doi:10.1038/nature13199

Role of Sympathetic Nerves in Psoriasiform Skin Inflammation

Since there was no detectable effect of sympathectomy on the local production of IMQ-induced inflammatory cytokines, it is highly probable that the effect of 6-OHDA was not at the level of the ear skin, but due to one or more systemic effects. For example, it is well established that the sympathetic nervous system regulates the anti-inflammatory splenic reflex arc[1], as well as, heart rate and systemic blood pressure[2,3]. Upon systemic sympathectomy, circulating myeloid cells increase[4,5] (providing an enhanced supply of inflammatory cells to the inflamed ear), while blood pressure drops leading to a diminished hydrostatic pressure differential between the intra- and extravascular compartments, which may translate to decreased edema formation in the ear skin.

Spatial Localization of Dendritic Cells within Skin

DCs in the ear reside preferentially close to nerves, when compared with other anatomical structures, such as lymph vessels and blood vessels. However, it is still possible that the observed difference is the result of distinct densities of these anatomical structures within the image volume. To address this possibility, we analysed the image volumes for the surface area and volume of nerves and blood vessels or nerves and lymph vessels co-stained in 3D ear skin confocal micrographs. While we didn't observe major differences in the surface area occupied by nerve and lymph vessels in the imaging space (Image volume 1: nerves occupy 525,497.60 μm^2; lymph vessels occupy 516,272.08 μm^2), blood vessels have a larger surface area than nerves (Image volume 2: nerves occupy 571,178.34 μm^2; blood vessels occupy 707,080.45 μm^2). When the volume occupied by the different structures is taken into account, lymph and blood vessels appear to be occupying a larger volume in the 3D image (Image volume 1: nerves occupy 2,024,046.48 μm^3; lymph vessels occupy 2,947,433.00 μm^3 and image volume 2: nerves occupy 2,022,104.42 μm^3; blood vessels occupy 3,480,004.76 μm^3). Altogether, these data suggest that a difference in density of the different anatomical structures cannot account for the difference in their distance to DCs we observed. The difference in surface area and volume of the structures highlight that one might expect a bias towards lymph and blood vessels if the DCs were randomly distributed in the ear tissue.

CGRP in the IMQ Model

What are the cellular and molecular tools of communication employed by nociceptors and dDCs? One candidate signal is CGRP, a neuropeptide released by sensory fibers, which has been implicated in promoting IL-23 production in a chronic psoriasis model[6]. However, a recent study on the impact of bacterial toxins and N-formylated peptides on $NaV1.8^+$ nociceptors ascribed an anti-inflammatory role to nociceptor-derived CGRP[7], suggesting that the pathophysiological role of this neuropeptide is complex and may be context dependent. In our hands, systemic treatment of mice with CGRP antagonists did not affect IMQ induced IL-23 production, so CGRP does not appear to be required for nociceptor function in the IMQ model (data not shown).

1 Rosas-Ballina, M. *et al.* Acetylcholine-Synthesizing T Cells Relay Neural Signals in a Vagus Nerve Circuit. *Science* **334**, 98-101, doi:10.1126/science.1209985 (2011).

2 Joyner, M. J., Charkoudian, N. & Wallin, B. G. A sympathetic view of the sympathetic nervous system and human blood pressure regulation. *Exp Physiol* **93**, 715-724, doi:10.1113/expphysiol.2007.039545 (2008).

3 Charkoudian, N. & Rabbitts, J. A. Sympathetic Neural Mechanisms in Human Cardiovascular Health and Disease. *Mayo Clin Proc.* **84**, 822-830 (2009).

4 Yamazaki, S. *et al.* Nonmyelinating Schwann cells maintain hematopoietic stem cell hibernation in the bone marrow niche. *Cell* **147**, 1146-1158, doi:10.1016/j.cell.2011.09.053 (2011).

5 Scheiermann, C. *et al.* Adrenergic nerves govern circadian leukocyte recruitment to tissues. *Immunity* **37**, 290-301, doi:10.1016/j.immuni.2012.05.021 (2012).

6 Ostrowski, S. M., Belkadi, A., Loyd, C. M., Diaconu, D. & Ward, N. L. Cutaneous denervation of psoriasiform mouse skin improves acanthosis and inflammation in a sensory neuropeptide-dependent manner. *J Invest Dermatol* **131**, 1530-1538, doi:10.1038/jid.2011.60 jid201160 [pii] (2011).

7 Chiu, I. M. *et al.* Bacteria activate sensory neurons that modulate pain and inflammation. *Nature*, doi:10.1038/nature12479 (2013).