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The Ongoing Saga of Osteoporosis Treatment

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SCENARIO

Over the past 15 years or so, there has been a surge in interest about osteoporosis. Evidence for this can been seen in newspapers, magazines, books, and television talk shows, or heard on the radio. The scientific community has responded to this increased awareness. For example, a growing number of scientists are attending conferences related to osteoporosis or other aspects of bone biology. With an aging population fortified by the "baby-boomers," there is a growing citizenry who want to maintain their skeletal integrity in order to remain active. The pharmaceutical industry has also responded to this scenario and is accelerating research in this area in order to deliver an increasing number of safe and effective therapies. The market for osteoporosis related drugs has grown remarkably from just under $1.0 billion several years ago to over $2.5 billion in 1998. Moreover, this market is projected to exceed $8.5 billion by $2005 and $12.0 billion by 2010. Thus, two major driving forces are in place to fuel the expansion of osteoporosis-related research in the pharmaceutical industry: a significant unmet medical need and business opportunity.

POTENTIAL TARGETS

Bone is in a continuous state of flux. It has remarkable capacities to respond to both internal and external signals in order to maintain its integrity, as well as to react to metabolic demands like the maintenance of constant serum calcium levels. The simplest way to describe this process of "bone remodeling" is to state that the activities of the osteoblast (or bone-forming cell) and the osteoclast (or bone resorbing cell) are "coupled" to each other and therefore kept in balance [Silverberg and Lindsay, 1987]. We know that bone remodeling is an ongoing process and results in a continuous resorption of mineralized matrix with the subsequent replacement of lost bone at numerous skeletal sites. Under normal physiological conditions, the amount of new calcified matrix that is produced by the osteoblasts is equal to the amount of bone that is resorbed by the osteoclast. However, at a point after peak bone mass has been achieved, and this temporal location is debatable but is thought to occur sometime during the third or fourth decades of life, formation begins to lag behind resorption and this results in a net loss of bone mass. In fact, as an individual proceeds past middle age, the amount of bone mass that is lost averages approximately 0.5–1.0% of the total skeletal mass per year [Dempster and Lindsay, 1993]. Thus, the peak bone mass that an individual achieves in the second or third decade of life is a major determinant for the subsequent development of osteopenia and osteoporosis in the sixth or seventh decade of life.

Many factors appear to have an effect on the attainment of peak bone mass and its subsequent maintenance: these include race, family history, hormonal status, exercise, alcohol consumption, smoking and corticosteroid use [Eastell, 1998]. Postmenopausal osteopenia and osteoporosis have received a great deal of attention over the past decade, even though it was first described almost 60 years ago by Fuller Albright in 1941 [Albright et al., 1941]. Unlike men, whose peak bone mass is higher and whose overall rate of bone loss is slower, women attain a lower peak bone mass and lose bone mass at an accelerated rate—especially during the first 5 years after menopause as a result of estrogen insufficiency. This reduction in bone mass may eventually lead to a point where the fracture threshold at weight-bearing skeletal sites is exceeded, which results in an increased frac-

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ture risk. Postmenopausal women have become a focus of the pharmaceutical industry due to the rising medical costs associated with the treatment of fractures in this rapidly enlarging population. This type of osteoporosis (type I) is associated primarily with ovarian (or testicular) insufficiency and results in increased bone resorption. Type I osteoporosis differs from type II (or senile) osteoporosis, since the later is due primarily to a reduction in bone formation capacity as a result of decreased osteoblast number and/or activity. Type II osteoporosis is a consequence of the aging process, where a small but continuous loss of mass occurs over time and may eventually result in a fracture [Riggs and Melton, 1986].

The underlying mechanisms associated with alterations in bone metabolism with age remain unknown. Besides the obvious notion that it is a natural part of the aging process, we do not fully understand the physiologic process associated with the change resulting in bone mass loss. Numerous studies have been performed preclinically and in the clinic in an effort to begin to understand the etiology of osteoporosis. From the perspective of the pharmaceutical industry, the complexity of bone physiology makes it essentially impossible to identify a single receptor, enzyme or hormone as a target to regulate the bone remodeling process. Instead, this complexity provides an abundance of potential targets. Without question, the selection of safe and efficacious drug targets is the primary challenge with which the industry is faced today.

In choosing a potential drug target for osteoporosis, a number of questions must be considered. For example, where in the scheme of bone remodeling should drug intervention be targeted, and what is the rationale for this decision? The available information on the regulation of osteoblast and osteoclast function is massive. Name a hormone, growth factor, or cytokine and chances are that it most likely has already been described as playing some function in bone physiology. Moreover, unlike the reproductive system, where hormonal changes result in rapid and dramatic modifications on a monthly basis, the effects of hormones on the skeleton are more subtle and require much longer periods of time. The delicacy of these responses on the bone remodeling process is certainly appropriate from a biological point of view, since extreme fluctuations would generate unwarranted alterations in skeletal homeostasis. However, these subtleties also create a difficult problem to surmount when attempting to select a pharmacologic target to modulate.

WHAT IS NEEDED? WHAT IS KNOWN? WHAT IS OBVIOUS?

What is needed for the treatment of osteoporosis is not a particularly difficult concept to understand. Therapies must either prevent the loss of bone in the case of antiresorbers or, if this loss has already occurred, they need to be able to stimulate new bone formation which occurs with osteogenic or anabolic agents. With everything that has been learned about the function of the osteoblast and osteoclast, along with their bone marrow precursors, this does not seem like it should be a difficult task. Yet, with all the therapies that are currently available, none of them completely solves the problem. So the question becomes, what is the situation that requires pharmacologic intervention? As we age, our bones begin to lose mass, and at some point, if enough mass is lost, they fracture. Fractures occur most commonly at the wrist, hip or spine, with the later two resulting in significant medical costs and, in fact, alarmingly high rates of morbidity and mortality [Peck et al., 1988]. The type of treatment that is appropriate, depends upon when a patient enters therapy and, perhaps, the patient's age. If fractures have already occurred, then there is a need to increase bone mass substantially with an osteogenic agent in order to move above the fracture threshold line. However, once formed, this new bone then needs to be maintained with an antiresorptive agent. The type of antiresorptive agent becomes an issue (ERT vs bisphosphonate), and patient-by-patient decisions have to be made. If a patient is considered to have normal bone mass, some type of preventive therapy, such as treatment with an antiresorber, would be in order. But another question that needs to be addressed is whether a patient with apparently normal skeletal mass should be treated first with an osteogenic agent for a brief period and then placed on an antiresorptive therapy. This treatment regimen would increase the "comfort zone" by distancing a patient's bone mass further above the fracture threshold line.

The roles, or at least the therapeutic effects, of several hormones have been well characterized. Vitamin D, calcitonin, estrogens, andro-

gens, and parathyroid hormone (PTH) are all known to affect skeletal metabolism in humans [Chapuy and Meunier, 1995]. The precise mechanism of action in bone for any one of these agents has not been entirely elucidated. As is often the case, the basic science (molecular mechanisms) lags behind the therapy.

It is known that the active metabolite of vitamin D, $1,25(OH)_2D_3$, affects calcium absorption in the gut, osteoclast differentiation, and bone resorption, as well as osteoblast differentiation and bone formation, yet the efficacy of this as a treatment for osteoporosis is marginal at best [Ott and Chestnut, 1989; Tilyard et al., 1992]. However, vitamin D is a proven supplement, especially in the aging population, in which calcium resorption becomes less efficient. The problem with vitamin D as a therapy for osteoporosis is the potential hypercalcemic effect. There is an ongoing effort to identify vitamin D analogues that may demonstrate tissue selectivity [Norman et al., 1993]. That is, analogues that don't increase calcium absorption (and perhaps also bone resorption), but do exhibit a beneficial effect on bone mineral density (i.e., increase bone formation). The concept of tissue selectivity is becoming increasingly accepted in the steroid/thyroid hormone receptor field. For example, some vitamin D analogues are purported to be anabolic towards the skeleton [Hansen and Maenpaa, 1997], which would be consistent with their positive effect on bone alkaline phosphatase and perhaps osteocalcin expression as well [Staal et al., 1998; Ducy et al., 1996].

While vitamin D therapy is not commonly used as a treatment for osteoporosis, another calciotropic hormone—calcitonin—is prescribed for this disease. Like vitamin D, the use of calcitonin to regulate bone remodeling is obvious. Calcitonin has been shown to have antiresorptive activity, both in vitro on isolated osteoclasts [Fenton et al., 1993] and in vivo [Overgaard et al., 1992]. Calcitonin is available in either a subcutaneous injectable form or in a nasal spray. Both are effective in reducing bone loss, especially in Paget's disease which shows an accelerated rate of resorption, however resistance has been reported after relatively short treatment periods [Singer et al., 1980]. Additionally, a beneficial side benefit of calcitonin use is its analgesic properties. While the mechanism for the side effect is unknown, it clearly provides fracture-associated pain relief and the onset is rapid. The improvement in bone mineral density (BMD) with calcitonin is considered moderate (1–2% increase in BMD), and some reports suggest a limited timeframe of effectiveness. The development of calcitonin mimetics or molecules that stimulate calcitonin synthesis and secretion have not yet been successful. An orally active small molecule that activates calcitonin pathways would most likely be an attractive pharmaceutical; however, the development of such molecules, if occurring, is a well-kept secret.

There is a great deal of ongoing effort in the characterization of the effects of estrogens and estrogen mimetics on the skeleton. It is quite clear that the sudden reduction in circulating estrogens at menopause results in a corresponding rapid loss of bone mass and a subsequent increase in fracture incidence. Estrogen replacement therapy (ERT), which includes Premarin (Wyeth-Ayerst, Radnor, PA), Ogen Estraderm (Pharmacia-Upjohn, Kalamazoo, MI), and 17β-estradiol, has been demonstrated to effectively reduce further loss of skeletal mass and are considered to be first-line therapies for the treatment of postmenopausal osteoporosis. However, ERT does not restore bone mass (i.e., estrogens are not osteogenic). What they appear to accomplish is the reestablishment of the normal balanced relationship between osteoblastic bone formation and osteoclastic bone resorption.

While estrogens certainly protect the skeleton, there are associated negative or unwanted side effects of unopposed ERT that are primarily associated with uterine hyperplasia and bleeding. The hyperplastic response to estrogens can be effectively reduced by combining estrogens with a progestin (hormone replacement therapy [HRT]); however, this does not entirely eliminate uterine bleeding. Unfortunately, there are additional side effects associated with progestin use, such as changes in mood (e.g., anxiety, depression) and water retention.

Considering the pros and cons of ERT/HRT, as well as the evolving understanding of estrogen receptor (ER) pharmacology, it has become reasonable to predict that one compound working through the ER could display tissue selectivity in its signal transduction to estrogen target tissues [Katzenellenbogen et al., 1993]. The concept of tissue selective estrogens has been exemplified by tamoxifen (first generation gen-

eration, Nolvadex, Zeneca, Wilmington, DE), and more recently raloxifene (Evista, second generation, Eli Lilly, Indianapolis, IN), which has an improved profile over tamoxifen [Jordan, 1998]. Other compounds in this class have also begun to emerge: these include droloxifene (Pfizer, New York), idoxifene (SmithKline Beecham, Philadelphia, PA), levormeloxifene (Novo Nordisk, Princeton, NJ), CP-336156 (Pfizer/Ligand, La Jolla, CA), and TSE-424 (Wyeth-Ayerst/Ligand, Radnor, PA). As a group, these compounds are bone sparing with varying potency and efficacy on the skeleton. Perhaps equally as important in terms of estrogen-like effect on the skeleton is the beneficial impact of these compounds on serum cholesterol reduction, without the stimulation of the uterus and the breast. A few years ago, these compounds would have been classified as anti-estrogens, because of their ability to antagonize estrogen action in standard in vitro reporter gene assays (i.e., estrogen response element-luciferase constructs). However, this simple classification no longer holds for more complex in vitro or in vivo responses, and the precise molecular mechanism(s) for the tissue-selective actions of these compounds remains unknown. Only one tissue selective estrogen, raloxifene, is approved for the prevention of postmenopausal osteoporosis, and although the clinical results demonstrated that this compound does protect the skeleton, it was not as effective as Premarin. The skeletal response of the other tissue-selective estrogens remains to be determined clinically. However, since the precise molecular target(s) for estrogen/estrogen-mimetic action remain unclear, improving the efficacy of these compounds on the skeleton while retaining or reducing side effects will be difficult. Moreover, improving efficacy on bone as measured by BMD may not even be necessary. Fracture prevention data with raloxifene are demonstrating that even with the relatively modest effect of 60 mg of drug on BMD, there is a 40–50% reduction in fracture risk, which is close to the 60% reduction associated with Premarin.

Is there room for improvement for the tissue-selective estrogens? From the point of view of the skeletal, probably yes. However, it is also important to maintain additional perspectives as well. This class of drugs will eventually serve multiple purposes when used as ERT/HRT, and bone is only one target. The uterus, breast, cardiovascular system, and central nervous system (CNS) are perhaps equally as important, if not more so. Improved tissue selectivity is therefore one area in which the next generation of selective estrogens will have to improve upon. Raloxifene sets the standard upon which future compounds will be judged, and new drugs will not only have to spare the skeleton, but they will also have to be less uterotropic, increase HDL cholesterol, and either have no effect or reduce hot flushes. Is this improved profile possible? A few years ago, it would not have been thought that one compound could do what raloxifene does. Today, with new in vitro and in vivo models and greater insight into the mechanism of estrogen action, the development of the ultimate tissue selective compound may yet be possible.

Although ERT/HRT is the first-line therapy for the treatment of osteoporosis, many women are either not willing or not able to use this therapy for various reasons. Besides calcitonin and ERT/HRT, the other major antiresorption therapy that is available are the bisphosphonates. The mechanism of action for bisphosphonates appears to be primarily manifested via the inhibition of osteoclastic activity. The competition in this area is as fierce as it is with the tissue selective estrogens, even though only a few bisphosphonates are currently available for the prevention and treatment of osteoporosis in the United States (e.g., alendronate, Fosamax, Merck/Wyeth-Ayerst, West Point, PA) with a few more also available in Europe. The positive effect of these compounds on BMD is well documented. For example, alendronate increases BMD in up to 7% of postmenopausal women, which is somewhat higher than what is reported for Premarin or 17β-estradiol [Liberman et al., 1995]. The fracture prevention data for alendronate are also impressive with a 60–80% reduction in risk. Unlike the tissue-selective estrogens, the bisphosphonates do not provide other types of protective effects such as reduction in LDL cholesterol or the prevention of breast cancer (preliminary studies suggest that they do appear to reduce bone metastasis). On the other hand, no uterine, breast, or CNS side effects are associated with the bisphosphonates. The major pitfalls are low oral bioavailability and gastrointestinal (GI) side effects. The newer generation bisphosphonates are more potent, which may reduce GI problems. Can these compounds be improved? Besides an

increase in potency, the use of patches as well as periodic dosing are also being considered as potential areas for development of this class of drugs. Since these compounds reside in the bone matrix and remain there for long periods of time, it has been postulated that daily dosing may not be required data. It is also possible that the bisphosphonates may be used to deliver other compounds to bone. So, one might derive the antiresorptive function not only of the bisphosphonate, but perhaps of the effects of another drug as well. For example, one could envision delivering an osteogenic agent, perhaps a peptide that may be protected from metabolic conversion when attached to a bisphosphonate, that is released in the bone microenvironment to stimulate formation. As with other antiresorbers, replacing bone that has already been lost does not occur with bisphosphonates, and some means to restore lost bone is still required.

This does not exhaust the possibilities for resorption inhibitors. For example, there is ongoing research in the pharmaceutical industry to identify and develop vitronectin receptor (VNR) antagonists. It is hypothesized that osteoclasts attach themselves to the bone surface via vitronectin receptors, and that blocking this interaction should therefore result in a reduction of bone resorption. In fact, nature has already provided a positive control to test this hypothesis: echistatin is a potent snake venom that interacts with this $\alpha_v\beta_3$ (i.e., vitronectin) receptor; treatment with this peptide inhibits osteoclastic bone resorption both in vitro [Sato et al., 1990] and in vivo [Engleman et al., 1997]. Searle/Monsanto has demonstrated a similar finding for a VNR antagonist, but only at high doses administered with a continuous fusion system. This is hardly the mode that one would envision for administration of a pharmaceutical. Never-the-less, the concept is sound and if specific compounds for the $\alpha_v\beta_3$ receptor can be developed that will not cross-react with the platelet integrin receptor (i.e., $\alpha_3\beta_2$), then these types of compounds have some potential. Just like the ER and VDR, there is no reason to believe that integrin selectivity cannot be engineered into ligands that block osteoclast binding to the VNR.

Essentially all osteoporosis therapies available today fall into the class of resorption inhibitors. Consequently, the major unmet need with which we are faced in this area is the replacement of lost bone.

BONE FORMATION/OSTEOGENESIS

Replacing bone that has been lost, especially trabecular struts that provide primary support in critical skeletal regions like the hip and spine, continues to be a major hurdle that is yet to be cleared. The list of potential anabolic or osteogenic agents is long and there are data demonstrating promise. While treatment with one such agent, sodium fluoride, has demonstrated that an increase in bone mass can be achieved [Pak et al., 1995], the new bone resulting from fluoride treatment is abnormal and, in fact, more prone to fracture. Consequently, newly developed anabolic bone agents will have to demonstrate fracture efficacy before approval, and this will be a serious challenge to the development of this class of drugs.

Over the past decade, the use of intermittent PTH has been increasingly applied to clinical situations of osteoporosis. After numerous demonstrations in rats that intermittent administration of PTH through daily injections resulted in significant increases in bone mass, a resurgence of interest in this hormone as a potential osteogenic agent has occurred. Recent data from a study in osteopenic women revealed a profound effect of PTH on increasing bone mass [Hodsman et al., 1997]. In fact, when given in combination with an antiresorber such as estrogen (Premarin, 0.625 mg/day) the results were even more impressive [Lindsay et al., 1997].

The pharmaceutical interest in PTH and its analogues is great. In fact, a number of PTH analogues are in various phases of preclinical and clinical development. However, a potential major drawback with these compounds, which at this point are all peptides, is that parenteral administration is required. Will this be a deterrent to those seeking treatment for osteoporosis? This question remains to be answered. Motivation is a powerful impetus, yet osteoporosis is not diabetes. Thus, the industry continues to strive to provide oral therapeutics. Consequently, a small molecule mimetic of PTH that is orally active would be the drug of choice. There are studies under way looking at nasal or deep lung delivery of PTH, which would be more attractive than daily subcutaneous injections, but efficacy and reliability with pulmonary delivery of peptides remain major issues.

It is envisioned that PTH or an analogue would be given over a certain prescribed period of time, perhaps in combination with an antiresorber. This would be followed by discontinuation of PTH, but continuation of the antiresorber in order to maintain the increases in skeletal mass. As discussed previously, the goal is to increase bone mass in order to reduce fracture liability. This will probably mean that PTH therapy will not be required on a permanent basis, but will instead be designed to meet individual need depending upon the level of BMD and fracture risk when therapy is begun. Consequently, temporary treatment for perhaps only 1–2 years with an anabolic peptide like PTH may lessen the concerns regarding parenteral administration.

PTH peptide analogues will lead the way for the next wave in osteoporosis therapy. In addition, other factors like insulin-like growth factor-1 (IGF-1), fibroblast growth factors (FGFs), transforming growth factor-β (TGFβ) bone morphogenetic protein-2 (BMP-2), and prostaglandin E_2 (PGE_2) are also capable of stimulating new bone formation. However, the benefit-to-risk ratio for some of these factors is a potential issue. These factors affect several tissues and, as noted previously, selectivity is a key issue when treating the skeleton. Nevertheless, we can learn much from these compounds regarding their mechanism of action on the skeleton, and future research may lead to the selective mimics that are required. For example, imagine a PGE_2 mimetic that only affects the skeleton and stimulates the production of new trabecular bone as prostaglandins have been shown to do in rodents [Jee and Ma, 1997]. The future targets for these and other mimetics may not be the receptors themselves, which are often expressed in many tissues, unless specific ligands can be developed that only modulate bone cell activity. Instead, the downstream postreceptor signaling apparatus may become the focus for future research in the development of drugs that regulate either osteoblast or osteoclast function.

Finding those downstream targets in bone will certainly not be easy. However, new gene profiling technologies such as differential display, DNA/RNA microarrays, and proteomics will aid in this search. These and additional new technologies will allow us to more completely characterize the model systems that we use to study the skeleton. In doing so, we will increase the chances of finding the ideal tissue-selective drug target for osteoporosis.

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