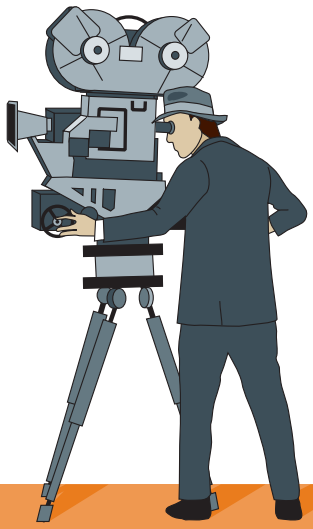

NATURAL KILLER T CELLS

Picturing activation in action

Two groups of investigators, reporting in *Nature Immunology*, have visualized invariant natural killer T (iNKT) cells being activated *in vivo* and provide new insights into how and where this occurs. Both studies reveal a crucial role for specialized tissue-resident macrophage populations — subcapsular sinus CD169$^+$ macrophages in the lymph nodes and Kupffer cells in the liver sinusoids — in early iNKT cell activation.

Using multiphoton microscopy, Barral *et al*. tracked the behaviour of fluorescently labelled iNKT cells in lymph nodes following adoptive transfer into wild-type mice. In resting lymph nodes, iNKT cells were mainly found in the paracortex alongside CD4$^+$ T cells and sometimes close to the subcapsular sinus (SCS), but were mainly excluded from the B cell follicles. The average speed and random pattern of their movements were similar to co-transferred fluorescently labelled CD4$^+$ T cells. The transferred iNKT cells were activated *in vivo* by injection of silica particles coated with the iNKT cell glycolipid antigen α-galactosylceramide (α-GalCer). After injection of these stimulatory particles, but not particles coated with non-stimulatory lipids, iNKT cell movements in the draining lymph nodes slowed, were more confined and involved more long-lasting stops. This did not occur when the cells were transferred into CD1d-deficient mice, indicating that this change in behaviour depended on the recognition of α-GalCer presented by CD1d molecules.

Further imaging revealed that, within minutes of injection, the lipid-coated particles could be detected in the SCS and medullary regions of the draining lymph nodes, in particular, co-localizing with CD169$^+$ macrophages. Within 6 hours, transferred iNKT cells were retained in the antigen-rich areas and established direct contacts with CD169$^+$ macrophages. Experiments involving depletion of macrophages in the lymph nodes or *in vitro* analysis of purified populations confirmed that SCS CD169$^+$ macrophages can present antigenic lipids — even those that require internalization and processing — to iNKT cells, inducing their activation, cytokine production and proliferation.

Lee *et al*. visualized the behaviour of iNKT cells in the liver following infection of mice with *Borrelia burgdorferi*, a spirochete responsible for human Lyme disease. In uninfected livers, iNKT cells were found to reside mainly in the sinusoids and to move in a random pattern, whereas Kupffer cells were largely immobile with extended processes reaching into several sinusoids. Within a few hours of infection, most *B. burgdorferi* spirochetes became trapped by Kupffer cells and iNKT cells became more stationary, forming clusters with Kupffer cells containing *B. burgdorferi*. The reduced motility and clustering of iNKT cells was shown to depend on CXC-chemokine receptor 3 and CD1d and resulted in the production of interferon-γ by the iNKT cells. Studies involving the depletion of Kupffer cells confirmed that these cells are responsible for rapid CD1d-mediated activation of iNKT cells in the liver in response to *B. burgdorferi*. Moreover, this pathway was shown to be crucial to limit the spread of *B. burgdorferi* from the blood to the liver parenchyma and joints.

Together, these studies establish a key role for tissue macrophages in the early activation of iNKT cells, which is likely to facilitate the induction of later immune responses.

Lucy Bird

ORIGINAL RESEARCH PAPERS Barral, P. *et al.* CD169$^+$ macrophages present lipid antigens to mediate early activation of iNKT cells in lymph nodes. *Nature Immunol.* 14 Mar 2010 (doi:10.1038/ni.1853) | Lee, W.-Y. *et al.* An intravascular immune response to *Borrelia burgdorferi* involves Kupffer cells and iNKT cells. *Nature Immunol.* 14 Mar 2010 (doi:10.1038/ni.1855)