

# Paget's disease of bone: an osteoimmunological disorder?

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**Abstract:** Osteoimmunology represents a large area of research resulting from the cross talk between bone and immune systems. Many cytokines and signaling cascades are involved in the field of osteoimmunology, originating from various cell types. The RANK/receptor activator of nuclear factor Kappa-B ligand (RANKL)/osteoprotegerin (OPG) signaling has a pivotal role in osteoimmunology, in addition to proinflammatory cytokines such as tumor necrosis factor- $\alpha$ , interleukin (IL)-1, IL-6, and IL-17. Clinically, osteoimmunological disorders, such as rheumatoid arthritis, osteoporosis, and periodontitis, should be classified according to their pattern of osteoimmunological serum biomarkers. Paget's disease of bone is a common metabolic bone disorder, resulting from an excessively increased bone resorption coupled with aberrant bone formation. With the exception of the cellular responses to measles virus nucleocapsid protein and the interferon-gamma signature, the exact role of the immune system in Paget's disease of bone is not well understood. The cytokine profiles, such as the increased levels of IL-6 and the interferon-gamma signature observed in this disease, are also very similar to those observed in other osteoimmunological disorders. As a potential osteoimmunological disorder, the treatment of Paget's disease of bone may also benefit from progress made in targeted therapies, in particular for receptor activator of nuclear factor Kappa-B ligand and IL-6 signaling inhibition.

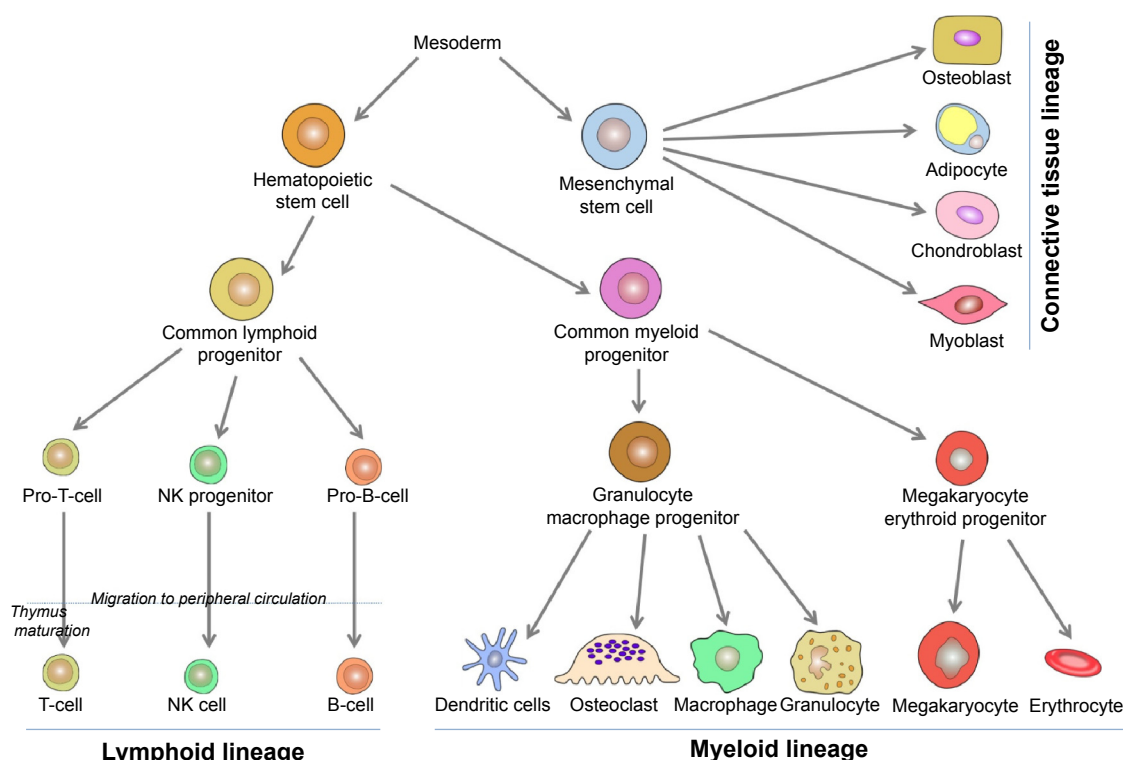
**Keywords:** Paget's disease of bone, SQSTM1/p62, osteoimmunology, osteoclast, RANKL

## Introduction

### Osteoimmunology at a glance

This narrative review of the literature presents first, data on osteoimmunology and osteoimmunological disorders, and second, discusses why Paget's disease of bone should be considered as a potential osteoimmunological disease. Osteoimmunology is an emerging research area that is the result of the cross talk regarding the relationship between bones and the immune systems. Various cell types are involved in osteoimmunology processes, but most of them originate from the hematopoietic tissue. At the third week of human embryonic development, stem cell lineages are formed in the yolk sac.<sup>1,2</sup> These lineages multiply asymmetrically, maintaining their original population and differentiating to other types of more specialized blood cells.<sup>3</sup> The immunological basic steps begin with the primary formation of hematopoietic stem cells lineage, which first appears in the yolk sac, followed by mesoderms of aorta, gonads, and nephrons (mesonephrons), and from there will migrate later to liver, spleen, and lymph nodes. Around the fourth month of fetal life, these hematopoietic stem cells migrate to bone marrow, and at the time of birth, bone marrow is responsible for all hematopoietic function.<sup>4,5</sup> Likewise, bone marrow pluripotent stem cells – which are stimulated by growth factors – are divided into two types of multipotent progenitor stem cells: common lymphoid cells and common myeloid cells (Figure 1).

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**Figure 1** Main osteoimmunological cell differentiations and cell lineages.  
**Abbreviation:** NK cell, natural killer cell.

## Lymphoid cells

Common lymphoid cells are further divided into three cell types: committed pro-natural killer stem cells, pro-T stem cells, and pro-B stem cells. Pro-natural killer stem cells migrate to peripheral circulation, where they become natural killer cells. Natural killer cells differ from B-cells and T-cells by not having clusters of differentiation 3 (CD3) like T-cells, nor CD19 and CD20 like B-cells. Pro-T stem cells (naïve T-cells) migrate from bone marrow to peripheral circulation, mature in thymus gland, and then some return to peripheral circulation where they become mature T-cells. Some of these mature cells will become T helper (TH) naïve (containing markers CD3, CD4) lymphocytes or T cytotoxic (containing markers CD3, CD8) lymphocytes. TH naïve cells differentiate into TH1 cells and TH2 cells. TH1 cells when activated by interleukin (IL)-4 and IL-5 contribute to convert B-cells to plasma cells called active B-cells producing immunological antibodies. TH2, when activated by interferon-gamma (IFN- $\gamma$ ) and tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ), contribute to activate monocytes to be highly active macrophages, epithelioid cells (modified monocytes), and giant cells. IFN- $\gamma$  receptor knockout mice showed exaggerated bone destruction in inflammatory arthritis in comparison with normal mice. IFN- $\gamma$  induces TNF receptor-associated factor 6 (TRAF6)

ubiquitination and degrades proteolytic TRAF6, ultimately leading to the inhibition of receptor activator of nuclear factor Kappa-B ligand (RANKL)-mediated osteoclastogenesis.<sup>6,7</sup> Activities of TH2 cells constitute the humoral immunity, while activities of TH1 cells and T cytotoxic cells create the cellular immunity. Pro-B stem cells differentiate in turn to be mature B-cells in peripheral blood circulation.

## Myeloid cells

Common myeloid progenitor cells differentiate into granulocyte/macrophage progenitor and megakaryocyte erythroid progenitor (MKEP) cells (Figure 1). Granulocyte/macrophage progenitor cells are divided into monocytes and granulocytes. Later, monocytes migrate to some tissues, reside there, and change their name depending on the tissue, such as monocytes that migrate to inflammation sites are called macrophages, monocytes that migrate to skin are called Langerhans cells, and monocytes that migrate and reside in bone tissue, which will be differentiated into bone resorbing cells are called osteoclasts.<sup>8,9</sup> Osteoclasts work mainly at the bone resorption activity controlling the bone turnover cycle.<sup>10</sup> Osteoclasts are large multinucleated cells of hematopoietic origin. They have the capability of removing organic and mineral components of bone. The macrophage

lineages and the myeloid dendritic cell originate hematopoietically, and they are affected by the cytokines and produce many of them. On the other hand, osteoblasts originate from mesenchymal stem cells and play important roles in bone formation, osteoclasts differentiation, and hematopoietic cell growth and differentiation.<sup>11,12</sup> The interaction or cross talk between osteoblast and osteoclasts play a central role in the osteoimmunological processes. Osteoblasts can control the osteoclastogenesis by two important cytokines; first the RANKL, which is a TNF member superfamily of proteins (Table 1). RANKL is a protein produced by *Tumor Necrosis Factor ligand Super-Family member 11 (TNFSF11)* gene. It has also been called TNF-related activation-induced cytokine or osteoprotegerin (OPG) ligand because it can be a ligand for osteoprotegerin decoy receptor. RANKL binds to RANK receptors, which normally are present at the pre-osteoclasts' cell membrane. Its crucial role as a transmembrane protein synthesized by osteoblasts is to perform maturation, differentiation, and activation of osteoclasts.<sup>13</sup> Second, OPG is also a member of the TNF superfamily and plays a role of a decoy receptor of RANKL leading to inhibition of osteoclasts maturation, differentiation, and activation and then leading to osteoclast apoptosis (Table 1). So, the balance between RANKL and OPG can modulate the level of bone resorption.<sup>14–16</sup> OPG works like a brake against the excessive bone resorption activity. A new inhibitory mechanism against OPG via autoantibodies has been revealed by studies of Riches et al. Indeed, they discovered autoantibodies against OPG in a man with celiac disease, severe osteoporosis, and high bone turnover.<sup>17</sup>

## Osteoimmunological cytokines

IL-1 is a very essential cytokine in osteoimmunological processes. The analysis of supernatants from phytohemagglutinin-stimulated peripheral blood monocytes in healthy humans suggested that IL-1 acts as the main stimulus of osteoclast-activating factor, which has a central role in osteoclastogenic activity. Subsequently, the same bone resorbing stimulating activity was found in TNF- $\alpha$  and IL-6. Indeed, IL-1, IL-6, and TNF- $\alpha$  increase the osteoclasts response to RANKL and consequently osteolysis (Table 1). Estrogen withdrawal after menopause has the same stimulating effect, increasing osteoclastic activity through IL-1, IL-6, and TNF- $\alpha$  effects.<sup>18</sup> Proinflammatory cytokines such as TNF- $\alpha$ , IL-1, IL-6, and IL-17 (Table 1) are also elevated in patients with rheumatoid arthritis, contributing to increased RANKL expression and subsequent osteolysis.<sup>19</sup> Schett et al have reviewed the important relation between autoimmunity and

joint erosion in rheumatoid arthritis, revealing the presence of anti-citrullinated protein antibodies and anti-carbamylated protein antibodies in serum of the patients with rheumatoid arthritis. Molecular interaction between anti-citrullinated protein antibodies and the surface of osteoclast precursor cells via citrullinated vimentin induces differentiation and production of bone-resorbing osteoclasts, resulting in excessive bone resorption.<sup>20</sup> Vitamin D<sub>3</sub>, prostaglandin E<sub>2</sub>, parathyroid hormone, in addition to IL-1, IL-6, IL-11, and TNF- $\alpha$ , can also induce RANKL expression, leading to excessive osteoclastogenesis (Figure 2).<sup>21</sup> Activated T-cells were also reported to regulate bone loss and activation of osteoclastogenesis in vitro through RANKL.<sup>22</sup> Contrariwise, TNF-stimulated gene 6 protein is an inflammation-induced protein that can inhibit osteoblastogenesis and osteoclast activation.<sup>23</sup> In addition, immunoreceptor tyrosine-based activation motif (ITAM) pathway may contribute to the relationship between immune system and bone as a co-stimulatory pathway in osteoclasts. ITAM-dependent receptors regulate myeloid-derived cells functions. Furthermore, ITAM-containing adapter proteins such as DNAX activation protein-12 and the Fc epsilon receptor I gamma chain (FCER1G) play an essential role in osteoclast differentiation. Suppression of calcineurin–nuclear factor of activated T-cells signaling can reduce the activity of ITAM pathway in the late stage of osteoclast differentiation, leading to the reduction of osteoclast differentiation and activity.<sup>24,25</sup> Calcium signaling induces the calmodulin-dependent kinase pathway role in osteoclast formation and plays a crucial role in the autoamplification of the transcription factor nuclear factor of activated T-cells cytoplasmic-1. Further, activation of TRAF6 and c-Fos pathways by RANKL leads to autoamplification of nuclear factor of activated T-cells cytoplasmic-1 and enhances osteoclastogenesis.<sup>21</sup>

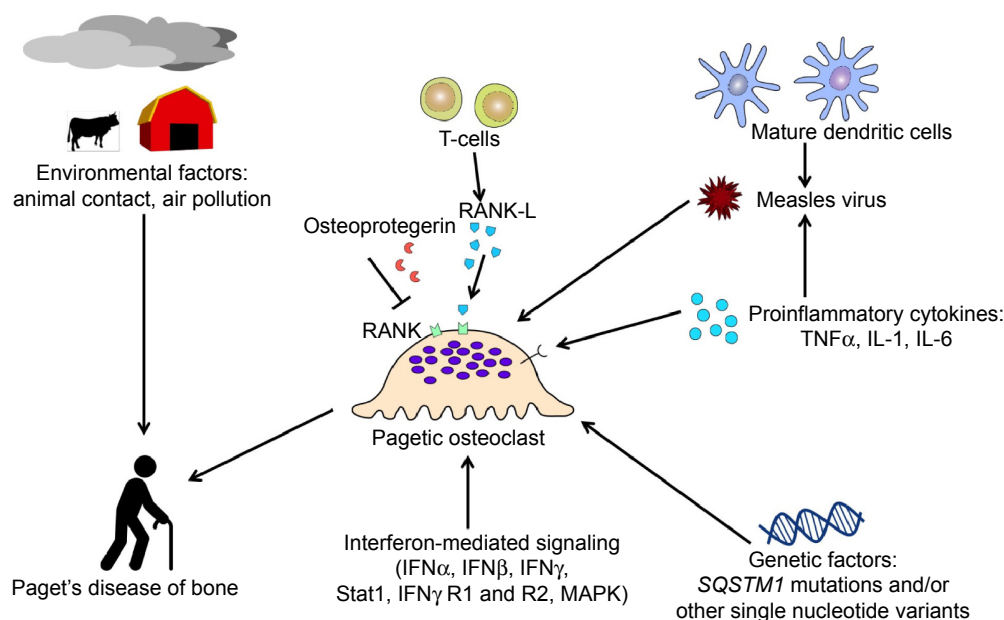
## Most frequent rheumatic osteoimmunological disorders and their related serum biomarkers

The most frequent rheumatic osteoimmunological disorders regroup bone metabolic diseases, such as osteoporosis and Paget's disease of bone, systemic autoimmune diseases such as rheumatoid arthritis, systemic lupus erythematosus and systemic sclerosis, and other rheumatic diseases including osteoarthritis and spondyloarthropathies, whereas periodontitis is frequently associated with systemic rheumatic conditions (Table 2). In almost all these disorders, serum levels of osteoimmunological biomarkers have been characterized in the literature (Table 2), and they can be combined to define a

**Table 1** Main cytokines involved in osteoimmunology processes, cells of production, and main roles in osteoimmunology

Cytokines	Cells or sites of production	Roles in osteoimmunology	References
Epidermal growth factor	Tumors	Increases osteoclasts formation and bone resorption	51
Fibroblast growth factor (acidic)	Bone matrix, osteoblasts	Increases bone formation	51
Fibroblast growth factor (basic)	Bone matrix, osteoblasts	Increases bone formation	51
GM-CSF	Stromal cells, Paneth's cells, macrophages, dendritic cell, mast cells, endothelial cells, smooth muscle cells, fibroblasts, chondrocytes, as well as IL-23-stimulated TH17 cells, IL-1 $\beta$ -stimulated TH1 and TH17 cells	Induces stem cells to produce granulocytes (neutrophils, eosinophils, basophils) and monocytes Induces macrophages and dendritic cells to differentiate and to activate RANKL signaling and osteoclastogenesis inhibition	52
Insulin growth factor- I	Osteoblasts, bone matrix	Increases bone formation	53
Interferon- $\gamma$	TH1 cells, natural-killer cells	Immunological natural antiviral and antitumor Aberrant expression leads to auto-inflammatory and autoimmune diseases	54, 55
Interleukin- I $\beta$	Leukocytes, osteoblasts, tumors	Induces TH1(T helper I) differentiation RANKL signaling and osteoclastogenesis inhibition	34
Interleukin-4	TH2 cells, NKT cells, and unclear but possible other sites are mast cells and basophils	Increases osteoclasts, formation, and bone resorption Helps in bone formation Induces TH2 to differentiate	35
Interleukin-5	Lymphocytes T Helper 2 (TH2)	Plays essential role for thymic stromal lymphopoietin in TH2-mediated immunity (lymphocyte development)	
Interleukin-6	TH2 cells, dendritic cells, leukocytes, osteoblasts, tumors	RANKL signaling and osteoclastogenesis inhibition Induces eosinophil to proliferate, to differentiate, to be mature, and to migrate then to survive Prevents apoptosis of eosinophil	56, 57
Interleukin-10	TH2 cells	Bone resorption may be increased via RANKL induction of mesenchymal cells	34
Interleukin-17	TH17 cells, memory T-cells	Its role in bone formation is unknown TH17 cell differentiation	58
M-CSF (CSF-1)	Osteoblasts/stromal cells	Induces expression of other cytokines such as IL-6, IL-8 and granulocyte colony-stimulating factor, and controls cartilage matrix turnover	59
Osteoprotegerin	Osteoblasts/stromal cells	RANKL induction of mesenchymal cells Helps mononuclear phagocytes to survive, to proliferate, and to differentiate Decoy receptor for RANKL	60, 61 62
Platelet derived growth factor	Platelets, osteoblasts, bone matrix, tumors	Inhibition of differentiation, activation, and bone resorption	51
RANKL	Osteoblasts/stromal cells, osteocytes, T-cells	Increases osteoclasts formation, and bone resorption, and increases bone formation	26
Transforming growth factor- $\beta$	Osteoblasts, bone matrix, leukocytes	Increases differentiation and maturation of osteoclasts, and bone resorption	63
Tumor necrosis factor-alpha (TNF- $\alpha$ )	Monocytes/macrophages, TH1 cells	Increases osteoclasts formation, and bone resorption, and increase bone formation Involved in lipid metabolism, coagulation, insulin resistance, and endothelial function RANKL induction of mesenchymal cells	63, 64
Tumor Necrosis Factor-beta (TNF- $\beta$ )	Leukocytes, tumors	Synergy with RANKL Increases osteoclasts formation, and bone resorption Helps in bone formation	51

**Abbreviations:** GM-CSF, Granulocyte-macrophage colony-stimulating factor; IL, Interleukin; TH, T helper; RANKL, Receptor Activator of Nuclear factor Kappa-B Ligand; NKT, natural-killer T; M-CSF, macrophage-colony stimulating factor.



**Figure 2** Pathophysiology of Paget's disease of bone and its relation to osteoimmunological cells and cytokines.

**Abbreviations:** RANKL, Receptor Activator of Nuclear factor Kappa-B Ligand; TNF, Tumor Necrosis Factor- $\alpha$ ; IFN, Interferon; MAPK, mitogen-activated protein kinase.

specific osteoimmunological pattern associated with a given rheumatic disease. For example, bone formation markers are usually increased in all these diseases, except in osteoporosis and rheumatoid arthritis (decreased level) and in systemic lupus erythematosus and systemic sclerosis (normal level). Bone resorption markers are also usually increased except for osteoarthritis and systemic lupus erythematosus (normal level). In addition, some proinflammatory cytokines may be of paramount importance at differentiating different rheumatic disorders. IL-17 and IL-6 levels are usually simultaneously increased in the same disorders, except for Paget's disease of bone and systemic sclerosis; in both diseases, IL-6 is elevated but not IL-17. Finally, the pattern of IFN- $\gamma$  serum levels in rheumatic diseases is very interesting: it is increased in almost all diseases, with the exception of osteoporosis and psoriatic arthritis (decreased level), and rheumatoid arthritis and ankylosing spondylitis (normal level). Overall, a combination of one serum biomarker of bone formation, one bone resorption biomarker, two proinflammatory cytokines such as IL-17 and IL-6 in addition to IFN- $\gamma$  serum levels would be able to classify with a good sensitivity the most frequent rheumatic osteoimmunological disorders. Adding other already available biomarkers in clinical practice, such as autoantibodies, would increase the specificity of such a combination, and its clinical utility (ie, combination of markers for outcome/prognosis prediction and/or a pharmacogenomic test to guide the choice of any targeted biotherapy) may further be validated in prospective cohorts.

## Paget's disease of bone as a potential osteoimmunological disorder

### Paget's disease of bone

Paget's disease of bone is the second most frequent metabolic bone disorder after osteoporosis,<sup>26</sup> where more than 3% of Caucasians older than 55 years are affected. This disorder is characterized by an excessive increased bone resorption by osteoclasts accompanied by aberrant osteoblastic bone formation. This aberrant bone remodeling causes fragile and weaker bones. To date, about 30 mutations in *SQSTM1/p62* gene have been reported in familial forms and unrelated patients with Paget's disease of bone. Furthermore, several common single nucleotide polymorphisms have been associated with Paget's disease of bone, in genome-wide association study, in particular in *CSF1*, *OPTN*, *TNFRSF11A*, *PML*, *RIN3*, and *NUP205* genes.<sup>26–28</sup> The consequences of these polymorphisms on osteoclast phenotype and activity are yet unknown.

### SQSTM1/p62 role and importance of osteoclastogenesis in Paget's disease

The SQSTM1/p62 protein anatomical structure has some important domains that regulate their essential functions such as Phox and Bem1p (PB1), ZZ, TRAF6 binding domain, LIR, KIR, and ubiquitin-associated (UBA) domains.<sup>29</sup> PB1 plays a role in adipogenesis by inhibiting ERK1, and it also activates NF- $\kappa$ B pathway through interaction with PKC $\zeta$ . ZZ domain activates NF- $\kappa$ B through the interaction with receptor interacting protein.

**Table 2** Pattern of serum biomarkers in most frequent rheumatic osteoimmunological disorders

	Bone metabolic diseases		Systemic autoimmune rheumatic diseases		Other rheumatic diseases				Periodontitis
	OP	PDB	RA	SLE	SSc	OA	PsA	AS	
References	65, 66	47, 67–71	72–80	78, 81–84	85–98	79, 99, 100	101–103	102, 104–113	114–127
<b>Bone formation markers</b>									
Total alkaline phosphatase	++			=					+
Bone alkaline phosphatase	++				=	+	+	+ or =	
Osteocalcin	+		–	+ or =	+ or =	+	=	+	+ or =
PINP	++				=				
PICP	+			=	=				
SPARC					+				
Sclerostin	+							=	+
Dickkopf-related protein 1	+		+	+			+	=	=
<b>Bone resorption markers</b>									
CTX-I	++			+	++	=	+	+	+
ICTP	+		+						+ or =
Urinary pyridinoline	+				+				
Osteoprotegerin	+		+	=	+	+	= or +	+ or –	
Urinary deoxypyridinoline	+			=	+		+	+	
TRAP5b								=	
RANKL	++				+	=	=	=	++
uNTX	++		+					=	
Matrix metalloproteinases					++				+
Osteopontin			+	+	++				++
<b>Cytokines</b>									
Interleukin-17	+	–	+ or =	+	–	= or +		+	+
Interleukin-6	+	+	++	++	+	+	+	++	+
Tumor necrosis factor- $\alpha$	+	=	=	++	+	=	+ or =	+	+ or –
Interleukin-1	+	=	=		+	=		=	+
Interferon- $\gamma$	–	+	=	+	++	+	–	=	+
Interleukin-4	–		=			=			

**Notes:** +, elevated levels; ++, very elevated levels; =, normal levels; –, decreased levels; —, very decreased levels.

**Abbreviations:** OP, osteoporosis; PDB, Paget's disease of bone; RA, rheumatoid arthritis; SLE, systemic lupus erythematosus; SSc, systemic sclerosis; OA, osteoarthritis; PsA, psoriatic arthritis; AS, ankylosing spondylitis; PINP, serum procollagen N-propeptide of type I collagen; PICP, carboxy-terminal propeptide of type I procollagen; SPARC, serum protein acidic and rich in cysteine; CTX-I, serum cross-linked C-telopeptide of type I collagen; ICTP, cross-linked carboxyterminal telopeptide of type I collagen; TRAP-5b, Tartrate-resistant acid phosphatase 5b; RANKL, Receptor activator of nuclear factor- $\kappa$ B ligand; uNTX, Urinary cross-linked N-telopeptide of type I collagen.

Interaction of TRAF6 with the TRAF6 binding domain can activate NF- $\kappa$ B pathway. SQSTM1/p62 can activate autophagy by interaction of LC3 with LIR domain, and autophagy can be inhibited by mTOR. The UBA domain of SQSTM1/p62 has a very important role for this protein function; it interacts non-covalently with ubiquitin protein to perform post-transcriptional modifications and degradation by 26S multisubunit protease or by autophagy. The UBA domain also has an important role in induction and activation of some transcription factors such as NF- $\kappa$ B. In osteoclasts, NF- $\kappa$ B–RANK signaling pathway is very important for osteoclastogenesis. With impairment of UBA functions, ubiquitin protein cannot interact with its domain in SQSTM1/p62 disrupting the autophagy and NF- $\kappa$ B signaling pathways, and consequently, osteoclastogenesis.<sup>26,30,31</sup> The KIR domain of SQSTM1/p62 plays a role in oxidative stress with Keap1 (cysteine-rich protein) that has antioxidant elements such as antioxidant response elements/electrophile response element in Nrf2 pathway. Keap1-Nrf2 has a cytoprotective action against oxidative stress, and Keap1 can be downregulated by SQSTM1/p62.<sup>32</sup> Autophagy dysfunction may result as a consequence of *SQSTM1* gene mutations and proteasomal pathway impairment. Ubiquitinated proteins are usually directed by SQSTM1/p62 protein to autophagosome. After binding to Atg8/LC3 at the surface of the autophagosome, they aggregate into polyubiquitinated non-functional proteins within the autophagosome in the autophagosome. Mutation in *SQSTM1/p62* can induce abnormal autophagy process, leading to the accumulation of aggregated ubiquitinated proteins, which stimulate osteoclastogenesis by triggering of NF- $\kappa$ B pathway, and may contribute to Paget's disease of bone.<sup>33</sup> Athanasiou's group reported very interesting data on pathogenic excessive osteoclastogenesis in Paget's disease of bone.<sup>34</sup> Neale et al found that macrophage-colony stimulating factor and IL-6 induce osteoclast formation and bone resorption in Paget's disease. Furthermore, elevated macrophage-colony stimulating factor in the serum may correlate with disease activity in patients with Paget's disease.<sup>34</sup> Kudo et al have also shown that IL-6 and IL-11 can enhance osteoclast formation and bone resorption through RANKL-independent mechanism. Then, the role of dexamethasone in osteoclastogenesis was suggested.<sup>35</sup> Indeed, dexamethasone can enhance proliferation and differentiation of human osteoclast precursors and suppress the bone resorption by mature osteoclasts.<sup>36</sup>

## Role of Optineurin in Paget's disease of bone

Mutations of *Optineurin (OPTN)* gene have been reported in glaucoma and amyotrophic lateral sclerosis, as well as

common genetic variants; in particular, the intronic variant rs1561570 was found to be associated with Paget's disease of bone.<sup>37</sup> The *OPTN* gene encodes a 67 kDa cytosolic protein that consists of 577 amino acids. The importance of OPTN was first raised after discovering disease-causing mutations in primary open-angle glaucoma.<sup>38</sup> In addition, OPTN may be involved in neurofibrillary tangles and dystrophic neuritis that leads to Alzheimer's disease, amyotrophic lateral sclerosis, Parkinson's disease, Creutzfeldt–Jakob disease, and glial cytoplasmic inclusions that lead to a rare neurological disorder called multiple system atrophy.<sup>39</sup> Although the exact role of OPTN in Paget's disease of bone pathogenesis is unknown yet, interaction between OPTN and TANK-binding kinase 1 may suggest a connection with the immune system. Indeed, TANK-binding kinase 1 is a TNF- $\alpha$ -activated protein kinase, which may be activated by viral double-stranded DNA or by lipopolysaccharide. On the other hand, induction of IFN- $\beta$  due to RNA virus infection can be suppressed by OPTN.<sup>39–41</sup>

## Environmental factors

Environmental factors may also contribute to Paget's disease of bone.<sup>26</sup> Although controversial in the literature, several studies have found a relationship between viral infections and excessive enhanced osteoclasts activity.<sup>42</sup> Inclusion bodies contained in osteoclasts were reported to be similar to *Paramyxoviral* nucleocapsids. Measles virus, respiratory syncytial virus, and canine distemper virus may play a role in Paget's disease of bone.<sup>43</sup> The expression of measles virus nucleocapsid protein (MVNP) in osteoclasts was reported to lead to the formation of pagetic-like osteoclasts. MVNP is known to increase the production of IL-6 that in turn leads to increase the production of TAFII-17 and increase the sensitivity of osteoclasts to 1,25-(OH)<sub>2</sub>D<sub>3</sub>. The pagetic phenotype of osteoclast is characterized by hypermultinucleation and hypersensitivity to 1,25-(OH)<sub>2</sub>D<sub>3</sub>. NF- $\kappa$ B signaling can be increased in cells by increasing the production of IL-6 and IL-1.<sup>44</sup>

## Paget's disease as a potential osteoimmunological disorder

Paget's disease of bone should be considered as a potential osteoimmunological disorder for several reasons. First, the RANKL-NF- $\kappa$ B signaling has a major role in pagetic osteoclast differentiation and activation, and the cytokine profile observed in this disease is very similar to those observed in other osteoimmunological diseases (Table 1). However, the exact role of the immune system in Paget's disease of bone is not very well understood, except for cellular responses to

**Table 3** Overview of the main clinical trials or case reports in which a cytokine or its receptor, involved in osteoimmunological process related to a rheumatic or musculoskeletal disorder, was targeted

Targeted signalling	Molecule name	Molecule description	Main rheumatic diseases treated	Level of evidence	References
IL-1	Anakinra	Human recombinant IL-1ra	Rheumatoid arthritis NOMID syndrome Still's disease Periodic fever syndromes Acute gout Behçet's disease	Randomized-controlled trials Observational study Case reports Case reports Observational study Case reports	40, 128–133
	Rilonacept	IL-1 trap, IL-1 inhibitor	CAPS syndromes Periodic fever syndromes Acute gout	Randomized-controlled trials Case reports Observational study	
	Canakinumab	Interleukin-1 $\beta$ blocker	CAPS syndromes Systemic juvenile idiopathic arthritis Acute gout Rheumatoid arthritis Behçet's disease	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Case reports	
			Rheumatoid arthritis	Randomized-controlled trials	
			Juvenile idiopathic arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Juvenile Paget's disease	Case report	
IL-6	Tocilizumab	IL-6 receptor inhibitor	Rheumatoid arthritis	Randomized-controlled trials	134
IL-17	Secukinumab	IL-17 inhibition	Juvenile idiopathic arthritis	Randomized-controlled trials	135–137
	Ixekizumab	IL-17 inhibition	Psoriatic arthritis	Randomized-controlled trials	
	Brodalumab	IL-17 receptor inhibition	Psoriatic arthritis	Randomized-controlled trials	
IL-23	Ustekinumab	IL-12 and IL-23 inhibition	Psoriatic arthritis	Randomized-controlled trials	138
OPG	Recombinant osteoprotegerin	Recombinant osteoprotegerin	Juvenile Paget's disease	Case report	139
RANKL	Denosumab	RANKL inhibition	Osteoporosis Treatment-induced bone loss Bone metastases Multiple myeloma Hypercalcemia of malignancy Giant cell tumor of the bone Rheumatoid arthritis Juvenile Paget's disease Paget's disease of bone	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Case report Case report	48, 140–143
			Rheumatoid arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Ankylosing spondylitis	Randomized-controlled trials	
			Juvenile idiopathic arthritis	Randomized-controlled trials	
			Rheumatoid arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Ankylosing spondylitis	Randomized-controlled trials	
			Behçet's disease	Case reports	
			Rheumatoid arthritis	Randomized-controlled trials	
TNF- $\alpha$	Etanercept	TNF inhibitor (decoy receptor)	Psoriatic arthritis Ankylosing spondylitis Juvenile idiopathic arthritis	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials	144–148
	Infliximab	TNF- $\alpha$ inhibition	Rheumatoid arthritis Psoriatic arthritis Ankylosing spondylitis Behçet's disease	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Case reports	
	Adalimumab	TNF- $\alpha$ inhibition	Rheumatoid arthritis Psoriatic arthritis Ankylosing spondylitis Juvenile idiopathic arthritis	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials	
	Golimumab	TNF- $\alpha$ inhibition	Rheumatoid arthritis Psoriatic arthritis Ankylosing spondylitis	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials	
	Certolizumab	TNF- $\alpha$ inhibition	Rheumatoid arthritis Psoriatic arthritis Ankylosing spondylitis	Randomized-controlled trials Randomized-controlled trials Randomized-controlled trials	
			Rheumatoid arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	
			Ankylosing spondylitis	Randomized-controlled trials	
			Rheumatoid arthritis	Randomized-controlled trials	
			Psoriatic arthritis	Randomized-controlled trials	

**Abbreviations:** IL, Interleukins; NOMID, Neonatal-onset multisystem inflammatory disease; CAPS, Cryopyrin-associated autoinflammatory syndrome; OPG, osteoprotegerin; RANKL, Receptor Activator of Nuclear factor Kappa-B Ligand; TNF- $\alpha$ , Tumor Necrosis Factor- $\alpha$ .

MNVP. Second, dendritic cells may also play a role in the pathogenesis of Paget's disease of bone.<sup>45</sup> Immature myeloid dendritic cells express CDw150, a signaling lymphocyte activation molecule acting as a receptor for *measles* virus.

Dendritic cells matured by stimulation of Toll-like receptors 2 and 4 will overexpress CDw150 up to fivefold. Then, human dendritic cells may increase the expression of measles virus, the latter contributing to Paget's disease of bone

(Figure 2).<sup>46</sup> In addition, Nagy et al were the first authors to show in the literature a remarkable increase in IFN-mediated signaling in Paget's disease of bone. Increasing expression of IFN- $\alpha$ , IFN- $\beta$ , and IFN- $\gamma$  messenger (m)RNA, STAT-1, IFN- $\gamma$  receptors 1 and 2, and mitogen-activated protein kinase were found in monocytes and lymphocytes from patients with Paget's disease in comparison with healthy controls, suggesting a possible post-viral reaction in Paget's disease of bone.<sup>47</sup>

## Main osteoimmunological cytokines as therapeutic targets

A large majority of the already known osteoimmunological cytokines or their receptors have been targeted by monoclonal antibodies, humanized or not, and they are now indicated in several rheumatic disorders, mostly in rheumatoid arthritis (Table 3). The treatment of Paget's disease of bone mostly relies on bisphosphonates to control disease activity, and no monoclonal antibodies have been investigated in this indication yet. But as a potential osteoimmunological disorder, the treatment of Paget's disease of bone may also benefit from progresses in targeted therapies. For instance, denosumab, a monoclonal antibody inhibiting RANKL, could be relevant to treat Paget's disease of bone as it prevents the osteoclastogenesis and promotes the apoptosis of mature osteoclasts. A case report has now been published on using denosumab in a patient with Paget's disease of bone and impaired renal function, which contraindicated the use of bisphosphonates. In this case, the authors reported that denosumab 60 mg subcutaneously administrated at 0 month, 6 months, 9 months, 12 months, and 15 months has rapidly decreased the activity of Paget's disease of bone as measured by biochemical markers and bone scan.<sup>48</sup> It is worth mentioning that IL-6 was found to be elevated in pagetic osteoclasts in bone marrow and in peripheral blood of patients with Paget's disease of bone.<sup>32</sup> However, some other studies by Neale et al found low serum levels of IL-1 beta, IL-6, and TNF- $\alpha$  in 13 patients with Paget's disease of bone in comparison with eight healthy controls.<sup>34</sup> As the IL-6 signaling is induced by MVNP, inhibition of IL-6 signaling should inhibit the development of pagetic osteoclasts.<sup>49</sup> Then, considering the high levels of IL-6 that usually characterize Paget's disease of the bone, inhibiting the IL-6 signaling by drugs should also be considered as a future therapeutic avenue that is as yet unexplored.<sup>50</sup>

## Conclusion

In conclusion, Paget's disease of bone should be considered as a new addition to the large family of osteoimmunological disorders. The cytokine profiles observed in this disease are

also very similar to those observed in other osteoimmunological disorders that should probably be classified accordingly. The treatment of Paget's disease of bone may also benefit from progresses in osteoimmunology-targeted therapies, in particular, RANKL and IL-6 signaling inhibition.

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## Disclosure

The authors report no conflicts of interest in this work.

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