Dendrimers To Treat Rheumatoid Arthritis

Xavier Bosch*

Department of Internal Medicine, Hospital Clínic, University of Barcelona, Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Barcelona, Spain

ABSTRACT In comparison with linear polymers, dendrimers' multivalency and nanostructure confer substantial advantages in drug delivery including rapid cell entry, targetability, and easier passage across biological barriers. Previous work has shown that phosphorus-containing dendrimers capped with anionic azabisphosphonate (ABP) end groups prompt anti-inflammatory activation of human monocytes. By using two mouse models of arthritis mimicking human rheumatoid arthritis (RA), Hayder *et al.* recently demonstrated that intravenous injection of dendrimer ABP inhibits the secretion of proinflammatory cytokines and osteoclastogenesis—two fundamental monocyte-dependent processes of inflammation and bone erosion in RA. While available biological therapies for RA target only one of the cytokines involved in inflammation or bone erosion, dendrimer ABP, by virtue of its double action on both processes in mice, might become a more active and cost-saving alternative for RA patients. This Perspective highlights this important development and the challenges that lie ahead.

Dendrimers are well-defined nanostructured macromolecules that have thin masses or size polydispersity and a tree-like design characterized by exponential numbers of separate dendritic branches radiating out from a joint core.[1,2] These molecules are characterized by a high level of synthetic control over critical nanoscale design parameters, including shape, size, surface functionality, and inner void. These characteristics mean they are archetypal structures for understanding an extensive variety of vital nanomedical issues. Their nanoarchitecture provides structural advantages such as fast entry into cells, abridged macrophage uptake, targetability, and easier passage through biological hurdles by transcytosis in comparison with linear polymers, larger nanoparticles, and liposomes.[1] Compared with linear polymers, dendrimers are multivalent, due to their multiplicity of reactive surface end groups, making them ideal drug carriers with potentially higher payloads.[3] The Tomalia-type poly(amidoamine) (PAMAM) Starburst series was the first group of dendrimers to be commercialized and remains the best-characterized and most-studied group. The use of dendrimers to transport therapeutic agents to intracellular target sites for pain control, inflammation, ocular, topical, oral, and transdermal delivery is shown in Table 1.

Dendrimers are multivalent, due to their multiplicity of reactive surface end groups, making them ideal drug carriers with potentially higher payloads.

However, dendrimers also turn into drugs themselves.[4] Dendrimer glucosamine conjugates have been used safely to avert the formation of scar tissue through antiangiogenic and immunomodulatory pathways.[5] Polyanionic dendrimers (with sulfonate groups) have been shown to act efficiently against some viruses.[6] Branched polyamines, such as PAMAM dendrimers, stimulate the elimination of prion proteins in brain homogenates of scrapie-affected mice.[7]

Some dendrimers, such as the PAMAM family, have *in vivo* anti-inflammatory activities, as shown by the inhibition of both proinflammatory cytokine secretion by human macrophages and dendritic cells and induced acute and chronic inflammatory disorders in rat models.[8,9] Poly(ethylene oxide) (PEO) dendrimers (also called poly(ethylene glycol), PEG, dendrimers) have shown anti-inflammatory properties by inhibiting the extravasation of inflammatory leukocytes across the endothelial barrier to sites of inflammation.[10]

Because dendrimers containing cationic end groups have demonstrated greater toxicity after repetitive intravenous (IV) injection, various dendrimers with anionic azabisphosphonate (ABP) endings have been synthesized.[8] In 2006, Poupot *et al.* reported that a phosphorus-containing dendrimer with an

* Address correspondence to xavbosch@clinic.ub.es.

Received for review August 19, 2011

Published online August 31, 2011
10.1021/nn203190x

TABLE 1. Applications and Functional Flexibility of Dendrimers[a]

use	agent	dendrimer	role
intracellular delivery	colchicine	glycopeptide	carrier
gene delivery and transfection agents	DNA	PAMAM	binding
	DNA	PAMAM	complexation
	DNA/siRNA	Priostar-PAMAM	complex
antibody conjugates	antibody	PAMAM	binding (anthrax detection)
	monoclonal sheep antibody	PAMAM	carrier
encapsulating agents	methotrexate	PAMAM	liposomes
	indomethacin	PEG-mesylate	micelles
	etoposide	core PAMAM —caprolactone-PEG	micelles
complexes	mefenamic acid	ClOC-PEG-COC	complexation
	diclofenac, amino salicylic acid	G1-G3[b]	
	furosemide	G0-3 PAMAM G4	
	indomethacin	PAMAM (folate surfacized)	
ocular	porphyrin	aryl ether dendrimer	carrier (neovasculature)
	pilocarpine and tropicamide	G1.5-4 PAMAM	vehicle
	G3.5 PAMAM-glucosamine	G3.5 PAMAM	therapeutic agent (scar tissue inhibition)
transdermal	indomethacin	G4 PAMAM (OH and NH_2)	permeation enhancers
oral	piroxicam	G3-4 PAMAM	complex/solubility enhancer
	naproxen	G0 PAMAM	permeation enhancers
	sulfamethoxazole	G3 PAMAM	solubility enhancer
	propranolol	G3 PAMAM	solubility enhancer
parenteral	*N*-acetyl cysteine	G4 PAMAM	carrier
	chloroquine phosphate	G3—4 p-lysine-PEG(1000)	encapsulating agent
topical gels	5-fluorouracil	G2-G6 PAMAM	permeation enhancer
	nifedipine	G5 PAMAM	solubility and permeation enhancers
	SPL7013	G4 lysine-based dendrimer	therapeutic agent (anti-HIV agent)
colon delivery	5-amino salicylic acid	G3 PAMAM	carrier
in vivo injectables	amine, hydroxyl surface	G4 PAMAM	therapeutic agent (anti-inflammatory)

[a] Adapted from Menjoge *et al.*[1] [b] Each branching shell is termed a generation (G).

N_3P_3 (cyclotriphosphazene) core and phenoxymethyl-methylhydrazone (PMMH) branches, and capped with ABP end groups, selectively targets monocytes when cultured with human peripheral blood mononuclear cells *in vitro*.[11] In particular, a dendrimer binds to isolated monocytes and undergoes internalization in a few seconds. These experiments also showed that dendrimers follow the phagolysosome path during internalization by monocytes. This internalization is followed by anti-inflammatory activation of human monocytes, as revealed by gene expression analysis of dendrimer ABP-activated monocytes.[4] Dendrimer-activated monocytes can enhance $CD4^+$ T cell amplification, producing IL-10, an immunosuppressive cytokine, which is a crucial differentiation factor for regulatory T cells.[4] This dendrimer also activates the amplification of human natural killer cells by inhibiting the interleukin-2 (IL-2)—dependent proliferation of $CD4^+$ T lymphocytes.[8]

Of importance, Poupot *et al.*[11] demonstrated that surface phosphonic groups represent an imperative determinant for the bioactivity of phosphorus-containing dendrimers since they are much less active when capped with carboxylic acid groups. Moreover, the whole dendritic structure is another significant structural requisite for this bioactivity, as surface groups or branches alone do not elicit monocyte activation.

What part could the monocyte-activating properties of dendrimer ABP play in disease? Monocytes play an essential role in inflammation and osteoclastogenesis in rheumatoid arthritis (RA).[12] The proinflammatory cytokines IL-1β and tumor necrosis factor (TNF)-α are principally produced by monocytes and macrophages and synergize with the receptor activator of nuclear factor-κB (RANK) ligand (RANKL) to produce the mature osteoclasts that are responsible for the bone resorption typical of RA.[13] Inflammation and bone erosion are narrowly connected. Cytokines play a role in each phase of RA pathogenesis: they promote autoimmunity, sustain chronic inflammatory synovitis, and drive the destruction of contiguous joint tissue.[14] In RA, osteoclasts at the boundary between articular bone and synovial tissue prompt the bone resorption that allows the invasion of synovial membrane cells, leading to pannus formation. This process relies on the incursion of osteoclast hematopoietic precursors into inflamed synovial tissue and their cytokine-driven differentiation into mature osteoclasts; the essential cytokine mediators are RANKL and macrophage colony-stimulating factor (M-CSF), which are expressed by T helper 1 (TH1) cells and synovial fibroblasts.[14] In RA, M-CSF, and RANKL, signaling acts synergistically with proinflammatory cytokines, TNF-α, IL-1β, and IL-17 (produced

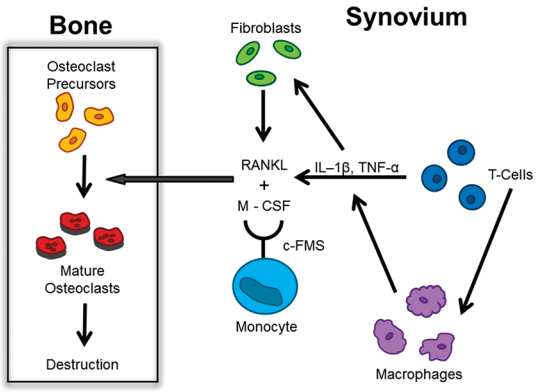

Figure 1. Mechanisms by which inflammation modulates osteoclast and bone resorption in RA. Cells of the myelomonocytic lineage (osteoclast precursors) under the influence of RANKL and M-CSF differentiate into osteoclasts. Activated T-cells promote osteoclastogenesis directly by production of RANKL. T-cells also stimulate the synovial macrophages to secrete proinflammatory cytokines that promote fibroblast production of RANKL. TNF-α, IL-1β, and IL-17 (not shown here) act synergistically with RANKL and M-CSF, which engages its receptor c-FMS on monocytes.

by T helper 17, TH17, cells).[8] Specifically, TNF induces M-CSF production by synovial fluid cells. M-CSF engages its receptor cellular-feline McDonough strain sarcoma virus oncogene homologue (c-FMS) on monocytes, prompting early differentiation into osteoclasts (Figure 1). RA therapy has been revolutionized by the introduction of monoclonal antibodies targeting these cytokines. In addition, TH17 cells have been identified as the selective osteoclastogenic T-cell subgroup.[15] In contrast, IL-4 and IL-10, which are produced by TH2 cells, as well as interferon (INF)-γ, inhibit osteoclast differentiation.[14]

Recently, Hayder *et al.*[8] tested the potential usefulness of dendrimer ABP in two established inflammatory arthritis models resembling human RA: IL-1 receptor antagonist deficient mice (IL-1ra−/− mice) and mice receiving K/BxN serum transfer. Initially, they compared the antiarthritic activity of weekly IV injections (10 mg/kg) of dendrimer ABP and two other dendrimer-based systems, dendrimer azamonophosphonate (AMP) and dendrimer polypropyleneimine (PPI), in mice. Only dendrimer ABP showed a significant reduction in inflammation and the external signs of arthritis at the age of 15 weeks, with complete inhibition at doses of 1 and 10 mg/kg (Figure 2). Microscopic analysis showed that mice treated with dendrimer ABP (10 mg/kg) had near-normal synovial membranes, preserved cartilage, normal joint appearance, and no osteoclasts in the bone matrix. In contrast, untreated IL-1ra−/− mice showed enlarged synovial membranes and inflammation with lymphocyte, macrophage, and neutrophil infiltration; complete cartilage erosion, indicating severe joint damage; and osteoclasts in the eroded bone matrix. Joints from dendrimer ABP-treated IL-1ra−/− mice showed no cellular expression of IL-17, a prominent pro-inflammatory cytokine in the pathogenesis of arthritis,[16] and no osteoclasts were identified in the bone matrix.

Hayder *et al.* then examined serum concentrations of proinflammatory and anti-inflammatory cytokines in untreated and dendrimer ABP-treated IL-1ra−/− mice. Dendrimer ABP-treated mice had dose-dependent reductions in serum levels of the proinflammatory TH1 cytokines IL-1β, TNF-α, IL-6, and IL-17 and reduced quantities of the enzymes responsible for cartilage degradation in RA, matrix metalloproteinase (MMP) 3 and 9, in comparison with untreated mice. When spleen immune cells (splenocytes) from IL-1ra−/− mice were cultured *ex vivo* in the presence of an anti-CD3ε monoclonal antibody, which targets T lymphocytes and initiates an activation stream of several cells, including B cells and monocytes, a reduction in the secretion of these proinflammatory cytokines, and of TH1 cytokines IL-2 and IFN-γ, was found following IV dendrimer ABP. In contrast, splenocyte secretion of anti-inflammatory TH2 cytokines IL-4 and IL-10 was augmented, suggesting that dendrimer ABP dose-dependently skews lymphocytes to an anti-inflammatory TH2 response. These results suggest that dendrimer ABP has antiarthritic and anti-inflammatory effects on spontaneous arthritis in IL-1ra−/− mice.

As a next step, Hayder *et al.* investigated whether dendrimer ABP has antiosteoclastic properties *in vitro*. Osteoclasts stem either from bone marrow precursors of monocytes or directly from monocytes, the chief cellular target of dendrimer ABP.[11] Bone marrow precursors of monocytes were obtained from IL-1ra−/− mice. Differentiation of these precursors into osteoclasts was induced by M-CSF and RANKL and was intensely inhibited *in vitro* by dendrimer ABP in a dose-dependent manner. Dendrimer ABP also inhibited human peripheral blood monocyte differentiation into osteoclasts induced by M-CSF and RANKL *in vitro*: this inhibition was related to the reduction in c-FMS expression.

Monocyte differentiation into osteoclasts is started through the interaction of M-CSF and its monocyte receptor c-FMS, which results in monocyte proliferation and the induction of RANK at the monocyte surface. RANK interacts with RANKL, which is expressed by bone-forming cells called osteoblasts and activated $CD4^+$ T cells, eventually promoting monocyte differentiation into osteoclasts.[13] Hayder *et al.* found that dendrimer ABP inhibited the proliferation of IL-1ra−/− arthritic mouse bone

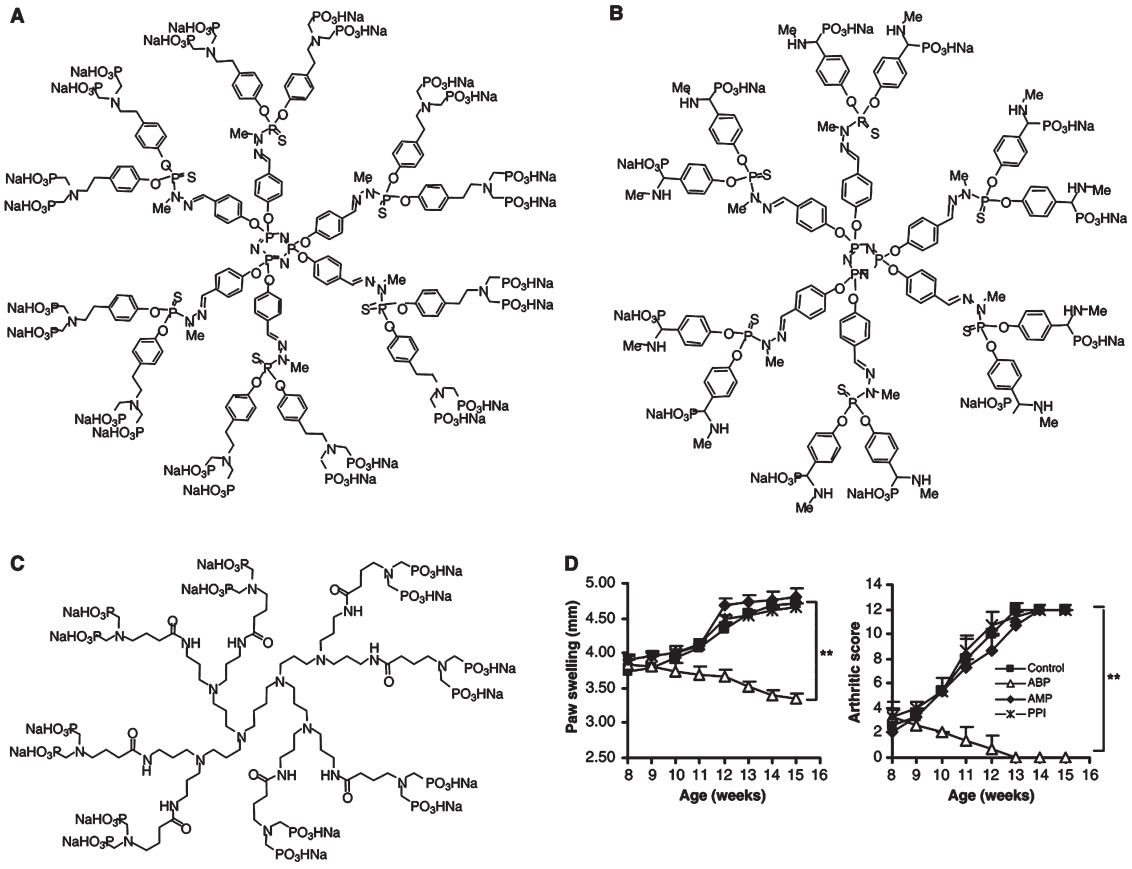

Figure 2. Structure of dendrimers used in the study by Hayder *et al*:[8] (A) dendrimer ABP; (B) dendrimer AMP; (C) dendrimer PPI; (D) comparison of the effect of dendrimer ABP, AMP, or PPI (10 mg/kg) on the development of external signs of arthritis (paw swelling and arthritic score) in IL-1ra−/− mice (n = 3 per treatment group). Control: untreated IL-1ra−/− mice (n = 3). Reproduced with permission from ref 8. Copyright 2011 American Association for the Advancement of Science.

marrow precursors of monocytes cultured in the presence of M-CSF and RANKL, and that the mechanism of action appeared to involve c-FMS. Dendrimer ABP dose-dependently reduced the expression of c-FMS and RANK, and the inhibition of c-FMS expression was also observed at the mRNA level in IL-1ra−/− mice compared with untreated IL-1ra−/− mice.

The inhibitory effect of dendrimer ABP on osteoclast differentiation was also demonstrated *ex vivo* in cultures of inflamed synovial tissue obtained from RA patients undergoing arthroplasty. In addition, both therapeutic and prophylactic doses of dendrimer ABP successfully suppressed arthritis and inflammation in the K/BxN serum transfer mouse model, suggesting that, in this model, arthritis flares could be prevented using this therapy.

> Dendrimer ABP (dendrimers with anionic azabisphosphonate endings) inhibited osteoclastogenesis and the secretion of proinflammatory cytokines, both of which are underlying monocyte-dependent processes in rheumatoid arthritis inflammation and bone damage.

Lastly, histologic comparison of several major organs (aorta, spleen, liver, lungs, and kidneys) between IL-1ra−/− mice treated with dendrimer ABP at the highest dose (10 mg/kg) for 12 weeks and normal mice showed that dendrimer ABP therapy had no off-target effects and remained nontoxic at 100 mM.

In summary, dendrimer ABP inhibited osteoclastogenesis and the secretion of proinflammatory cytokines, both of which are underlying monocyte-dependent processes in RA inflammation and bone damage. Mouse serum MMP levels were normalized after weekly dendrimer ABP injections. Dendrimer ABP-activated monocytes increased the secretion of the anti-inflammatory cytokines IL-10 by $CD4^+$ T cells and also IL-4, which suggested TH2 twisting. Dendrimer ABP's anti-inflammatory

activity and antiosteoclastogenesis effects appear to be mediated by inhibition of c-FMS expression both *in vivo* in mice and *in vitro* in human cells. Major organs showed no overt toxicity. Hayder *et al*. attributed the improvement in inflammation and suppression of joint damage to the general three-dimensional structure of dendrimer ABP, since other dendrimers such as AMP (identical branches and core as dendrimer ABP, but different surface groups) and PPI (different branches and core, but ABP surface groups) were ineffective in resolving arthritis in IL-1ra−/− mice.

Do these promising results suggest that dendrimer ABP may be a novel and potentially effective alternative to the newest therapies in RA? Rheumatoid arthritis is a chronic, systemic inflammatory disease affecting 0.8% of adults throughout the world. Disease onset is normally between 30 and 50 years of age, and the estimated incidence is 25 per 100 000 males and 54 per 100 000 females in the United States.[17] Although the pathogenesis of RA is still thought to be the initiation of a pathologic response due to incitement by a stimulus in people with genetic susceptibility, the disease has become a prototype for the application of new therapies based on elucidation of the molecular pathogenesis, rather than a disease whose pathogenesis is uncertain.[18]

Recent years have shown that effective therapies in RA involve early, aggressive pharmacotherapy with one or more disease-modifying antirheumatic drugs (DMARDs), usually together with nonsteroidal anti-inflammatory drugs (NSAIDs) and/or corticosteroids for pain relief and a degree of control of inflammation.[17] Most of these combination therapies still include methotrexate (MTX), an older oral DMARD, which remains the gold standard of treatment. However, traditional DMARDs have a slow onset to action combined with toxic effects, which requires enhanced monitoring.

In the past decade, the introduction of biological DMARDs, especially TNF-targeting agents, both as monotherapy and in combination with MTX, has substantially improved outcomes in RA. Biological therapies have much greater tolerability and efficacy than previous DMARDs.

Nevertheless, current biological therapies in RA have limitations related to safety, efficacy, and cost. Although TNF inhibitors have resulted in improved outcomes for many patients, they may be expensive, with treatment often costing more than US $1,000 monthly in the United States.[17] Likewise, TNF inhibitors are associated with safety risks in the long term, including cancer, especially lymphoma, and serious infections such as tuberculosis. Biological agents with alternative mechanisms of action are available for RA. These include the interleukin 1 receptor antagonist anakinra, a monoclonal antibody; rituximab, which binds to CD20 on B cells; abatacept, a fusion protein that binds to CD28 on T cells; and tocilizumab, a humanized monoclonal antibody that binds to the IL-6 receptor. An anti-RANKL antibody, denosumab, has shown that targeting RANKL efficiently delays osteoclast-mediated joint destruction, but has failed to resolve inflammation.[8]

Current biological therapies have not achieved the goal of uniform remission in RA and prolonged treatment is still associated with a lack of uniform induction of even a low disease activity state. Available biological therapies target only one of the cytokines involved in inflammation or bone erosion, but it seems highly improbable that one cytokine alone drives the pathogenesis of RA.[19] Therefore, novel alternatives are required in the continued search for true remission, the ultimate goal of RA therapy.

It is plausible to speculate that dendrimer ABP, by virtue of its direct dual action on both inflammation and bone erosion, might be more active in RA than current biological agents and could be an effective and cost-saving alternative in the future.

The study by Hayder *et al*. suggests that it is plausible to speculate that dendrimer ABP, by virtue of its direct dual action on both inflammation and bone erosion, might be more active in RA than current biological agents and could be an effective and cost-saving alternative in the future.

OUTLOOK AND FUTURE CHALLENGES

Although the results reported by this group point to early success in treating RA-like disease in mice with dendrimer ABP, they are also a pointer to future challenges. Methotrexate, the current standard of care for RA therapy is administered weekly, either orally or intramuscularly and can also be administered as a first-line drug in combination with TNF inhibitors. Hayder *et al*. demonstrate that dendrimer ABP therapy for RA can be administered in the same time frame as MTX.[8] Furthermore, they show that, although the need for repeated injections is one possible barrier to the use of dendrimer ABP in clinical practice, IV injections could be administered every 3 weeks, similar to current anticytokine therapies. Assessment of possible oral administration is

required. Future studies of the toxicity, tolerance, and pharmacokinetic properties of dendrimer ABP will provide a clearer picture of clinical feasibility.

The results found by Hayder *et al*. also suggest some possible applications of dendrimer ABP in other diseases. Psoriatic arthritis (PsA), which occurs in 10−15% of patients with psoriasis, a chronic immune-mediated disease, is also characterized by joint damage, with two-thirds of patients presenting with initial bone erosions.[20] In contrast, in systemic lupus erythematosus (SLE), a paradigm of systemic autoimmune disease, radiographs rarely display erosive changes, although nearly half of SLE patients present with joint pain.

Schwarz *et al*.[20] used a TNF transgenic mouse model of erosive arthritis and anti-TNF clinical trials in PsA patients to show that systemic TNF induces the migration of $CD11b^+$ osteoclast precursors from the bone marrow into peripheral blood. These osteoclast precursors then enter the joints through blood vessels, translocate across the RANKL-rich inflamed synovium, and differentiate into active osteoclasts. In contrast, SLE patients appeared to possess innate resistance to bone resorption: The authors' hypothetical explanation is that systemic IFN-α diverts the bone marrow-derived myeloid precursors away from the osteoclast lineage and stimulates their differentiation into dendritic cells. Furthermore, Schwarz *et al*. suggest that systemic TNF prompts osteoclastic differentiation of peripheral blood mononuclear cells in patients with PsA that correlates with the erosive behavior, and that the erosive phenotype of autoimmune disease is determined by the innate TNF/IFN axis.

Psoriatic arthritis patients also have higher circulating M-CSF levels and a positive correlation between M-CSF and RANKL levels and radiographic erosion. Circulating M-CSF concentrations also correlate with the percentage of peripheral blood $CD14^+/CD11b^+$ cells.[21]

Other cytokines secreted from activated T- and B-cells and other immunocompetent cells have been shown to be crucial in the pathogenesis of PsA through the induction of proliferation and activation of synovial and epidermal fibroblasts. These cytokines include TNF-α and IL-1, which may play a significant role in joint destruction.[22]

These results suggest that investigation of the effects of dendrimer ABP in PsA would be of interest. As there is still no animal model that closely mimics PsA; studies that explore the inhibitory effect of dendrimer ABP on *ex vivo* osteoclast differentiation in cultures of synovial tissue from surgical interventions in PsA patients could be the first step. As in the experiments by Hayder *et al*., synovial membrane could be cultured alone; in the presence of M-CSF and RANKL; or in the presence of M-CSF, RANKL, and dendrimer ABP.

In conclusion, the study by Hayder *et al*. illuminates the potential of phosphorus-containing dendrimers and, in particular, dendrimer ABP, as a promising nanotherapeutic that modulates innate immunity through its effects on monocytes. Determining whether it becomes a new drug candidate for combating the bone-eroding and inflammatory characteristics of RA in humans is a more distant goal. Studies such as this ratify an even higher level of assurance about the impending role of dendrimers in nanomedicine.

REFERENCES AND NOTES

1. Menjoge, A. R.; Kannan, R. M.; Tomalia, D. A. Dendrimer-Based Drug and Imaging Conjugates: Design Considerations for Nanomedical Applications. *Drug Discovery Today* **2010**, *15*, 171–185.
2. Tomalia, D. A.; Fréchet, J. M. J. Discovery of Dendrimers and Dendritic Polymers: A Brief Historical Perspective. *J. Polym. Sci., Part A: Polym. Chem.* **2002**, *40*, 2719–2728.
3. Yang, H.; Lopina, S. T. Stealth Dendrimers for Antiarrhythmic Quinidine Delivery. *J. Mater. Sci. Mater. Med.* **2007**, *18*, 2061–2065.
4. Fruchon, S.; Poupot, M.; Martinet, L.; Turrin, C. O.; Majoral, J. P.; Fournié, J. J.; Caminade, A. M.; Poupot, R. Anti-Inflammatory and Immunosuppressive Activation of Human Monocytes by a Bioactive Dendrimer. *J. Leukoc. Biol.* **2009**, *85*, 553–562.
5. Shaunak, S.; Thomas, S.; Gianasi, E.; Godwin, A.; Jones, E.; Teo, I.; Mireskandari, K.; Luthert, P.; Duncan, R.; Patterson, S.; *et al*. Polyvalent Dendrimer Glucosamine Conjugates Prevent Scar Tissue Formation. *Nat. Biotechnol.* **2004**, *22*, 977–984.
6. McCarthy, T. D.; Karellas, P.; Henderson, S. A.; Giannis, M.; O'Keefe, D. F.; Heery, G.; Paull, J. R.; Matthews, B. R.; Holan, G. Dendrimers as Drugs: Discovery and Preclinical and Clinical Development of Dendrimer-Based Microbicides for HIV and STI Prevention. *Mol. Pharm.* **2005**, *2*, 312–318.
7. Supattapone, S.; Nguyen, H. O.; Cohen, F. E.; Prusiner, S. B.; Scott, M. R. Elimination of Prions by Branched Polyamines and Implications for Therapeutics. *Proc. Natl. Acad. Sci. U.S.A.* **1999**, *96*, 14529–14534.
8. Hayder, M.; Poupot, M.; Baron, M.; Nigon, D.; Turrin, C. O.; Caminade, A. M.; Majoral, J. P.; Eisenberg, R. A.; Fournié, J. J.; Cantagrel, A.; *et al*. A Phosphorus-Based Dendrimer Targets Inflammation and Osteoclastogenesis in Experimental Arthritis. *Sci. Transl. Med.* **2011**, *3*, 81ra35.
9. Chauhan, A. S.; Diwan, P. V.; Jain, N. K.; Tomalia, D. A. Unexpected *In Vivo* Anti-Inflammatory Activity Observed for Simple, Surface Functionalized Poly(amidoamine) Dendrimers. *Biomacromolecules* **2009**, *10*, 1195–1202.
10. Dernedde, J.; Rausch, A.; Weinhart, M.; Enders, S.; Tauber, R.; Licha, K.; Schirner, M.; Zügel, U.; von Bonin, A.; Haag, R. Dendritic Polyglycerol Sulfates as Multivalent Inhibitors of Inflammation. *Proc. Natl. Acad. Sci. U.S.A.* **2010**, *107*, 19679–19684.
11. Poupot, M.; Griffe, L.; Marchand, P.; Maraval, A.; Rolland, O.; Martinet, L.; L'Faqihi-Olive, F. E.; Turrin, C. O.; Caminade, A. M.; Fournié, J. J.; *et al*. Design of Phosphorylated Dendritic Architectures to Promote Human Monocyte Activation. *FASEB J.* **2006**, *20*, 2339–2351.
12. Sato, K.; Takayanagi, H. Osteoclasts, Rheumatoid Arthritis, and Osteoimmunology. *Curr. Opin. Rheumatol.* **2006**, *18*, 419–426.
13. Takayanagi, H. Osteoimmunology: Shared Mechanisms and Crosstalk between the Immune and Bone Systems. *Nat. Rev. Immunol.* **2007**, *7*, 292–304.
14. McInnes, I. B.; Schett, G. Cytokines in the Pathogenesis of Rheumatoid Arthritis. *Nat. Rev. Immunol.* **2007**, *7*, 429–442.
15. Takayanagi, H. New Immune Connections in Osteoclast Formation. *Ann. N. Y. Acad. Sci.* **2010**, *1192*, 117–123.
16. Nakae, S.; Saijo, S.; Horai, R.; Sudo, K.; Mori, S.; Iwakura, Y. IL-17 Production

from Activated T Cells Is Required for the Spontaneous Development of Destructive Arthritis in Mice Deficient in IL-1 Receptor Antagonist. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 5986–5990.
17. Stoll, J. G.; Yasothan, U. Rheumatoid Arthritis Market. *Nat. Rev. Drug Discovery* **2009**, *8*, 693–694.
18. Klareskog, L.; Catrina, A. I.; Paget, S. Rheumatoid Arthritis. *Lancet* **2009**, *373*, 659–672.
19. Lipsky, P. E. Are New Agents Needed to Treat RA?. *Nat. Rev. Rheumatol.* **2009**, *5*, 521–522.
20. Schwarz, E. M.; Looney, R. J.; Drissi, M. H.; O'Keefe, R. J.; Boyce, B. F.; Xing, L.; Ritchlin, C. T. Autoimmunity and Bone. *Ann. N.Y. Acad. Sci.* **2006**, *1068*, 275–283.
21. Dalbeth, N.; Pool, B.; Smith, T.; Callon, K. E.; Lobo, M.; Taylor, W. J.; Jones, P. B.; Cornish, J.; McQueen, F. M. Circulating Mediators of Bone Remodeling in Psoriatic Arthritis: Implications for Disordered Osteoclastogenesis and Bone Erosion. *Arthritis Res. Ther.* **2010**, *12*, R164.
22. Szodoray, P; Alex, P.; Chappell-Woodward, C. M.; Madland, T. M.; Knowlton, N.; Dozmorov, I.; Zeher, M.; Jarvis, J. N.; Nakken, B.; Brun, J. G.; *et al*. Circulating Cytokines in Norwegian Patients with Psoriatic Arthritis Determined by a Multiplex Cytokine Array System. *Rheumatology (Oxford)* **2007**, *46*, 417–425.