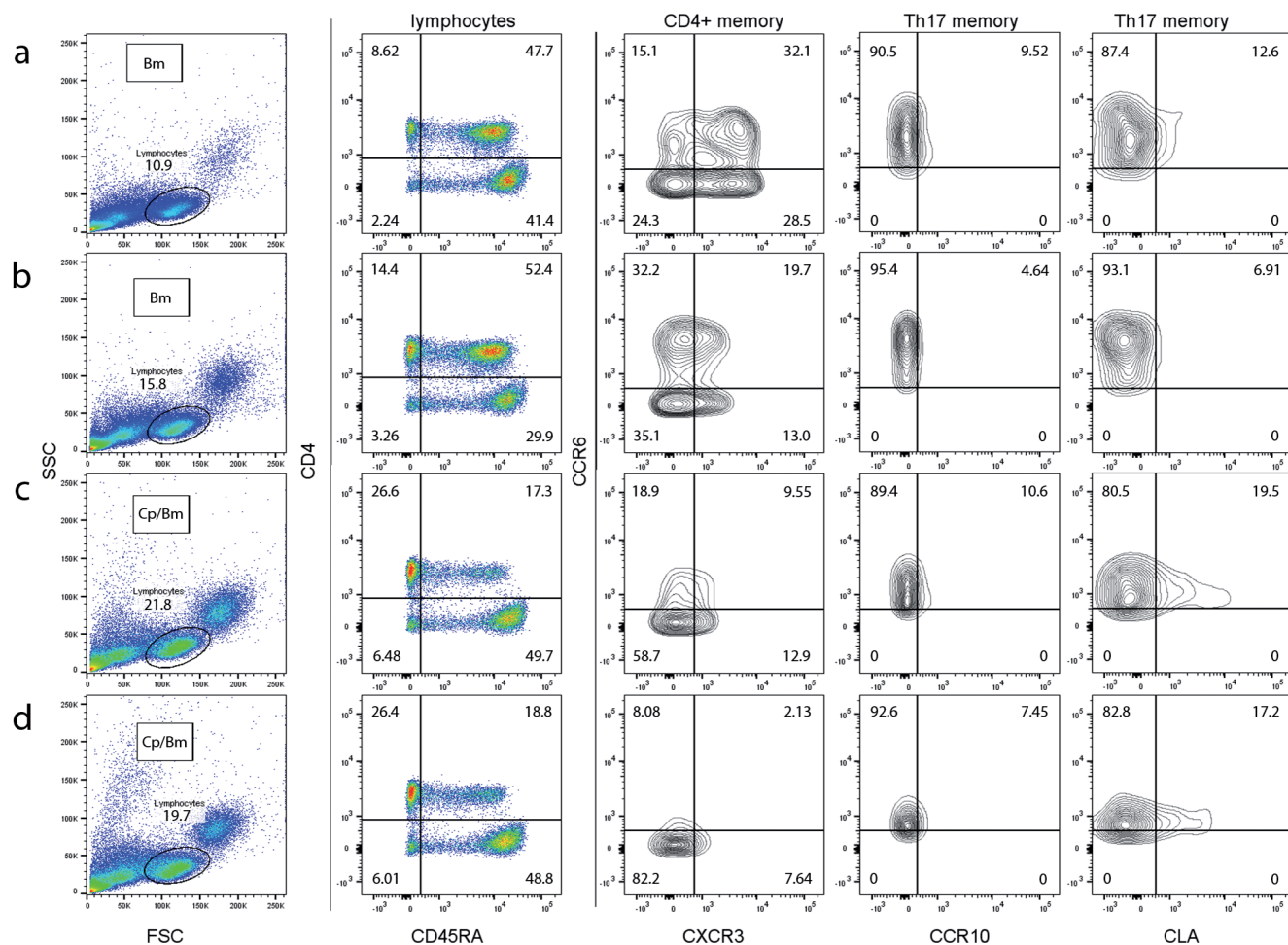
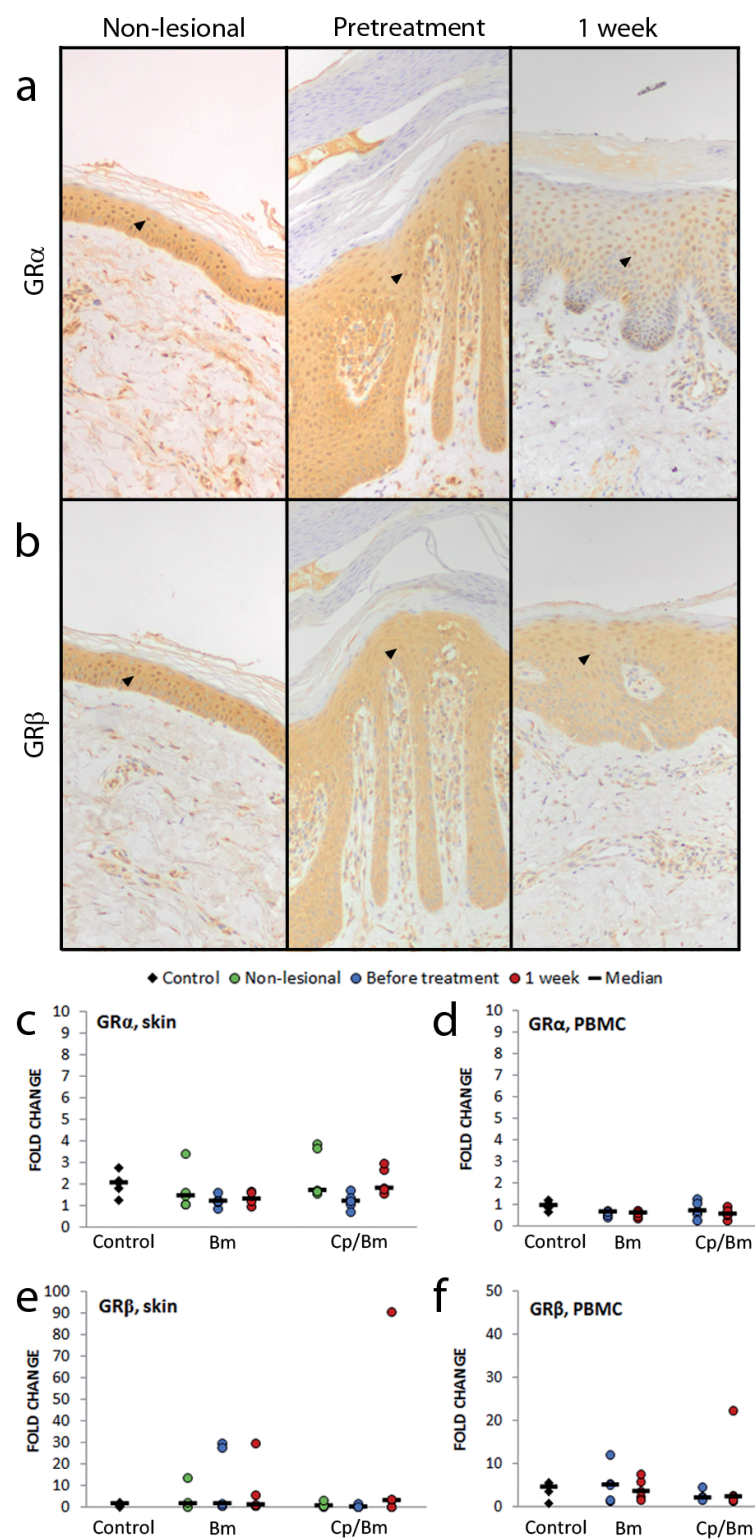




**Fig. S1. Immunohistochemical staining of skin biopsies from a psoriatic patient; non-lesional healthy skin (left-hand column), pretreatment lesional sample (middle column) and lesional sample (right-hand column) after one week of therapy with calcipotriol/betamethasone ointment.** (a) Staining with anti-IL-17A showed no positive cells in the epidermis in these skin samples. In the dermis, there were more positive cells in lesional than non-lesional skin samples. After the treatment period with calcipotriol/betamethasone, the number of IL-17A positive cells in the dermis increased. Staining with (b) anti-CD3, (c) anti-CD4 and (d) anti-CD8 showed greater numbers of positive cells in psoriatic lesional samples compared with non-lesional samples in both epidermis and dermis. Calcipotriol/betamethasone reduced the numbers of CD3<sup>+</sup>, CD4<sup>+</sup> and CD8<sup>+</sup> cells in the epidermis and dermis. (e) Staining with anti-FoxP3 was negative in epidermis in the non-lesional sample, few cells were positive in the dermis. Positive cells were found in both epidermis and dermis in lesional samples, calcipotriol/betamethasone reduced the numbers of positive cells.



**Fig. S2. The numbers of Th17 and Th22 cells in peripheral blood mononuclear cell (PBMC) samples analysed by flow cytometry.** As representative samples, PBMC samples from a psoriatic patient (a) before and (b) after one week of betamethasone monotherapy and samples from another psoriatic patient (c) before and (d) after 1 week of calcipotriol/betamethasone combination therapy are shown. CD4<sup>+</sup> T cells were selected from PBMC samples (*first column*). Memory CD4<sup>+</sup> T cells were identified as CD4<sup>+</sup>, CD45RA<sup>-</sup> T cells (*second column*). Th17 memory cells were identified from memory CD4<sup>+</sup> T cells as CD4<sup>+</sup>, CD45RA<sup>-</sup>, CXCR3<sup>-</sup>, CCR6<sup>+</sup> cells (*third column*). Th22 memory cells were identified from Th17 memory cells as CD4<sup>+</sup>, CD45RA<sup>-</sup>, CXCR3<sup>-</sup>, CCR6<sup>+</sup>, CCR10<sup>+</sup> cells and (*fourth column*) and skin-homing Th17 cells as CD4<sup>+</sup>, CD45RA<sup>-</sup>, CXCR3<sup>-</sup>, CCR6<sup>+</sup>, CLA<sup>+</sup> cells (*fifth column*). Both topical therapies reduced the numbers of Th22 cells (*fourth column*: combination therapy from 10.6% to 7.45% and monotherapy from 9.52% to 4.64%) and skin homing CLA<sup>+</sup> Th17 cells (*fifth column*: combination therapy from 19.5% to 17.2% and monotherapy from 12.6% to 6.91%).



**Fig. S3. Expression of glucocorticoid receptor (GR) isoforms in skin and peripheral blood mononuclear cell (PBMC) samples from psoriatic patients and healthy controls.** Immunohistochemical staining of skin biopsies from a psoriatic patient; non-lesional healthy skin (*left-hand column*), pretreatment lesional sample (*middle column*) and lesional sample (*right-hand column*) after one week of therapy with calcipotriol/betamethasone combination. (a) Epidermal nuclear staining with the GR $\alpha$  antibody (arrowheads) was more prominent in psoriatic lesions than in non-lesional samples. (b) Staining with GR $\beta$  antibody showed no difference. Combination therapy decreased the nuclear staining of keratinocytes with GR $\alpha$  and GR $\beta$  antibodies (*right-hand column, a, b, arrowheads*). (c, d) Expression of GR $\alpha$  mRNA in skin and PBMC samples. (e, f) Expression of GR $\beta$  mRNA in skin and PBMC samples. mRNA levels were measured using quantitative real-time PCR. Control: healthy control; Non-lesional: skin biopsy from non-lesional skin of patient with psoriasis; Before treatment: skin biopsy from untreated lesional skin/PBMC sample before treatment; 1 week: skin biopsy/PBMC sample taken after one week of treatment with either betamethasone or calcipotriol/betamethasone; fold change: the normalized expression level compared with normal skin/PBMCs.

**Table SI. Primers used for quantitative real-time PCR**

| Primer name                      | Gene bank access number | 5'-3' sequence           |
|----------------------------------|-------------------------|--------------------------|
| IL-6 upstream primer             | NM_000600               | GGCACTGGCAGAAAACAACC     |
| IL-6 downstream primer           |                         | GCAAGTCTCCTCATTGAATCC    |
| IL-17A upstream primer           | NM_002190               | CTCATTGGTGCTCACTGCTACTG  |
| IL-17A downstream primer         |                         | CCTGGATTTCGTGGGATTGTG    |
| IL-23A upstream primer           | NM_016584               | GACTACAAATGATGTTCCCC     |
| IL-23A downstream primer         |                         | CCTGTGAAAATATCCGATCC     |
| IFN- $\gamma$ upstream primer    | NM_000619               | TAATGCAGGTCATTGAGATG     |
| IFN- $\gamma$ downstream primer  |                         | CTCTCCTCTTTCCAATTCTTC    |
| TNF- $\alpha$ upstream primer    | NM_000594               | CTCAGCCTCTCTCCTTC        |
| TNF- $\alpha$ downstream primer  |                         | AGAAGATGATCTGACTGCC      |
| T-bet upstream primer            | NM_013351               | TCCAACACGCATATCTTTAC     |
| T-bet downstream primer          |                         | TTGGCAAAGGGGTTATTATC     |
| FoxP3 upstream primer            | NM_001114377            | CTTCATCTGTGGCATCATC      |
| FoxP3 downstream primer          |                         | ATGTTGTGGAGGAAGCTCTG     |
| S100A7 upstream primer           | NM_002963               | TTAGTGCCTGTGACAAAAG      |
| S100A7 downstream primer         |                         | GTAAGTCTGTGGCTATGCTC     |
| CCL-20 upstream primer           | NM_001130046            | TATATTGTGCGTCTCCTCAG     |
| CCL-20 downstream primer         |                         | GCTATGTCCAATTCCATTCC     |
| hGrA upstream primer             | X03225                  | CTATGCATGAAGTGGTTGAAAA   |
| hGrA downstream primer           |                         | TTTCAGCTAACATCTCGGG      |
| hGr $\beta$ upstream primer      | X03348.1                | GAAGGAACTCCAGCCAGAA      |
| hGr $\beta$ downstream primer    |                         | CCACATAACATTTTCATGCATAGA |
| GAPDH upstream primer            | NM_002046               | GGAGCCAAAAGGGTCATCATC    |
| GAPDH downstream primer          |                         | GTCATGAGTCCTTCCACGATA    |
| $\beta$ -actin upstream primer   | NM_001101.3             | AGAGCTACGAGCTGCCTGAC     |
| $\beta$ -actin downstream primer |                         | AGCACTGTGTTGGCGTACAG     |

**Table SII. Antibodies used for immunohistochemical staining of skin biopsies**

| Antibody name | Catalogue number and producer |                                                               | Dilution |
|---------------|-------------------------------|---------------------------------------------------------------|----------|
| CD3           | Monoclonal mouse, antihuman   | NCL-CD3-PS1, Novocastra, Newcastle, UK                        | 1:50     |
| CD4           | Monoclonal mouse, antihuman   | NCL-CD4-368, Novocastra, Newcastle, UK                        | 1:40     |
| CD8           | Monoclonal mouse, antihuman   | NCL-CD8-4B11, Novocastra, Newcastle, UK                       | 1:200    |
| FoxP3         | Monoclonal mouse, antihuman   | ab20034, Abcam Ltd., Cambridge, UK                            | 1:100    |
| IL17A         | Polyclonal goat, antihuman    | AF-317-NA, R&D systems, Minneapolis, MN, USA                  | 1:100    |
| GR $\alpha$   | Polyclonal rabbit, antihuman  | sc-1002 (p-20); Santa Cruz Biotechnology, Santa Cruz, CA, USA | 1:500    |
| GR $\beta$    | Polyclonal rabbit, antihuman  | Ab3581, Abcam Ltd, Cambridge, UK                              | 1:100    |

**Table SIII. Clinical characteristics of psoriatic patients and controls**

| Treatment                | Age, years/sex | BMI  | PASI before | PASI after 1 week | Total amount of ointment used, g |
|--------------------------|----------------|------|-------------|-------------------|----------------------------------|
| Combination <sup>a</sup> | 52/F           | 31.4 | 6.4         | 3.6               | 23.4                             |
| Combination              | 56/F           | 19.0 | 13.8        | 8.0               | 73.3                             |
| Combination              | 38/M           | 21.9 | 7.0         | 5.2               | 9.3                              |
| Combination              | 34/M           | 25.0 | 5.4         | 3.6               | 14.7                             |
| Combination              | 57/F           | 24.0 | 12.0        | 9.3               | 37.7                             |
| Median                   |                | 24.0 | 7.0         | 5.2               | 23.4                             |
| Monotherapy <sup>b</sup> | 25/M           | 23.8 | 12.6        | 8.4               | 24.9                             |
| Monotherapy              | 55/M           | 26.5 | 6.5         | 4.3               | 9.4                              |
| Monotherapy              | 55/F           | 39.5 | 10.5        | 9.9               | 27.9                             |
| Monotherapy              | 24/M           | 34.4 | 8.5         | 6.4               | 7.6                              |
| Monotherapy              | 34/F           | 18.0 | 8.6         | 8.4               | 38.0                             |
| Median                   |                | 26.5 | 8.6         | 8.4               | 24.9                             |
| Control                  | 52/M           | 33.1 | –           | –                 | –                                |
| Control                  | 64/F           | 26.7 | –           | –                 | –                                |
| Control                  | 55/F           | 26.0 | –           | –                 | –                                |
| Control                  | 26/M           | 22.9 | –           | –                 | –                                |
| Control                  | 40/F           | 29.4 | –           | –                 | –                                |
| Median                   |                | 26.7 | –           | –                 | –                                |

<sup>a</sup>Calcipotriol/betamethasone. <sup>b</sup>Betamethasone.

BMI: body mass index; PASI: Psoriasis Area Severity Index.

**Table SIV. Analysis of the immunostained skin biopsies of normal controls' skin and psoriatic patients' lesional and non-lesional skin**

| Antibody/localization                     | Control (n = 5)               | Psoriasis lesion (n = 10) | Non-lesional skin (n = 10) | p-value <sup>a</sup> | p-value <sup>b</sup> | p-value <sup>c</sup> |
|-------------------------------------------|-------------------------------|---------------------------|----------------------------|----------------------|----------------------|----------------------|
| <i>Immunopositive cells</i>               |                               |                           |                            |                      |                      |                      |
| IL-17A/epidermis                          | 0 (0–0) <sup>d</sup>          | 0 (0–0.25)                | 0 (0–0)                    | 0.524                | 0.500                | 1.000                |
| IL-17A/dermis                             | 2.0 (1.25–7.5)                | 8.17 (6.13–12.5)          | 3.0 (2.38–4.5)             | 0.036                | 0.004                | 0.657                |
| CD3/epidermis                             | 1.0 (0.5–3.0)                 | 24.5 (13.75–46.0)         | 0 (0–1.0)                  | 0.001                | 0.002                | 0.068                |
| CD3/dermis                                | 21.5 (12.0–26.5)              | 80.42 (44.25–104.75)      | 4.75 (3.38–8.63)           | 0.001                | 0.002                | 0.005                |
| CD4/epidermis                             | 0 (0–1.5)                     | 6.5 (5.0–13.0)            | 0 (0–0)                    | 0.001                | 0.002                | 0.333                |
| CD4/dermis                                | 31.0 (27.1–40.75)             | 125.25 (93.88–160.5)      | 11.25 (9.13–19.75)         | 0.001                | 0.002                | 0.008                |
| CD8/epidermis                             | 0.5 (0–1.0)                   | 15.0 (9.25–39.75)         | 0.5 (0–1.0)                | 0.001                | 0.002                | 1.000                |
| CD8/dermis                                | 13.5 (4.5–17.0)               | 43.5 (36.38–60.38)        | 3.75 (3.38–5.5)            | 0.001                | 0.002                | 0.082                |
| FoxP3/epidermis                           | 0 (0–0)                       | 2.5 (1.0–6.5)             | 0 (0–0)                    | 0.003                | 0.008                | 1.000                |
| FoxP3/dermis                              | 4.0 (3.25–5.0)                | 26.0 (12.0–58.0)          | 2.0 (0.5–3.88)             | 0.001                | 0.002                | 0.072                |
| <i>Immunopositive keratinocyte nuclei</i> |                               |                           |                            |                      |                      |                      |
| GR $\alpha$ /epidermis                    | 50.0 (40.0–60.0) <sup>e</sup> | 80.0 (72.5–90.0)          | 62.5 (47.5–70.0)           | 0.002                | 0.004                | 0.194                |
| GR $\beta$ /epidermis                     | 60.0 (47.5–62.5)              | 55.0 (43.75–61.25)        | 55.0 (45.0–65.0)           | 0.763                | 0.930                | 0.792                |

<sup>a</sup>Control vs. psoriasis, calculated by Mann-Whitney test. <sup>b</sup>Lesional vs. non-lesional skin in same patient, calculated by Wilcoxon signed-rank test. <sup>c</sup>Control vs. non-lesional sample from psoriatic patient, calculated by Mann-Whitney test. <sup>d</sup>Number of immunopositive cells /20 $\times$  field image; median (interquartile range). <sup>e</sup>Percentage of immunopositive keratinocyte nuclei; median (interquartile range).

**Table SV. Analysis of the immunostained skin biopsies of psoriatic patients’ lesional skin before and after treatment**

| Antibody/<br>localization                 | Before treatment                            |                                             | After treatment                |                                | <i>p</i> -value <sup>c</sup> (before vs. after) |             |
|-------------------------------------------|---------------------------------------------|---------------------------------------------|--------------------------------|--------------------------------|-------------------------------------------------|-------------|
|                                           | Combination <sup>a</sup><br>( <i>n</i> = 5) | Monotherapy <sup>b</sup><br>( <i>n</i> = 5) | Combination<br>( <i>n</i> = 5) | Monotherapy<br>( <i>n</i> = 5) | Combination                                     | Monotherapy |
| <i>Immunopositive cells</i>               |                                             |                                             |                                |                                |                                                 |             |
| IL-17A/epidermis                          | 0 (0–0.5) <sup>d</sup>                      | 0 (0–0.5)                                   | 0 (0–0)                        | 0 (0–1.0)                      | 1.000                                           | 1.000       |
| IL-17A/dermis                             | 8.3 (7.25–23.2)                             | 6.5 (4.0–13.3)                              | 12.0 (5.5–16.3)                | 12.5 (3.5–14.3)                | 0.875                                           | 1.000       |
| CD3/epidermis                             | 45.0 (15.5–58.0)                            | 15.0 (13.0–36.0)                            | 5.0 (1.5–36.5)                 | 23.0 (15.0–25.0)               | 0.188                                           | 0.875       |
| CD3/dermis                                | 70.3 (54.25–119.5)                          | 90.5 (37.5–103.25)                          | 50.0 (23.8–74.5)               | 70.5 (40.25–96.5)              | 0.063                                           | 0.813       |
| CD4/epidermis                             | 9.0 (4.0–21.0)                              | 5.0 (5.0–9.5)                               | 2.0 (0.5–3.75)                 | 6.0 (3.0–14.0)                 | 0.063                                           | 1.000       |
| CD4/dermis                                | 150.0 (83.5–205.75)                         | 117.0 (87.75–159.0)                         | 61.5 (36.75–188.0)             | 72.0 (48.75–148.25)            | 0.438                                           | 0.625       |
| CD8/epidermis                             | 15.5 (9.0–41.0)                             | 14.5 (7.25–43.0)                            | 3.0 (1.25–14.0)                | 11.0 (3.5–33.25)               | 0.063                                           | 1.000       |
| CD8/dermis                                | 38.0 (24.0–61.1)                            | 45.0 (40.5–57.5)                            | 33.5 (5.17–55.5)               | 45.0 (29.5–57.0)               | 0.313                                           | 0.625       |
| FoxP3/epidermis                           | 3.0 (0.5–12.0)                              | 2.0 (1.0–5.5)                               | 0.0 (0.0–4.0)                  | 1.0 (0.5–4.0)                  | 0.125                                           | 0.625       |
| FoxP3/dermis                              | 47.5 (22.0–61.5)                            | 22.0 (5.5–49.75)                            | 22.5 (12.75–38.0)              | 22.5 (12.7–36.75)              | 0.313                                           | 0.625       |
| <i>Immunopositive keratinocyte nuclei</i> |                                             |                                             |                                |                                |                                                 |             |
| GR $\alpha$ /epidermis                    | 80.0 (70.0–92.5) <sup>e</sup>               | 80.0 (70.0–87.5)                            | 60.0 (50.0–77.5)               | 70.0 (57.5–77.5)               | 0.250                                           | 0.063       |
| GR $\beta$ /epidermis                     | 50.0 (42.5–62.5)                            | 60.0 (47.5–62.5)                            | 45.0 (30.0–50.0)               | 55.0 (40.0–60.0)               | 0.250                                           | 0.500       |

<sup>a</sup>Combination: combination of calcipotriol/betamethasone. <sup>b</sup>Monotherapy: betamethasone. <sup>c</sup>*p*-values are calculated by Wilcoxon signed-rank test. <sup>d</sup>The number of immunopositive cells /20 $\times$  field image; median (interquartile range). <sup>e</sup>Percentage of immunopositive keratinocyte nuclei; median (interquartile range).