

EDITORIAL REVIEW

Neutrophils and adjuvant arthritis

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The role of neutrophils in immunologically mediated inflammatory reactions has been controversial, especially for reactions that involve a morphological predominance of lymphocytes. Another matter of controversy has been the extent to which infiltration of monocytes and lymphocytes into tissues requires a prior presence of neutrophils, as opposed to the complete independence of a mononuclear cell/lymphocyte influx on the prior accumulation of neutrophils. The resolution of these questions as well as the extent to which cytokine/chemokine-dependent inflammatory reactions exclusively reflect monocyte, macrophage and lymphocyte involvement has been made more difficult by the realization that neutrophils can also express mRNA and proteins for a wide array of cytokines/chemokines [1]. Early approaches to address the role of neutrophils in inflammatory models involved the use of cytotoxic drugs, such as nitrogen mustard or cyclophosphamide, which tended to show, at least within the first 72 h, selective depletion of neutrophils. However, it is well known that such drug-related effects are not specific for this cell lineage, making observed results difficult to interpret. A key to resolving some of these questions has been access to blocking reagents that are highly specific for epitopes contained on neutrophils and devoid of reactivity with epitopes on monocytes and lymphocytes.

Requirements for neutrophils have been established in a large number of immunologically mediated types of tissue injury in which the key event is triggering of an inflammatory response [2]. Neutrophil involvement is especially prominent in lesions associated with deposition of immune complexes, and often in other conditions featuring the presence of complement activation products. In rats antigen-induced arthritis and collagen type II-induced arthritis have been found to be neutrophil- and complement-dependent [3]. Adjuvant arthritis is a type of progressive inflammatory polyarthritis that displays many of the features of human rheumatoid arthritis, with a prominence of synovial thickening and hypercellularity and numerous inflammatory cells, including neutrophils as well as mononuclear and lymphoid cells. Progressive erosion of articular cartilage is a prominent feature of this disease. Not surprisingly, immunosuppressive approaches such as blockade of CD4⁺ lymphocytes effectively reduce the intensity of damage and the progression of adjuvant arthritis. The current report of Santos *et al.* [4] convincingly demonstrates a requirement not only for CD4⁺ lymphocytes but also for neutrophils, the latter being determined by the protective

effects of neutrophil depletion, even though another report suggested that methotrexate-induced neutropenia was not effective in reducing the arthritic symptoms in adjuvant arthritis [5]. However, these discrepant findings may be a reflection of insufficiently sensitive endpoints of measurement or inadequate levels of neutrophil depletion. In the studies of Santos *et al.* [4] it is clear that interventions with antibody either to neutrophils or to CD4⁺ lymphocytes, while significantly attenuating evidence of arthritis, provided incomplete protection, especially when paw volume was used as the endpoint (e.g. 29% and 28% reductions in paw volume as a result of neutrophil or CD4⁺ lymphocyte depletion, respectively). Accordingly, either insufficient neutrophil depletion occurred, or neutrophils contribute only partly to outcomes such as increases in paw volume and other changes related to development of arthritis. Although one might be concerned that the antibody to neutrophils could be reactive with a lymphocyte-associated epitope, thus reducing the critical immune response required to activate the chain of events leading to development of arthritis, this possibility seems unlikely to be the case, for two reasons: (i) the anti-neutrophil antibody treatments were not administered until days 7 and 9, long after establishment of an immune response to antigen(s) in the *Mycobacteria* suspension; and (ii) the DTH skin reaction to tuberculin purified protein derivative (PPD) remained intact and unaltered in the face of the neutrophil depletion procedures. For these reasons, it seems unlikely that the protective effects of treatment with anti-neutrophil antibody in the adjuvant arthritis model could be the result of induced defects in immune effector functions by the anti-neutrophil antibody. The sequence of events would seem to be that CD4⁺ cells are necessary for establishment of the immune response, the outcomes of which are products which lead to the recruitment of neutrophils, perhaps with major involvement of cytokines (tumour necrosis factor- α (TNF- α), IL-1) and the IL-8 family of chemokines, especially the α chemokines. The combination of products (oxidants, proteinases, cytokines) from stimulated neutrophils, synovial macrophages and lymphocytes would then seem to set the stage for acute and progressive polyarthritis.

What is especially intriguing in the adjuvant arthritis model is why after injection of *M. tuberculosis* into the base of the tail of rats the inflammatory response, which because progressive, is primarily restricted to joints and not to other locales. There appears to be evidence that rats develop an immune response to products of *M. tuberculosis*, including heat shock proteins, and perhaps to other *Mycobacterial* products [6], the result of which is a cellular and humoral response with cross-reactivity to antigens in articular cartilage proteoglycans [7–9]. Interestingly, in patients with rheumatoid

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arthritis there appears to be autoreactivity to these heat shock proteins [8,10]. It also seems clear that in the 'passive' immune form of adjuvant arthritis reactive lymphocytes home to joints, where they induce development of arthritis [11]. In aggregate, these data seem to suggest that adjuvant arthritis has intriguing ties to immune responses found in patients with rheumatoid arthritis. How to tie together the mechanisms of neutrophil recruitment with the T cell-mediated immune responses in adjuvant arthritis and how more precisely to define the role of neutrophils in this form of inflammatory arthritis are the next challenges.

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