CHAPTER 18
RANKL INHIBITION: FROM MICE TO MEN (AND WOMEN)

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1. ABSTRACT

RANKL, the primary mediator of osteoclast formation, function and survival, is implicated in bone loss across a broad range of conditions. RANK and RANKL are expressed by cells involved in bone remodeling, by cells of the immune system, and by cells in other tissues. Preclinical and clinical data support the following conclusions: 1) The immune and skeletal phenotypes associated with RANKL inhibition differ in important ways from those associated with the complete absence or ablation of RANK or RANKL. 2) Immune challenge performed in animals in the presence of RANKL inhibition demonstrates normal immune function, consistent with the interpretation that RANKL inhibition does not impair the ability of animals to mount an effective immune response. 3) In animal models of inflammatory disease, inhibition of RANKL prevents bone loss but does not show a detectable effect on immune mediators or inflammation. 4) A phase 2 study in postmenopausal women with low BMD using the RANKL inhibitor denosumab showed an increase in BMD with an incidence of adverse events that was similar to placebo and to open-label alendronate. In addition, in a subset of patients tested for immunological markers, there were no clinically meaningful differences in T, B, or NK cell numbers or in immunoglobulin levels across dose or treatment groups.

2. INTRODUCTION

The signaling pathway that is activated upon receptor activator of NF-kappa B ligand (RANKL) binding to its receptor RANK is central to bone remodeling (Boyle, Simonet, and Lacey 2003). RANKL is produced by osteoblasts, stromal cells, and other cells and is an essential...
mediator of osteoclast formation, function, and survival (Fuller, Wong, Fox, et al. 1998; Lacey, Timms, Tan, et al. 1998; Yasuda, Shima, Nakagawa, et al. 1998; Lacey, Tan, Lu, et al. 2000; Boyle, Simonet, and Lacey 2003). RANK is expressed on mature osteoclasts and their precursors. RANKL binding to RANK results in increased formation, function, and survival of osteoclasts, leading to increased bone-resorbing activity. Osteoprotegerin (OPG) is an endogenous inhibitor that binds to and prevents RANKL from binding RANK, thereby reducing osteoclast activity.

The essential requirement of RANKL and RANK in bone resorption was demonstrated using knockout animals. Mice lacking RANK or RANKL are unable to produce mature osteoclasts and are born with osteopetrosis. Transgenic mice overexpressing the RANKL inhibitor, OPG, have osteosclerosis, but in contrast to the knockout, have normal tooth eruption and normal bone shape (Simonet, Lacey, Dunstan, et al. 1997; Dougall, Glaccum, Charrier, et al. 1999; Kong, Yoshida, Sarosi, et al. 1999). Mice lacking OPG are severely osteopenic due to unopposed RANKL action driving bone resorption (Bucay, Sarosi, Dunstan, et al. 1998; Mizuno, Amizuka, Irie, et al. 1998).

There are many tissues in which RANK or RANKL mRNA is expressed. In the immune system, RANKL is expressed by activated T and B cells, whereas expression of RANK is mostly confined to mature dendritic cells—cells which share a common lineage with osteoclasts and macrophages (Anderson, Maraskovsky, Billingsley, et al. 1997; Wong, Josien, Lee, et al. 1997; Choi, Woo, Ko, et al. 2001). Although these molecules are expressed, a functional and non-redundant role in the adult immune system has yet to be identified.

3. INHIBITION VS ABSENCE OF RANKL

Inhibition of the RANKL pathway (as with OPG administration or overexpression) differs from complete absence or ablation of RANKL or RANK molecules, as shown by the phenotypes of transgenic (RANKL inhibition) and knock-out (ablation of RANK or RANKL) animals. Ablation of the RANKL or RANK genes in mice results in absence of lymph nodes (Dougall, Glaccum, Charrier, et al. 1999; Kong, Yoshida, Sarosi, et al. 1999). The interaction of RANK and RANKL appears to be an essential mediator of the differentiation of lymph node ‘inducer’ cells during embryogenesis of the lymph node anlagen in mouse at approximately fetal day 14 (Mebius 2003). Altered development and differentiation of lymphocytes, absence of tooth eruption, osteopetrosis, and absence of lactation are also seen in these animals (Kong, Yoshida, Sarosi, et al. 1999). Animals with the absence of RANK or RANKL have normal Peyer's patches, normal splenic architecture, and normal dendritic cell numbers and function, suggesting that RANK/RANKL is not essential for formation of these organs or for dendritic cell development or function.

In contrast, inhibition of this pathway by transgenic overexpression of OPG in mice does not affect lymph node formation or lymphocyte function (Simonet, Lacey, Dunstan, et al. 1997). Thus, the phenotype of RANKL inhibition is very different from that of RANKL absence in terms of embryogenesis of the immune system. In addition, as discussed below, numerous studies with exogenous RANKL inhibitors support the idea that RANKL inhibition in adult animals does not affect the immune system.

4. RANKL INHIBITION AND IMMUNE CHALLENGE

Immune challenges performed in animals in the presence of RANKL inhibition demonstrate normal immune function, consistent with the interpretation that the RANKL/RANK pathway is not essential for an animal to mount a normal immune response. In studies of mice