SUPPLEMENTAL MATERIAL

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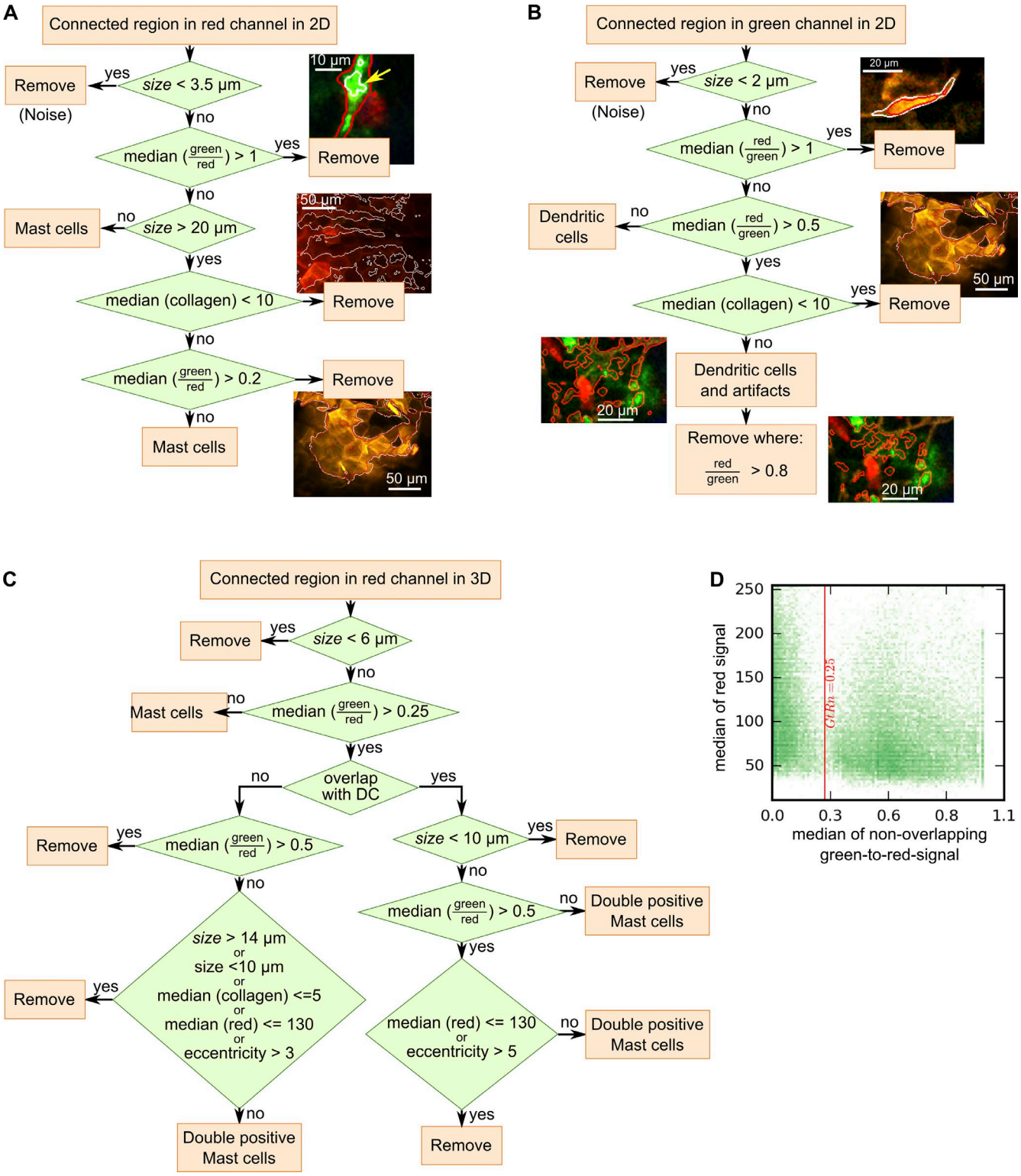


Figure S1. **Decision workflow for artifacts removal.** (A) Red channel in 2D. (B) Green channel in 2D. (C) Red channel in 3D. (D) Distribution of red signal versus green-to-red signal motivating the choice of the threshold value 0.25 for the green-to-red signal to distinguish between ordinary MC (<0.25) and GFP$^+$RFP$^+$MCs or artifacts (>0.25). Selection of other thresholds was done analogously based on distributions of corresponding parameters in all objects. Related to Fig. 1.

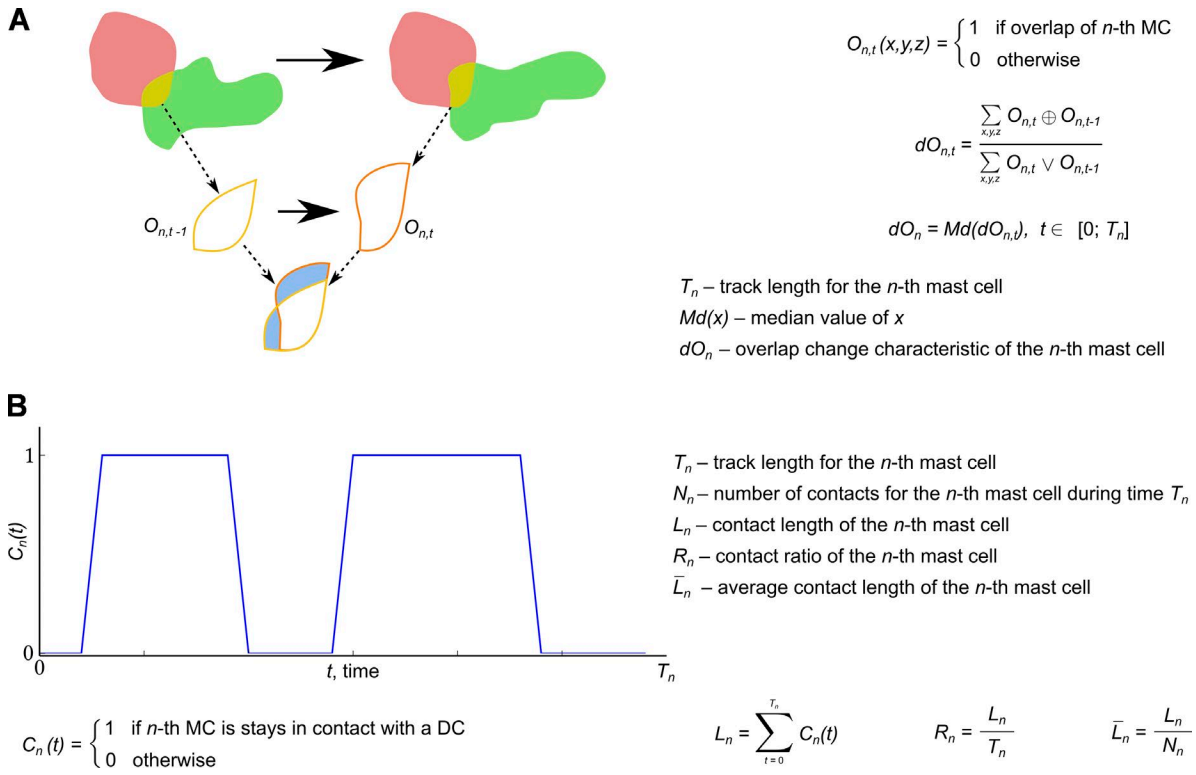


Figure S2. **Dynamic analysis of DC–to–MC interactions over time.** (A) Computing of overlap change between MCs and DCs in time-lapse series. (B) Computing characteristics of contacts between MCs and DCs. Related to Fig. 5.

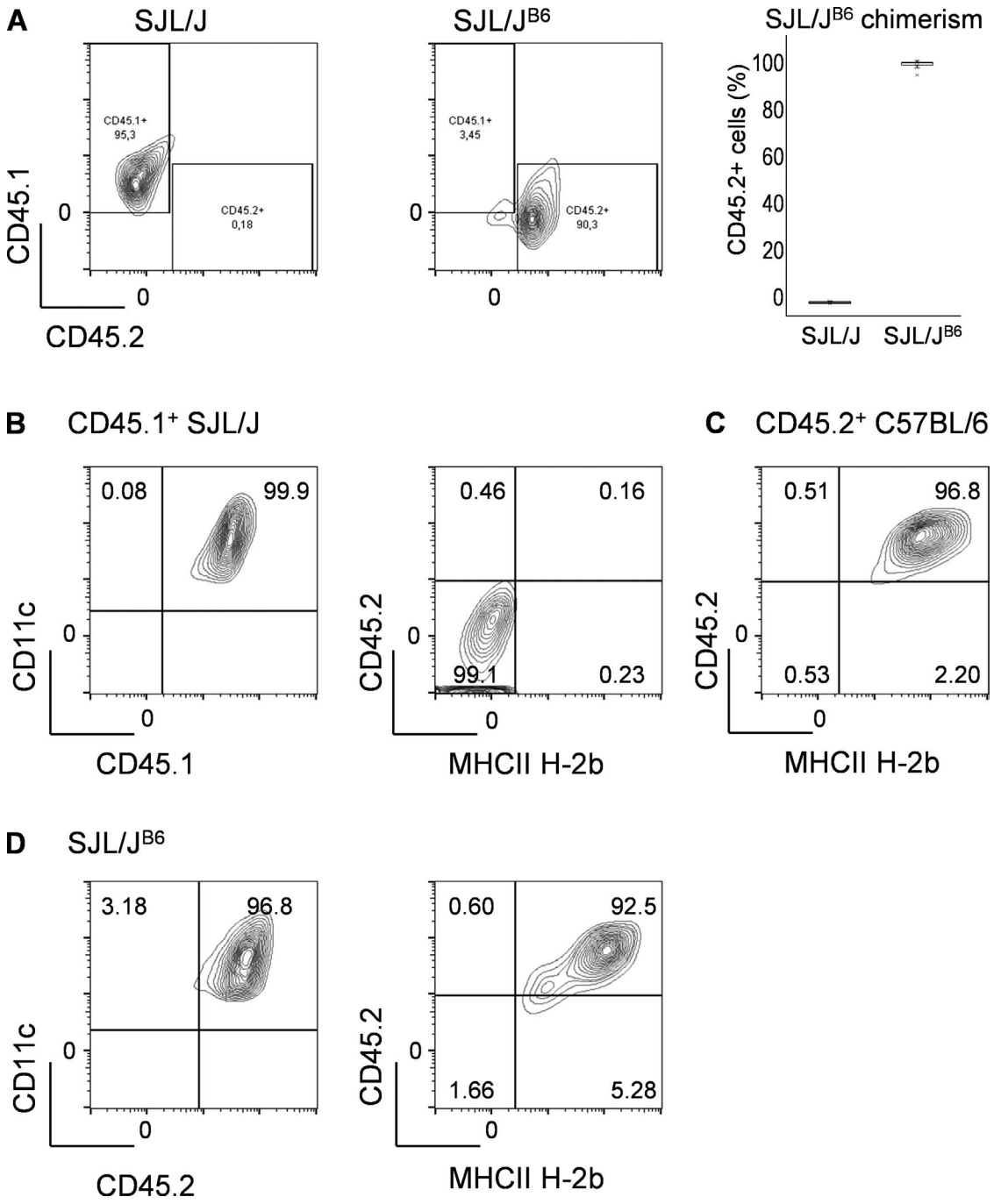


Figure S3. **Analysis of CD45.2 and H-2^b MHCII expression by SJL/J^{B6} BM chimera DCs.** (A) The chimerism of SJL/JB6 mice was determined and quantified according to the expression of CD45.1 and CD45.2 on blood leukocytes 2 wk after BM transplantation. (B–D) CD45.1, CD45.2, and MHCII haplotype H2^b expression in ear skin CD11c$^+$DCs of untreated SJL/J mice (B) and C57BL/6 mice (C) and in ear skin of SJL/J^{B6} BM chimera mice (D) 24 h after DNFB administration was analyzed using flow cytometry. Related to Fig. 8.

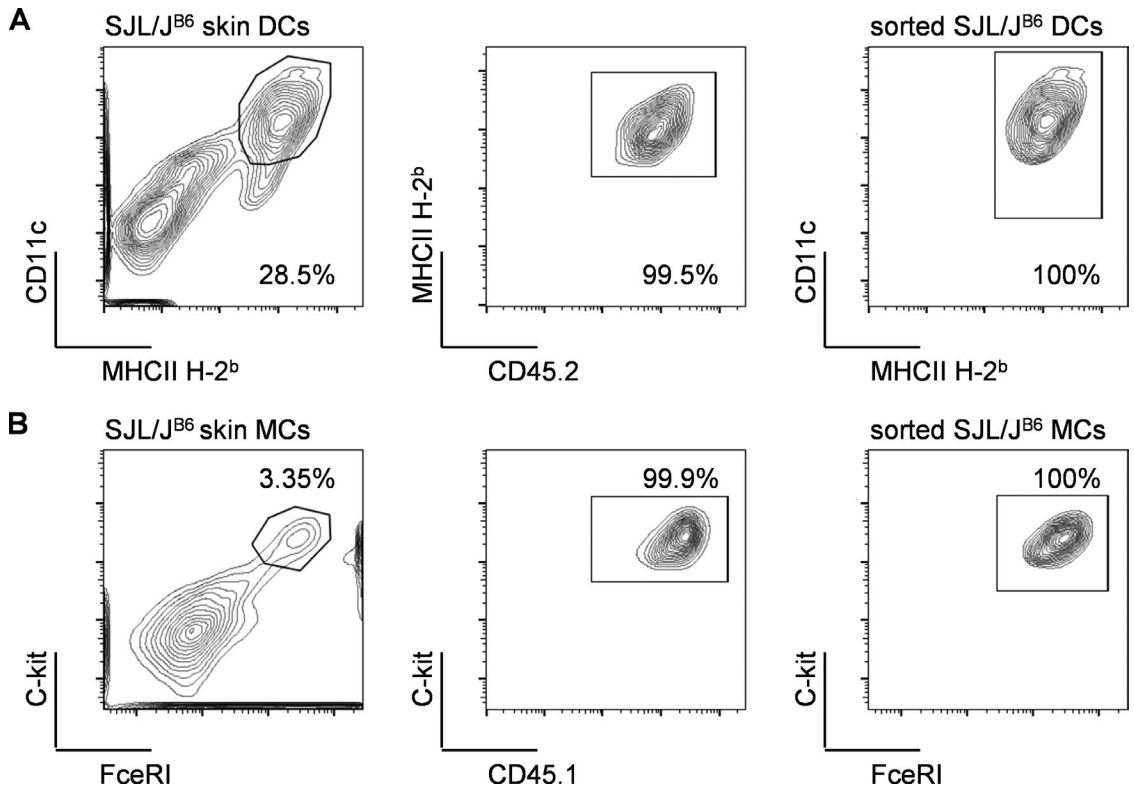
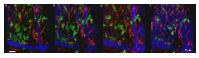
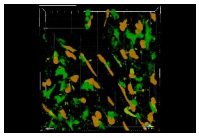


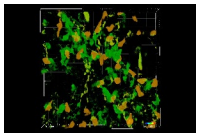
Figure S4. **Sorting of skin DCs and MCs from SJL/J^{B6} BM mice.** (A) Donor BM replaced ear skin DCs (CD11c$^+$, MHCII H2^{b+}, CD45.2$^+$) were sorted from SJL/J^{B6} BM chimera 24 h after DNFB. The sorting control proved 100% expression of CD11c and H2^b, thereby excluding contamination with other cell subsets. Control DCs were sorted from SJL/J (H2^s) mice 24 h after DNFB as cell subset expressing CD11c and CD45.1, but no expression of MHCII H2^b and CD45.2 (not depicted). The sorting control proved 100% expression of CD11c and CD45.1. (B) Skin MCs from SJL/J mice and recipient skin MCs from SJL/J^{B6} BM chimera mice were sorted 24 h after DNFB as cell subset expressing c-kit, FcεRI, and CD45.1. The sorting control proved 100% expression of c-kit, FcεRI, and CD45.1, thereby excluding contamination with other cell subsets. Related to Fig. 9.



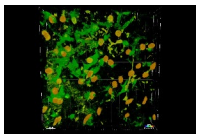
Video 1. **DC migrational arrest upon DNFB administration.** (A–D) Longitudinal and side-matched time-lapse series of DC^{GFP}/MC^{RFP} mouse ear skin after DNFB (A), and 1 h (B), 6 h (C), and 7.5 h (D) after DNFB administration. MCs are represented in red and DCs in green. Blood vessels are depicted by a vascular tracer (blue). Skin inflammation is associated with a pronounced increase in plasma (and vascular dye) leakage (B–D). DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as maximum intensity projection; time of observation, 29 min, each. Bar, 30 µm. Refers to Fig. 2.



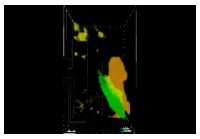
Video 2. **Time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin before DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 30 min. Bar, 20 µm. Refers to Fig. 3 B.



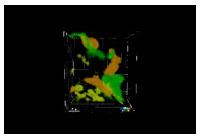
Video 3. **Time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin 12 h after DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 60 min. Bar, 20 µm. Refers to Fig. 3 B.



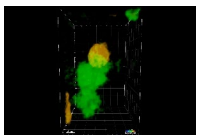
Video 4. **Time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin 24 h after DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 60 min. Bar, 20 µm. Refers to Fig. 3 B.



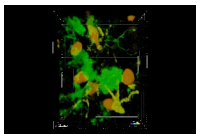
Video 5. **DC/MC single-pair time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin before DNFB treatment.** MC is represented in orange, DC in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 30 min. Bar, 5 µm. Refers to Fig. 4 A.



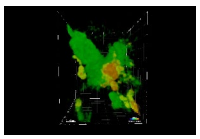
Video 6. **Close-up time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin before DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 30 min. Bar, 10 µm. Refers to Fig. 4 A.



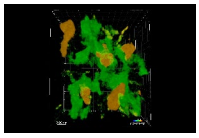
Video 7. **DC/MC single-pair time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin 8 h after DNFB treatment.** MC is represented in orange, DC in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D rendered z-stack; time of observation, 30 min. Bar, 5 µm. Refers to Fig. 4 B.



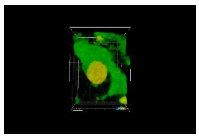
Video 8. **Close-up time-lapse series of DC^{GFP}/MC^{RFP} mouse ear skin 8 h after DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 34 min. Bar, 10 µm. Refers to Fig. 4 B.



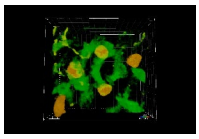
Video 9. **DC/MC single-pair time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin 14 h after DNFB treatment.** MC is represented in orange, DC in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 60 min. Bar, 5 µm. Refers to Fig. 4 C.



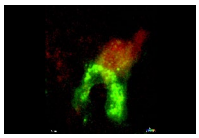
Video 10. **Close-up time-lapse series of DC^{GFP}/MC^{RFP} mouse ear skin 14 h after DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 60 min. Bar, 8 µm. Refers to Fig. 4 C.



Video 11. **DC/MC single-pair time-lapse series in DC^{GFP}/MC^{RFP} mouse ear skin 24 h after DNFB treatment.** MC is represented in orange, DC in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 60 min. Bar, 7 µm. Refers to Fig. 4 D.



Video 12. **Close-up time-lapse series of DC^{GFP}/MC^{RFP} mouse ear skin 24 h after DNFB treatment.** MCs are represented in orange, DCs in green; areas of DC-to-MC colocalization appear as yellow contact zones; DC/MC-independent yellow structures correspond to autofluorescence (including hairs, corneocytes, melanocytes, and structures in blood flow). Time-lapse series is depicted as a 3D-rendered z-stack; time of observation, 30 min. Bar, 10 µm. Refers to Fig. 4 D.



Video 13. **Single z-plane time-lapse series of a GFP^+RFP^+MC in contact with DCs in DC^{GFP}/MC^{RFP} mouse ear skin 24 h after DNFB treatment, showing the intercellular transfer of GFP^+ vesicles.** MC is represented in red, DCs in green. Time of observation, 22 min. Bar, 5 µm. Refers to Fig. 5 D.