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Therapeutic targets for osteoporosis

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CONTENTS

Abstract

Osteoporosis is the most common type of metabolic bone disease. It is characterized by low bone mass and deterioration of bone microarchitecture that leads to bone fragility and increased susceptibility to fractures. Generally, it increases progressively with age and decreased gonadal function and is the result of a negative skeletal balance observed when osteoclasts increase in number or activity, osteoblasts decrease in number or activity, or a combination of these factors. Most drugs marketed for the treatment of osteoporosis act by decreasing bone resorption or by increasing bone formation. The search for effective treatment strategies for osteoporosis continues, with special attention focused on the identification of novel targets for drug development. In the interest of facilitating access to information on the major targets for therapeutic intervention in osteoporosis, this article presents those targets that are currently under active investigation.

Introduction

Osteoporosis, the most common type of metabolic bone disease, is defined as a skeletal disorder characterized by low bone mass and deterioration of bone microarchitecture, leading to bone fragility and increased susceptibility to fractures, especially of the hip, spine and wrist, caused by relatively low impact (1, 2). It can be classified as a primary disorder (typically postmenopausal or senile osteoporosis) related to aging and decreased gonadal function, or as a secondary condition arising in patients taking glucocorticoids or other drugs or who suffer from hypogonadism, inflammatory bowel disease, cystic fibrosis, vitamin D deficiency, Cushing's syndrome, primary hyperparathyroidism or malignancies, transplant recipients and others (3, 4).

Bone is a porous, mineralized structure comprised of cells, vessels and hydroxyapatite crystals in varying proportions depending upon the type and location of the bone in the body. Throughout the normal human lifetime, old bone is removed by resorption and new bone is formed. Osteoclasts, a specialized class of bone-eroding cells derived from pluripotent hematopoietic cells, invade the bone's surface and create small cavities. After a period known as reversal, during which mononuclear cells appear on the bone surface, the cavitated bone is subsequently filled in by osteoblasts, or bone-forming cells. The entire process can take up to 6 months to be completed (5, 6). A series of molecular signals, some of which are only beginning to be understood, direct the processes of cell replication, differentiation, function, survival and death (5). When osteoclasts increase in number or activity, osteoblasts decrease in number or activity, or a combination thereof, the result is a negative skeletal balance. Although bone mineral density (BMD) is the most important and easily assessable factor determining bone strength and risk of fracture, another cluster of factors, together known as bone quality, is gaining increasing recognition for its contribution to bone strength (2). Bone quality encompasses the following variables: bone macroarchitecture (shape, geometry), bone microarchitecture (trabecular and cortical bone), matrix and mineral composition, degree of mineralization, accumulated microdamage and bone turnover rate.

Osteoporosis is a preventable disease. Furthermore, it is generally more difficult to restore lost bone in patients with established osteoporosis than it is to prevent bone loss. The most important components of prevention include calcium and vitamin D supplementation, as well as overall good nutrition, weight-bearing exercise, and reducing tobacco and alcohol consumption (7). Most drugs marketed for the treatment of osteoporosis (see Table I), *e.g.*, estrogens, bisphosphonates and calcitonin, act by decreasing bone resorption. Others, such as parathyroid hormone, act by increasing bone formation (8).

The search for effective treatment strategies for osteoporosis continues, with special attention focused on the identification of novel targets for drug development. Those targets which are currently under active investigation are discussed below (see Figure 1). Table I shows a selection of products under active development for each target.

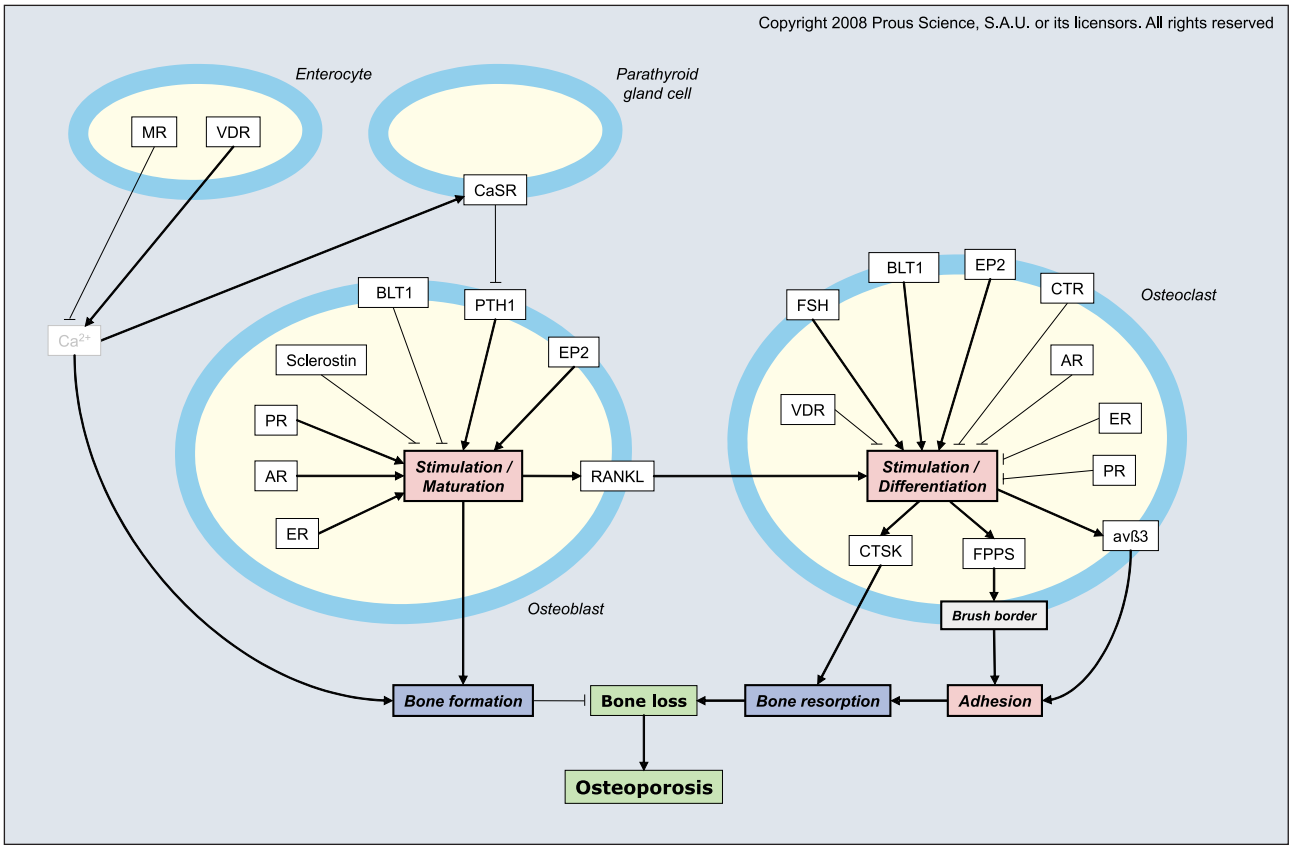

Fig. 1. Osteoporosis targetscape. Diagram showing an overall cellular and molecular landscape or comprehensive network of connections among the current therapeutic targets for the treatment of osteoporosis and their biological actions. Arrow, positive effect; dash, negative effect; αvβ3, integrin $\alpha_v\beta_3$ (vitronectin) receptor; AR, androgen receptor; BLT1, leukotriene B_4 receptor; CaSR, calcium-sensing receptor; CTR, calcitonin receptor; CTSK, cathepsin K; EP2, prostanoid EP_2 receptor; ER, estrogen receptor; FPPS, farnesyl pyrophosphate synthetase; FSH, follicle-stimulating hormone receptor; MR, mineralocorticoid receptor; PR, progesterone receptor; PTH1, parathyroid hormone receptor; RANKL, receptor activator of NF-κB ligand; VDR, vitamin D receptor.

Targets

Androgen receptor

The androgen receptor (AR) is the nuclear receptor that binds androgens (*e.g.,* testosterone, dihydrotestosterone, androstenedione), a class of sex hormones involved in the development and maintenance of the secondary male sex characteristics, sperm induction and sexual differentiation. Binding of these hormones to their receptor results in increased virility, libido and nitrogen and water retention, and stimulation of skeletal growth. When the androgen receptor is inactive, it is bound to heat shock proteins (HSPs) in the cytoplasm of prostate cells. After androgen binding, the receptor dissociates from HSPs and translocates to the nucleus, where it dimerizes and binds to the androgen response elements in the DNA, thereby activating genes involved in cell growth. Expression of ARs has been demonstrated in nearly all bone cells, including osteoblasts and osteoclasts. Stimulation of ARs has been described to exert proapoptotic effects on osteoclasts, whereas it prevents apoptosis of osteoblasts, thus modulating their lifespan and decreasing the number of bone remodeling cycles. Also, ARs appear to play a relevant role in the growth of periosteal cortical bone, although androgens stimulate both longitudinal and radial growth. It is interesting to note that both AR and estrogen receptor ERα pathways generate similar and additive effects on bone. In conclusion, AR agonists may exert a protective effect against bone loss, including in osteoporosis (9, 10).

Calcitonin receptors

Calcitonin receptors are G protein-coupled receptors (GPCRs) for calcitonin that couple to a $G_{s\alpha}$. Calcitonin is a peptide hormone (32 amino acids) synthesized and secreted in response to high blood calcium levels by the parafollicular cells of the thyroid gland in mammals. It increases calcium and phosphate deposition in bone and reduces plasma calcium levels. The binding of calcitonin to calcitonin receptors leads to the activation of adenylyl cyclase, thereby favoring the production of cAMP in target cells. In general, elevated levels of cAMP oppose any cell stimulation response. Calcitonin receptors are found in

Table I: Selected targets and products launched or being actively investigated for osteoporosis (from Prous Science Integrity®).

Target	Product	Source	Phase
Androgen receptor	LGD-2941	Ligand/TAP	I
	ACP-105	ACADIA	Preclinical
	RAD-140	Radius	Preclinical
Calcitonin receptor	Calcitonin, salmon	Menarini/Novartis/Rottapharm	Launched
	Elcatonin	GlaxoSmithKline	L-1985
Calcium-sensing receptor	JTT-305	Japan Tobacco	II
	Ronacaleret HCl	GlaxoSmithKline/NPS Pharmaceuticals	II
Cathepsin K	Odanacatib	Merck & Co.	III
	ONO-5534	Ono	II
	MIV-701	Medivir	I
	VEL-0230	Velcura	I
Estrogen receptor	Raloxifene HCl	Lilly	L-1998
	Bazedoxifene acetate	Ligand/Wyeth Pharmaceuticals	Prereg.
	Lasofoxifene tartrate	Pfizer	Prereg.
	Arzoxifene HCl	Lilly	III
	Bazedoxifene/conjugated estrogens	Ligand/Wyeth Pharmaceuticals	III
	Estetrol	Pantarhei Bioscience	II
	CHF-4227	Chiesi	I
	RAD-1901	Radius	Preclinical
Estrogen receptor/progesterone receptor	Estradiol valerate/medroxyprogesterone acetate	Orion Pharma	L-1984
	Estradiol/dydrogesterone	Solvay	L-1995
	Norethindrone acetate/ethinyl estradiol	Warner Chilcott	L-2000
Farnesyl pyrophosphate synthetase	Risedronate sodium	sanofi-aventis	L-1999
Follicle-stimulating hormone receptor	ADX-68693	Addex Pharmaceuticals	Preclinical
Integrin $\alpha_v\beta_3$ (vitronectin) receptor	NVX-188	Novelix	Preclinical
Leukotriene B_4 receptor	DW-1350	Dong-Wha	II
Mineralocorticoid receptor	Drospirenone/estradiol	Bayer Schering Pharma	L-2003
Parathyroid hormone receptor	Teriparatide	Lilly	L-2002
	Recombinant human parathyroid hormone(1-84)	Nycomed	L-2006
	Teriparatide acetate	Asahi Kasei Pharma	III
	BA-058	Radius	II
	ZT-031	Zelos Therapeutics	II
	PTH-131A	GlaxoSmithKline/Unigene	I
Prostanoid EP_2 receptor	CP-533536	Pfizer	II
RANKL	Denosumab	Amgen/Daiichi Sankyo	III
	RANKL AutoVac vaccine	Pharmexa	Preclinical
Sclerostin	Sclerostin Ab	Amgen/UCB	I
Vitamin D receptor	Calcitriol	Fontus	L-1978
	Eldecalcitol	Chugai	III
	BXL-093	Bioxell	Preclinical

osteoclasts and their activation by calcitonin is associated with a decrease in the rate of bone resorption. In more detail, osteoclasts undergo a loss of the ruffled border, cessation of motility and pseudopodial and margin retraction, and inhibition of osteoclast secretion of proteolytic enzymes and the proton pump. Calcitonin receptor agonists such as calcitonin of different origin have been marketed as effective antiosteoporotic strategies, thus demonstrating the importance of this therapeutic target (11-13).

Calcium-sensing receptor

The calcium-sensing receptor (CaS, CaSR) is a 7-transmembrane-spanning GPCR that transduces signals mainly through the G_q, G_i and, to a lesser extent, $G_{12/13}$ pathways. Both G_i and G_q signaling pathways result in elevated cytosolic calcium levels and activated protein kinase C (PKC), key elements causing cell responses. However, the biological effects derived from CaS recep-

tors are considerably more complex and cannot be explained solely by the basic signaling pattern of GPCRs. Interaction with additional proteins, such as filamin, receptor activity-modifying proteins and potassium channels, is believed to better explain their behavior. CaS receptors are present in chief cells of the parathyroid gland, although they can be found in many tissues, and their major role is the regulation of endogenous parathyroid hormone (PTH) levels. Elevated concentrations of extracellular calcium activate CaS receptors and PTH secretion is inhibited, and subsequently the release of calcium and phosphorus from bone. CaS receptor antagonists may increase BMD and decrease the risk of fractures in osteoporosis (14, 15).

Cathepsin K

A lysosomal cysteine protease (EC 3.4.22.38), cathepsin K is expressed predominantly on osteoclasts, cleaves bone matrix proteins and is involved in the process of bone resorption and bone remodeling. Agents that stimulate the osteoclast to produce increased amounts of cathepsin K include receptor activator of NF-κB ligand (RANKL), tumor necrosis factor (TNF), retinoic acid, interleukin-1 (IL-1), peroxisome proliferator-activated receptors PPARδ and PPARβ, the transcription factor c-jun, MITF (microphthalmia-associated transcription factor) and extracellular matrix proteins. Synthesized cathepsin K is transported from the endoplasmic reticulum within lysosomes and other secretory vesicles and then released to the extracellular compartment, where it digests several components of bone matrix, including type I collagen, osteopontin and osteonectin. Animal models for evaluating bone-active compounds were assayed for this enzyme and potent inhibitors are being developed, especially newer classes of low-molecular-weight compounds with a high degree of selectivity for cathepsin K, as potential therapeutic agents to treat diseases associated with excess bone resorption such as osteoporosis (16-18).

Estrogen receptor

The estrogen receptor (ER) family includes two structurally similar although not identical receptors, ERα and ERβ, that mediate the effects of estrogens, natural or synthetic substances that have activity (*i.e.,* estrogenic) similar to that of the most potent naturally occurring estrogen 17β-estradiol. Estrogens are produced by the ovary, testis, placenta, adrenal cortex and by certain plants. Estrogen actions include stimulation of secondary sex characteristics, growth and maturation of long bones, and control of the menstrual cycle. Upon ligand binding, ER dissociates from its inactive binding to HSP90, is activated, undergoes conformational change, dimerizes and autophosphorylates. Subsequently, ER dimers translocate to the nucleus, where they bind to estrogen response elements that regulate promoter regions of various genes, which, in general, are responsible for cell growth, inhibition of apoptosis and promotion of angiogenesis. Expression of ER has been detected in osteoblasts and osteocytes. Stimulation of ER has been described to exert proapoptotic effects in osteoclasts, whereas it prevents apoptosis of osteoblasts, thus modulating their lifespan and decreasing the number of bone remodeling cycles. During skeletal remodeling, ERα is the crucial pathway for longitudinal bone growth, although both the ERα and AR stimulate periosteal growth. In conclusion, ERs can be considered appropriate targets to prevent bone loss in osteoporosis (9, 19, 20).

Farnesyl pyrophosphate synthetase

Farnesyl pyrophosphate synthetase, or geranyltranstransferase, is the enzyme responsible for the synthesis of the linear isoprenoid farnesyl pyrophosphate (FPP) in the HMG-CoA reductase pathway, a precursor of steroids, cholesterol, sesquiterpenes, farnesylated proteins, heme and vitamin K_2. Isoprenoids such as FPP and geranylgeranyl pyrophosphate (GGPP) are required for the posttranslational modification of the small GTPases (*e.g.*, Ras, Rho and Rac), which are essential for the bone-resorbing activity and survival of osteoclasts. There are two types of posttranslational modification: farnesylation and geranylgeranylation, both affecting G protein-dependent cellular activation and numerous signaling pathways. Rho protein plays a key role in the organization of the osteoclast cytoskeleton, which is required for the configuration of the membrane brush border that allows osteoclasts to attach to bone and form the sealed resorption lacuna. Activation of Rho requires the attachment of geranylgeraniol, necessary for the translocation of inactive Rho from the cytosol to the membrane. Activation of Rho results in contractile forces and reorganization of the actin cytoskeleton, osteoclast shape change and formation of the ruffled border. Farnesyl pyrophosphate synthetase is a therapeutic target for well-known antiosteoporotic agents such as N-containing bisphosphonates (N-BPs). N-BPs interfere with the biosynthetic mevalonate pathway by inhibiting farnesyl pyrophosphate synthetase, thus blocking an essential step for Rho activation. Osteoclasts cannot form the tight-sealing zone required for bone resorption and bone loss is then stopped (17, 21-23).

Follicle-stimulating hormone receptor

The follicle-stimulating hormone receptor (FSH) is a member of the glycoprotein hormone receptor family that binds FSH, which is synthesized and secreted from the anterior pituitary gland in response to luteinizing hormone-releasing hormone (LHRH). FSH is a typical member of the GPCR family. In ovarian cells, FSH has been described to couple with a $G_{s\alpha}$, leading to stimulation of adenylyl cyclase, production of cyclic AMP (cAMP) and activation of PKA, which phosphorylates structural proteins, enzymes and transcriptional activators. In women, during the first half of the menstrual cycle, FSH stimulates

the production of graafian follicles in the ovary and is involved in subsequent oocyte maturation and the secretion of estrogen. In men, FSH stimulates the epithelium of the seminiferous tubules and is involved in the induction of spermatogenesis. However, recent studies demonstrate that osteoclasts also express FSH, which couples with $G_{i\alpha}$ instead of $G_{s\alpha}$. G_i activation is linked to the stimulation of the MEK/ERK, NF-κB and Akt pathways, resulting in enhanced osteoclast formation and function, and in consequence, increased bone loss. During early menopause and in hypogonadism, bone loss is observed, which may result in part from elevated levels of circulating FSH. All these observations suggest that the modulation of FSH may be an interesting strategy for the treatment of postmenopausal and hypogonadal osteoporosis (24-26).

Integrin $\alpha_v\beta_3$ (vitronectin) receptor

Integrins are a large family of widely expressed transmembrane receptors for extracellular matrix (ECM) and plasma proteins. Like other integrins, $\alpha_v\beta_3$ is comprised of two noncovalently linked subunits (α_v and β_3) that span the plasma membrane and interact with the actin cytoskeleton and focal adhesion kinase (FAK) complex through their cytoplasmic domains, which are highly conserved and provide specificity of interactions and fine tuning of subsequent downstream signaling. A characteristic feature of all integrins, including $\alpha_v\beta_3$, is the ability to transmit signals bidirectionally, both inside-out and outside-in, regulating the processes of cell survival, growth and motility. Integrin $\alpha_v\beta_3$ interacts with various ECM proteins, including fibronectin, osteoponin, tenascin and vitronectin. When the mature osteoclast adheres to bone, a tight-sealing zone between the cell and the bone surface, which is an extracellular compartment called the resorption lacuna, forms after integrin $\alpha_v\beta_3$ binds to ECM proteins. In the resorption lacuna, the collagen bone matrix is enzymatically digested and calcium hydroxyapatite is dissolved under acid conditions, leading to bone resorption. As a strategy against osteoporosis, antagonizing integrin $\alpha_v\beta_3$ would prevent the attachment of osteoclasts to bone and thus reduce bone resorption (23, 27).

Leukotriene B_4 receptor

The leukotriene B_4 receptor, or BLT_1, is a GPCR for LTB_4 that signals via the IP_3 second messenger (phospholipase C [PLC]-activating). It plays a role in inflammation and immunity. Leukotrienes are physiologically active substances that mediate inflammation and allergic reactions and have been implicated in inflammation-related diseases. The leukotrienes are 5-lipoxygenase metabolites of arachidonic acid and differ from prostaglandins and thromboxanes in that they do not have a central ring. Six metabolites have been named –LTA-LTF– with the number of double bonds indicated as a subscript (*e.g.,* LTB_4). They are classified into two subclasses: peptidoleukotrienes (*e.g.*, LTC_4, LTD_4, LTE_4) and hydroxyleukotrienes (LTB_4). LTB_4 is involved in immune responses such as stimulation of leukocytes (*i.e.*, chemotaxis, aggregation, etc.). High levels of LTB_4 are observed in rheumatoid arthritis, gout, psoriasis and inflammatory bowel disease. Curiously, a negative bone remodeling balance has been observed in patients with inflammation-related diseases in which LTB_4 plays a significant role. Studies have shown LTB_4 to increase bone resorption both *in vitro* and *in vivo*, probably by stimulating the recruitment, formation and activation of osteoclasts, as well as by inhibiting the differentiated function of the osteoblasts. Thus, inhibition of the BLT_1 receptor may be an attractive strategy for the treatment of osteoporosis (28-30).

Mineralocorticoid receptor

The mineralocorticoid receptor (MR) is a nuclear receptor of the NR3 class also known as the aldosterone receptor, that exists as a dimer coupled with chaperone molecules (*e.g.,* HSP90, HSP70). It binds corticosteroid, cortisol, aldosterone and progesterone with comparable affinity. Upon binding, the receptor–ligand complex dissociates from HSP and becomes activated, dimerizes and translocates to the nucleus, where it interacts with hormone response elements in the promoter regions of target genes involved in the regulation of electrolyte and water balance, modulating their transcription rate. Hence, the receptor affects ion transport in epithelial cells and renal tubules, causing retention of sodium and loss of potassium. Based on its role in water balance, MR modulators are being investigated for the treatment of hypertension, heart failure and Cushing's syndrome. Interestingly, it has been observed that osteoporosis is a characteristic clinical feature in patients with Cushing's syndrome or in those on glucocorticoid treatment. Moreover, glucocorticoids have been described to increase bone resorption, inhibit bone formation and have an indirect action on bone by decreasing intestinal calcium absorption, but also inducing a sustained renal calcium excretion. Thus, the MR is an attractive target for the prevention and treatment of glucocorticoid-induced osteoporosis (31, 32).

Parathyroid hormone receptor PTH1

PTH1 is a GPCR for PTH that couples to a $G_{s\alpha}$. PTH is a peptide produced by the parathyroid gland that regulates calcium and phosphorus metabolism, with resulting anabolic effects in bone. PTH1 also binds parathyroid hormone-related hormone (PTHrH). PTH raises serum salt levels by dissolving the salts in bone and preventing their renal excretion. Although excess PTH can be deleterious for bone formation, it has been reported that intermittent exposure of bone to PTH can increase bone mass. PTH1 is expressed in mature osteoblasts and PTH binding leads to an antiapoptotic signaling that involves stimulation of adenylyl cyclase, production of cAMP and activation of PKA, which phosphorylates structural proteins, enzymes and transcriptional activators. Studies in mice have demonstrated that PTH treatment inhibits

osteoblast apoptosis, leading to an increase in osteoblast numbers and a subsequent increase in bone formation. PTH also seems to increase osteoblast differentiation and to stimulate osteoclast-released factors that activate osteoblasts. PTH1 agonists such as recombinant PTH and PTH analogues are currently under active development for the treatment of osteoporosis (33, 34).

Progesterone receptor

A nuclear receptor of the NR3 class that exists as a dimer coupled with chaperone molecules (*e.g.*, HSP90, HSP70), the PR exists as two separate isoforms (A and B) and binds the steroid progesterone (4-pregnene-3,20-dione), a naturally occurring antiestrogenic steroid produced by the corpus luteum and placenta. Chaperone molecules are shed subsequent to ligand binding, PR undergoes a conformational change, dimerizes and becomes activated. Subsequently, PR dimers translocate to the nucleus, where they bind to progesterone response elements that regulate promoter regions of various genes associated with the preparation of the uterus for implantation and growth of fertilized ovum, as well as the maintenance of pregnancy by inhibiting uterine contractions during gestation. PRs have been identified in primary cultures of human osteoblasts and osteoclasts and it has been indicated that progesterone appears to act directly on bone remodeling, playing a role in the coupling of bone resorption with bone formation. However, it is not clear whether these activities can be ascribed to its progestational or other intrinsic hormonal actions. It is important to point out that progesterone acts synergistically with estrogen on the skeleton. PR modulation, problably with additional ER modulation, may be an effective treatment for osteoporosis and postmenopausal syndrome (35, 36).

Prostanoid EP_2 receptor

The prostanoid EP_2 receptor is a GPCR that mediates the actions of prostaglandin E_2 (PGE_2) and is characterized by a compact structure when compared to other prostanoid receptors (EP_1, EP_3 and EP_4). EP_2 receptors mainly couple to G_s and mediate elevations in cAMP concentration, although they participate in other pathways as well. In the skeleton, PGE_2 effects have been linked to EP_2 and/or EP_4 receptor subtypes. PGE_2 has been found to stimulate both bone formation and resorption *in vitro* and *in vivo*. For example, both receptors induce PGE_2-mediated RANKL expression in osteoblasts through cAMP, thus inducing osteoclast maturation and bone loss. Conversely, PGE_2-stimulated bone formation most likely results from recruitment of osteoblasts from their bone marrow stromal precursors. The balance of this dual action of PGE_2 seems to be biased towards favoring bone formation because when PGE_2 is administered systemically or locally to the skeleton in a rat model it causes significant increases in bone mass and strength. However, due to its side effects, PGE_2 administration is not an appropriate therapy. An alternative therapeutic approach that is being considered is the development of EP_2 receptor agonists able to mimic the effects of PGE_2 as a treatment for osteoporosis and bone loss (37-39).

RANKL

A member of the TNF family of cytokines that is produced by cells of osteoblastic lineage, RANKL is a crucial modulator of bone remodeling and required for the development and activation of osteoclasts. RANK is present in osteoclast progenitors and mature osteoclasts. RANKL expression on the plasma membrane of osteoblasts is induced by bone-resorbing factors such as vitamin D_3, PTH, PGE_2, IL-1 and -11, TNF-α and glucocorticoids. RANKL binding to RANK induces receptor trimerization and activation that results in the recruitment of several adaptor proteins, which trigger the activation of downstream signaling pathways such as p38 MAPK/ MK2, NF-κB, Jun kinase 1 (JNK1) and c-Src/PI3K/Akt. Signaling ends with the activation of transcription factors that translocate to the nucleus and induce the expression of genes that regulate activation, survival and differentiation of osteoclasts. Based on its key role as a regulator of osteoclast development, RANKL is a highly attractive target for the treatment of osteolytic bone disorders such as osteoporosis and rheumatoid arthritis (40-42).

Sclerostin

Sclerostin is a bone morphogenetic protein (BMP) antagonist that inhibits the differentiation of osteoprogenitor cells and reduces osteoblast activity. Sclerostin is the product of the *SOST* gene that is produced in osteocytes buried in the bone and is a powerful inhibitor of bone formation. As osteoblasts become embedded in the mineralized matrix, they transform into osteocytes and begin expressing sclerostin, which controls bone formation and phosphate metabolism. Runt-related transcription factor 2 (RUNX2) controls its expression throughout osteoblast maturation. Sclerostin inhibits Wnt signaling, a complex network of proteins that has been described to play a role in embryogenesis and cell growth and proliferation, and is also involved in controlling physiological processes such as osteoblast differentiation. Wnt protein is a ligand for the frizzled receptor (FZD), whose coreceptor is the low-density lipoprotein receptor-related protein 5 (LRP5). Ligand binding induces signal transduction that leads to stabilization of β-catenin, the accumulation of which promotes transcriptional events that increase osteoblast number and activity, and ultimately bone formation. Sclerostin antagonizes this signaling by inducing the dissociation of LRP5 from FZD and Wnt. Current therapeutic approaches to treat bone loss include neutralization of sclerostin by antibodies and drugs (33, 43, 44).

Vitamin D receptor

The vitamin D receptor, or VDR, also known as the calcitriol receptor, belongs to the superfamily of

steroid/thyroid hormone nuclear receptors. It is activated by vitamin D (1,25-dihydroxyvitamin D), a general name for any steroid with activity similar to vitamin D_2 or D_3. Upon activation by vitamin D, VDR heterodimerizes with the retinoid X receptor and binds to hormone response elements on DNA, resulting in the expression of specific genes and products that regulate calcium and phosphorus absorption from the bowel, promote reabsorption of calcium in the kidneys and mediate the mineralization of osteoid tissue. Calcium provides mechanical strength to the skeleton, and bone is the main reservoir of the body's calcium, accounting for over 99%. Hence, the importance of vitamin D in maintaining blood calcium levels and preventing fractures and bone loss has been widely recognized. Moreover, vitamin D, acting through the VDR, has been described to suppress bone resorption through a direct antiosteoclastic action, by interfering with signaling through RANK receptors on osteoclast precursor cells. Although vitamin D analogues and VDR agonists have long been used for the oral treatment of hypocalcemia and resulting metabolic bone disease, new products targeting VDR are currently under active development for the treatment of osteoporosis (45, 46).

References

1. Lane, N.E., Kelman, A. *A review of anabolic therapies for osteoporosis.* Arthritis Res Ther 2003, 5(5): 214-22.

2. Sambrook, P., Cooper, C. *Osteoporosis.* Lancet 2006, 367(9527): 2010-8.

3. Painter, S.E., Kleerekoper, M., Camacho, P.M. *Secondary osteoporosis: A review of the recent evidence.* Endocr Pract 2006, 12(4): 436-45.

4. Pfeilschifter, J., Diel, I.J. *Osteoporosis due to cancer treatment: Pathogenesis and management.* J Clin Oncol 2000, 18(7): 1570-93.

5. Canalis, E., Giustina, A., Bilezikian, J.P. *Mechanisms of anabolic therapies for osteoporosis.* N Engl J Med 2007, 357(9): 905-16.

6. Hadjidakis, D.J., Androulakis, I.I. *Bone remodeling.* Ann NY Acad Sci 2006, 1092: 385-96.

7. Keen, R. *Osteoporosis: Strategies for prevention and management.* Best Pract Res Clin Rheumatol 2007, 21(1): 109-22.

8. Kirk, D., Fish, S.A. *Medical management of osteoporosis.* Am J Manag Care 2004, 10(7, Pt. 1): 445-55.

9. Manolagas, S.C., Kousteni, S., Jilka, R.L. *Sex steroids and bone.* Recent Prog Horm Res 2002, 57: 385-409.

10. Vanderschueren, D., Vandenput, L., Boonen, S., Lindberg, M.K., Bouillon, R., Ohlsson, C. *Androgens and bone.* Endocr Rev 2004, 25(3): 389-425.

11. Purdue, B.W., Tilakaratne, N., Sexton, P.M. *Molecular pharmacology of the calcitonin receptor.* Receptors Channels 2002, 8(3-4): 243-55.

12. Stepan, J.J., Alenfeld, F., Boivin, G., Feyen, J.H., Lakatos, P. *Mechanisms of action of antiresorptive therapies of postmenopausal osteoporosis.* Endocr Regul 2003, 37(4): 225-38.

13. Poyner, D.R., Sexton, P.M., Marshall, I. et al. *International Union of Pharmacology. XXXII. The mammalian calcitonin gene-related peptides, adrenomedullin, amylin, and calcitonin receptors.* Pharmacol Rev 2002, 54(2): 233-46.

14. Huang, C., Miller, R.T. *The calcium-sensing receptor and its interacting proteins.* J Cell Mol Med 2007, 11(5): 923-34.

15. Rodriguez, M., Nemeth, E., Martin, D. *The calcium-sensing receptor: A key factor in the pathogenesis of secondary hyperparathyroidism.* Am J Physiol Renal Physiol 2005, 288(2): F253-64.

16. Stoch, S.A., Wagner, J.A. *Cathepsin K inhibitors: A novel target for osteoporosis therapy.* Clin Pharmacol Ther 2008, 83(1): 172-6.

17. Lee, S.W. et al. *Participation of protein kinase C beta in osteoclast differentiation and function.* Bone 2003, 32(3): 217-27.

18. Selent, J., Kaleta, J., Li, Z., Lalmanach, G., Brömme, D. *Selective inhibition of the collagenase activity of cathepsin K.* J Biol Chem 2007, 282(22): 16492-501.

19. Osborne, C.K., Schiff, R. *Estrogen-receptor biology: Continuing progress and therapeutic implications.* J Clin Oncol 2005, 23(8): 1616-22.

20. Ohmichi, M., Tasaka, K., Kurachi, H., Murata, Y. *Molecular mechanism of action of selective estrogen receptor modulator in target tissues.* Endocr J 2005, 52(2): 161-7.

21. Liang, P.H., Ko, T.P., Wang, A.H. *Structure, mechanism and function of prenyltransferases.* Eur J Biochem 2002, 269(14): 3339-54.

22. Green, J.R. *Bisphosphonates: Preclinical review.* Oncologist 2004, 9(Suppl. 4): 3-13.

23. Faccio, R., Novack, D.V., Zallone, A, Ross, F.P., Teitelbaum, S.L. *Dynamic changes in the osteoclast cytoskeleton in response to growth factors and cell attachment are controlled by beta3 integrin.* J Cell Biol 2003, 162(3): 499-509.

24. Sun, L., Peng, Y., Sharrow, A.C. et al. *FSH directly regulates bone mass.* Cell 2006, 125(2): 247-60.

25. Ulloa-Aguirre, A., Timossi, C. *Structure-function relationship of follicle-stimulating hormone and its receptor.* Hum Reprod Update 1998, 4(3): 260-83.

26. Simoni, M., Gromoll, J., Nieschlag, E. *The follicle-stimulating hormone receptor: Biochemistry, molecular biology, physiology, and pathophysiology.* Endocr Rev 1997, 18(6): 739-73.

27. Mostafavi-Pour, Z., Askari, J.A., Parkinson, S.J., Parker, P.J., Ng, T.T., Humphries, M.J. *Integrin-specific signaling pathways controlling focal adhesion formation and cell migration.* J Cell Biol 2003, 161(1): 155-67.

28. Bos, C.L., Richel, D.J., Ritsema, T., Peppelenbosch, M.P. *Prostanoids and prostanoid receptors in signal transduction.* Int J Biochem Cell Biol 2004, 36(7): 1187-205.

29. Traianedes, K., Dallas, M.R., Garrett, I.R., Mundy, G.R., Bonewald, L.F. *5-Lipoxygenase metabolites inhibit bone formation in vitro.* Endocrinology 1998, 139(7): 3178-84.

30. Garcia, C., Boyce, B.F., Gilles, J., Dallas, M., Qiao, M., Mundy, G.R., Bonewald, L.F. *Leukotriene B4 stimulates osteoclastic bone resorption both in vitro and in vivo.* J Bone Miner Res 1996, 11(11): 1619-27.

31. Fuller, P.J., Young, M.J. *Mechanisms of mineralocorticoid action.* Hypertension 2005, 46(6): 1227-35.

32. Ferrari, P. *Cortisol and the renal handling of electrolytes: Role in glucocorticoid-induced hypertension and bone disease.* Best Pract Res Clin Endocrinol Metab 2003, 17(4): 575-89.

33. Khosla, S., Westendorf, J.J., Oursler, M.J. *Building bone to reverse osteoporosis and repair fractures.* J Clin Invest 2008, 118(2): 421-8.

34. Cranney, A., Papaioannou, A., Zytaruk, N. et al. *Clinical Guidelines Committee of Osteoporosis Canada. Parathyroid hormone for the treatment of osteoporosis: A systematic review.* CMAJ 2006, 175(1): 52-9.

35. Balasch, J. *Sex steroids and bone: Current perspectives.* Hum Reprod Update 2003, 9(3): 207-22.

36. Chabbert-Buffet, N., Meduri, G., Bouchard, P., Spitz, I.M. *Selective progesterone receptor modulators and progesterone antagonists: Mechanisms of action and clinical applications.* Hum Reprod Update 2005, 11(3): 293-307.

37. Sugimoto, Y., Narumiya, S. *Prostaglandin E receptors.* J Biol Chem 2007, 282(16): 11613-7

38. Paralkar, V.M., Borovecki, F., Ke, H.Z. et al. *An EP2 receptor-selective prostaglandin E2 agonist induces bone healing.* Proc Natl Acad Sci USA 2003, 100(11): 6736-40.

39. Ushikubi, F., Sugimoto, Y., Ichikawa, A., Narumiya, S. *Roles of prostanoids revealed from studies using mice lacking specific prostanoid receptors.* Jpn J Pharmacol 2000, 83(4): 279-85.

40. Rauner, M., Sipos, W., Pietschmann, P. *Osteoimmunology.* Int Arch Allergy Immunol 2007, 143(1): 31-48.

41. Blair, J.M., Zheng, Y., Dunstan, C.R. *RANK ligand.* Int J Biochem Cell Biol 2007, 39(6): 1077-81.

42. Kiechl, S., Werner, P., Knoflach, M., Furtner, M., Willeit, J., Schett, G. *The osteoprotegerin/RANK/RANKL system: A bone key to vascular disease.* Expert Rev Cardiovasc Ther 2006, 4(6): 801-11.

43. Martin, T.J., Sims, N.A., Ng, K.W. *Regulatory pathways revealing new approaches to the development of anabolic drugs for osteoporosis.* Osteoporos Int 2008, 19(8): 1125-38.

44. Shoback, D. *Update in osteoporosis and metabolic bone disorders.* J Clin Endocrinol Metab 2007, 92(3): 747-53.

45. Francis, R.M., Anderson, F.H., Patel, S., Sahota, O., van Staa, T.P. *Calcium and vitamin D in the prevention of osteoporotic fractures.* QJM 2006, 99(6): 355-63.

46. Ikeda, K. *Vitamin D, osteoclastogenesis and bone resorption: From mechanistic insight to the development of new analogs.* Endocr J 2007, 54(1): 1-6.