Figure S1, related to Figure 1. T<sub>H</sub> cells convert monocytes into DCs in the presence of IL-2, IL-15 or tetanus toxoid antigen. CD14<sup>+</sup> monocytes were cultured as indicated. After 6 days, cells were analyzed for surface expression of DC-associated molecules after excluding T<sub>H</sub> cells. Isotype controls are shown as dashed lines. (A) CD14<sup>+</sup> monocytes were cultured alone (blue), with autologous T<sub>H</sub> cells (orange), IL-2 (black), autologous T<sub>H</sub> cells and IL-2 (red), or GM-CSF and IL-4 (purple). Data are representative of 4 independent experiments and >10 donors. (B) CD14<sup>+</sup> monocytes were cultured alone (Mono), with autologous T<sub>H</sub> cells (Mono + T<sub>H</sub>), IL-2 (Mono + IL-2), or autologous T<sub>H</sub> cells and IL-2 (DC<sub>Th</sub>). Median fluorescence intensities (MFIs) of DC-associated molecules are shown. Data are from 4 independent experiments and >10 donors (mean and SEM); *p<0.05, **p<0.01, ***p<0.001 as compared to all other subsets. (C) CD14<sup>+</sup> monocytes were cultured with IL-2 alone (black, open), tetanus toxoid alone (Tet, black, filled), autologous T<sub>H</sub> cells and IL-2 (DC<sub>Th</sub>, red, open), or autologous T<sub>H</sub> cells and tetanus (Tet, red, filled). Data are representative of 3 independent experiments with 5 donors. (D) FACS sorted T<sub>H</sub> cells were cultured in medium alone (Unstim) or IL-2. After 5 days, supernatants were collected for measurement of GM-CSF by ELISA. Data are from 2 independent experiments and 2 donors (mean and SEM). (E) CD14<sup>+</sup> monocytes were cultured with IL-2 alone (black, open), tetanus toxoid alone (Tet, black, filled), autologous T<sub>H</sub> cells and IL-2 (DC<sub>Th</sub>, red, open), or autologous T<sub>H</sub> cells and tetanus (Tet, red, filled). Data are from 2 independent experiments with 6 donors (mean and SEM); *p<0.05 as compared to Mono Alone in its respective condition.

Figure S2, related to Figure 3. T<sub>H</sub> subsets interact with monocytes and DCs in inflamed tissues, and T<sub>H</sub> subsets isolated from peripheral blood elicit DC formation. (A, B) Representative skin lesions from psoriasis and acute atopic dermatitis patients were stained as indicated. Images are shown at 200X magnification. Scale bar equals 40 µm. (A) Skin lesions were stained with CD4 or T-bet, ROR<sub>γ</sub>, GATA-3 to identify T<sub>H1</sub>, T<sub>H17</sub> or T<sub>H2</sub> cells, respectively. (B) Skin lesions were stained with DC-LAMP (1:100 dilution, Dendritics, clone 104G4) to identify DCs and T-bet (left column), ROR<sub>γ</sub> (middle column) or GATA-3 (right column) to identify T<sub>H1</sub>, T<sub>H17</sub> and T<sub>H2</sub> cells, respectively. Red arrows indicate potential DC/T<sub>H</sub> interactions. (C) Memory CD4<sup>+</sup>CD45RO<sup>+</sup>CD45RA<sup>-</sup> T cells were isolated by magnetic cell sorting from PBMCs derived from healthy donors and subsequently labeled with surface markers specific for T<sub>H</sub> subsets. After gating out CD4<sup>+</sup>CD45RA<sup>+</sup>CD25<sup>+</sup>CD127<sup>-</sup> cells to deplete Tregs, T<sub>H2</sub> cells were defined as PI<sup>-</sup>CD4<sup>+</sup>CD45RA<sup>-</sup>CD25<sup>-</sup>CRTH2<sup>-</sup>CD161<sup>-</sup>CCR4<sup>-</sup>CCR6<sup>-</sup>. T<sub>H1</sub> cells were defined as PI<sup>-</sup>CD4<sup>+</sup>CD45RA<sup>-</sup>CD25<sup>-</sup>CRTH2<sup>-</sup>CD161<sup>-</sup>CCR4<sup>-</sup>CCR6<sup>-</sup>, while T<sub>H17</sub> cells were defined as PI<sup>-</sup>CD4<sup>+</sup>CD45RA<sup>-</sup>CD25<sup>-</sup>CRTH2<sup>-</sup>CD161<sup>-</sup>CCR4<sup>-</sup>CCR6<sup>-</sup>. Shaded boxes indicate sorted populations. (D) Bulk memory CD4<sup>+</sup>CD45RA<sup>-</sup> T cells (Bulk) and T<sub>H</sub> subsets were stimulated with 25 ng/mL PMA and 750 ng/mL ionomycin for 12 hours, and supernatants were collected for measurement of IL-4, IL-5, IL-13, IL-17A, IFN-γ, TNF-α and GM-CSF by ELISA. Data are from 4 independent experiments and 4 donors (mean and SEM), *p<0.05 as compared to the other T<sub>H</sub> subsets for the same cytokine. (E) CD14<sup>+</sup> monocytes were cultured with IL-2 alone (Mono); with IL-2 and allogeneic (filled columns) or autologous (open columns) T<sub>H1</sub> cells (DC<sub>Th1</sub>), T<sub>H2</sub> cells (DC<sub>Th2</sub>) or T<sub>H17</sub> cells (DC<sub>Th17</sub>). After 6 days, each culture was analyzed for surface expression of DC-associated molecules after excluding T<sub>H</sub> cells. Isotype controls are shown as dashed lines. Data are from 3 donors (mean and SEM).

Figure S3, related to Figure 4. Cytokine secretion profile of different DC<sub>Th</sub> subsets. Monocytes (Mono) or each DC<sub>Th</sub> co-culture was stimulated with 2 µg/mL LPS at day 6. (A, B) Brefeldin A (BFA, eBioscience) was added after (A) 12 or (B) 4 hours to Mono and allogeneic
DC\textsubscript{Th} co-cultures. Cells were analyzed 6 hours later for intracellular expression of IL-1β and IL-12p40 (eBioscience) or TNF-α (BD Pharmingen) after excluding T\textsubscript{H} cells. Data are representative of 3 independent experiments and 7 donors. (C) Supernatants from autologous co-cultures were collected for measurement of IL-1β, IL-6, IL-10, IL-12p70, IL-23p19 and TNF-α by ELISA 36 hours after LPS stimulation. Data are from 3 independent experiments and 5 donors (mean and SEM), *p<0.05 as compared to all other subsets; †p<0.05 as compared to Mono and DC\textsubscript{Th2}. (D) Prior to LPS stimulation, T cells were magnetically depleted from DC\textsubscript{Th} co-cultures. Mono and DC\textsubscript{Th} co-cultures were subsequently stimulated with 2 µg/mL LPS. After 36 hours, cell-free supernatants were collected for measurement of IL-1β, IL-6, IL-10 and TNF-α by ELISA. Data are from 3 independent experiments and 5 donors (mean and SEM), *p<0.05 as compared to Mono and DC\textsubscript{Th2}, †p<as compared to DC\textsubscript{Th1} and DC\textsubscript{Th17}.

Figure S4. DC\textsubscript{Th} subsets promote distinct T\textsubscript{H}-cell responses. T-cell depleted DC\textsubscript{Th} subsets from autologous co-cultures were stimulated with LPS for 36 hours. (A) DC\textsubscript{Th} subsets were subsequently cultured with allogeneic naïve CD4\textsuperscript{+} T cells for six days and stimulated with 25 ng/mL PMA and 750 ng/mL ionomycin for 4 hours prior to the addition of BFA and subsequent intracellular cytokine analysis of IFN-γ, IL-13 and IL-17. Percent cytokine positive cells (% Cytokine Pos.) are shown after gating on CFSE\textsuperscript{+} T cells. Data are from 4 independent experiments and 4 donors (mean and SEM). (B) DC\textsubscript{Th} subsets were subsequently cultured alone (open columns) or with allogeneic naïve CD4\textsuperscript{+} T cells (filled columns) for six days. Cell free supernatants were collected for measurement of IL-5, IL-13, IL-17A and IFN-γ by ELISA. Instances where the level of cytokine present was below the level of detection are indicated as not detected (N.D). Data are from 2 independent experiments and 2 donors (mean and SEM).
Figure S1, related to Figure 1. T_\text{H} cells convert monocytes into DCs in the presence of IL-2, IL-15 or tetanus toxoid antigen.
Figure S2, related to Figure 3. \( T_H \) subsets interact with monocytes and DCs in inflamed tissues, \( T_H \) subset isolation from peripheral blood and allogeneic \( T_H \) cells elicit DC formation.
Figure S3, related to Figure 4. Cytokine secretion profile of different DC_{Th} cultures
Figure S4. DC<sub>Th</sub> subsets promote distinct T<sub>H</sub>-cell responses