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Review Article

Hylonome

Rationale for the potential use of calcitonin in osteoarthritis

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Abstract

This review provides evidence that osteoarthritis (OA) or a major subset of OA is not only a disease of cartilage but also a disorder of subchondral bone. This review also discusses the potential efficacy of a bone and cartilage active agent, calcitonin, and discusses how calcitonin might be useful in the pharmaceutical treatment of OA.

Keywords: Calcitonin, Osteoarthritis, Subchondral Bone, Cartilage

Introduction

Calcitonin may be an ideal agent for treatment of osteoarthritis as preliminary data show it to be safe and effective in osteoarthritis (OA). The safety profile of calcitonin is well established. In a recent study[1], oral calcitonin given daily to healthy postmenopausal women for up to 3 months was very well tolerated at doses intended to be given in therapeutic use. This point is worth stressing as any pharmaceutical intervention in patients over 50 years of age who usually have comorbid medical conditions carries an enhanced risk of drug interactions and serious side-effects. Secondly, there is experimental evidence, both *in vivo* and *in vitro*, that calcitonin acts on both cartilage and subchondral bone. Calcitonin not only decreases markedly the enhanced turnover of OA subchondral bone trabeculae but also reduces significantly the severity of cartilage OA lesions in several models of OA and hampers major alterations in the biochemical composition and supramolecular organization of OA cartilage matrix. Finally, calcitonin is analgesic to bone[2] and has been reported to be superior to naproxen in relieving OA knee pain[3]. This may be an additional benefit in the treatment of osteoarthritis which is often painful.

Osteoarthritis is a common and expensive musculoskeletal disease. OA in humans increases in frequency after age 40 and is nearly universal by age 80. OA may result in decreased quality of life. Although the therapeutic options for managing OA have expanded considerably in recent years, most, if not all, currently available non-surgical treatments remain palliative. Our rather limited therapeutic approach includes the control of pain, physical therapies, patient education as well as, whenever possible, the decrease or elimination of local and general risk factors[4,5].

Although inflammatory processes do play a role on provoking signs and symptoms[6-8], it is still debated whether simple analgesics such as paracetamol or non-steroidal anti-inflammatory drugs (NSAIDs) should be used as the oral drugs of first choice and, if effective, as the preferred long-term analgesics[9,10]. The use of NSAIDs is associated with the occurrence of serious adverse events particularly gastrointestinal. When compared to conventional NSAIDs, the selective cyclo-oxygenase-2 (COX-2) inhibitors reduce the incidence of ulcerations of the upper gastro-intestinal tract but they offer no apparent advantage with respect to other toxicities such as fluid retention, hypertension, congestive heart failure and renal insufficiency[8]. Further, some of the selective COX-2 inhibitors increase the risk for cardiovascular thrombotic events[11].

The intra-articular injections of preparations of high molecular weight hyaluronan in patients with knee OA seem to be effective in reducing pain, but there is no convincing evidence that this therapeutic approach slows the progression of joint damage[12]. The therapeutic role of chondroitin sulfate, glucosamine and other compounds is also debated[13-15]. When pain becomes intractable or when the joint has reached the end stages of the disease process, total joint arthroplasty may be helpful for many OA sufferers[16]. However, the arthroplasty has a limited life span and the surgical procedure, that does not restore a normal joint, relieves pain more pre-

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dictably than it improves function and can be associated with serious general and local complications.

The following discussion stresses the concept of the joint as an organ consisting of cartilage, subchondral bone and other tissues, outlines our current understanding of cartilage metabolism both in health and OA, explores the potential role of the underlying bone in the pathophysiology of OA, and reviews the effects of calcitonin on cartilage and subchondral bone in animal models of OA and in human OA as well.

Normal function of diarthrodial joint function depends on the health of its different components

Diarthrodial or synovial joints are complex organs. The articular capsule, muscle/tendon elements and ligaments link the bones, provide joint stability and attenuate the forces delivered to the joint. The ends of bone are covered by hyaline cartilage, a viscoelastic tissue that not only permits the smooth movement of bone on bone, but also distributes the load-bearing forces more evenly on the underlying bone. The biomechanical properties of articular cartilage depend on the structure and composition of its abundant extracellular matrix whose major components are type II collagen and the proteoglycan termed aggrecan[17]. The formation of pyridinium cross-links between neighboring collagen molecules strengthens the dense network of collagen fibrils which imparts shape and tensile properties to the cartilage tissue while the filamentous hyaluronan molecules interact non-covalently with many aggrecan molecules to form aggrecan aggregates which then become firmly immobilized at high concentration within the collagen network and, thereby, endow cartilage with viscoelastic properties. Because articular cartilage is avascular, nutrients to the chondrocytes must be provided by the well vascularized subchondral bone and by the synovial fluid which link together the different joint components.

Obviously, changes in the metabolism, architecture, supramolecular organization and/or architecture of any one joint tissue will change the mechanical performance of this joint tissue components and will have consequences on the loads experienced by other joint components. Therefore, joint diseases should not be considered as the disease of a single joint component but rather as the failure of a whole organ.

An evolving picture of joint metabolism

For many years, cartilage had been thought to be a metabolically inactive tissue that becomes degraded by being subjected to the wear and tear of everyday life. However, during the last decade, the use of sensitive assays to quantify in body fluids the molecules or fragments derived from cartilage and other joint components has pointed out that the different joint components are in a constant, dynamic state of adaptation and response to the patterns of load distribution in the joint[18,19]. Indeed, throughout adult life, chondrocytes orchestrate the turnover of cartilage matrix molecules, and, in turn, are protected from the potentially damaging forces of mechanical function by the extracellular matrix they produce. The rate of turnover of proteoglycans (PGs) is much more rapid than that of collagen fibrils, and, during normal turnover, PG degradation is rather a conservative process in order to permit renewal of the PG molecules without weakening the tissue. Although proteinases belonging to different classes (disintegrins and metalloproteinases with thrombospondin-like motifs or ADAMTs as well as serine-, cysteine-, aspartic- and metallo-proteinases) have been detected in cartilage, it is generally believed that, both in health and diseases, collagenase-3 or MMP-13 initiates type II collagen degradation whereas ADAMT4 is the major enzyme responsible for aggrecan degradation[20].

Synovium is a major source of cytokines and growth factors which reach the cells by diffusion through the matrix[21]. These molecules have a major influence on the expression of matrix components and they also regulate matrix turnover by controlling proteinase synthesis, secretion, activation and inhibition.

Mechanical forces associated with joint loading can dramatically alter chondrocyte synthesis, assembly, and degradation of extracellular matrix components in a manner dependent upon the type, magnitude, duration, and frequency of applied load[22].

Pathophysiology of the osteoarthritic disease process

Previously considered as a "degenerative" disease that must be accepted as the inevitable consequence of trauma and ageing, OA is now viewed as a dynamic, essentially reparative process with exciting potential for health intervention. Although OA prevalence increases with aging, cartilage changes seen in OA differ from age-related cartilage changes: with aging, the water content goes down, the hyaluronan content increases and the biomechanical properties remain unchanged whereas, in OA, the water content increases, the hyaluronan content goes down and the tensile, compressive, and shear moduli of the matrix are significantly decreased.

During the OA disease process, the anatomy and composition of all the different joint components are altered by a complex combination of degradative and reparative processes which depend upon an interplay between, on the one hand, local biomechanical factors acting on the joint and, on the other hand, generalized factors making a predisposition to the disease[23,24]. However, relatively little is known about which of the disease processes and etiological factors initiate or control progression of the disease process in OA joint[25].

Because studies of human OA are largely confined to examinations of tissue obtained postmortem and at joint replacement surgery, animal models have been created to know more about the early events of the OA disease process[26]. The cruciate deficient dog model is well-established: transection of the anterior cruciate ligament of the right knee (ACLT) leads to progressive pathologic changes that are unambiguously those of OA, the cartilage being lost from the articular surface after a

time period of 4 to 5 years[27,28]. Within weeks following ACLT, the cartilage from the operated knee joint exhibits a progressive depletion of its hyaluronan content as well as a disappearance of its fast sedimenting PG aggregates[29-31]. These striking changes in the biochemical composition and supramolecular organization of articular cartilage precede anatomical lesions and have also been observed in human OA[32-34].

This model of post-traumatic OA is also characterized by an early and sustained increase in the urinary levels of deoxypyridinoline, a specific marker of bone resorption, and in blood levels of antigenic keratan sulfate, a marker of aggrecan turnover, and hyaluronan, a marker of synovial proliferation and hyperactivity[35-37]. These findings are worth stressing since, in humans, a sustained increase in the turnover of subchondral bone as well as a sustained increase in the serum levels of hyaluronan both predict disease progression in OA of the knee[38,39]. However, thus far, it is still unclear whether OA changes occur first in cartilage, in bone, or are concurrent.

Osteoarthritis and the failure of biomechanical properties of articular cartilage

Biomechanical studies point out that normal cartilage subjected to normal biomechanical forces does not break down whereas normal cartilage will fail in response to single or multiple traumatic events, and defective cartilage will gradually disintegrate in the presence of normal wear[17]. Accordingly OA could be considered as the final common pathway of several different but possibly interacting etiological factors leading to failure of mechanical properties and breakdown of cartilage matrix. These factors may include inflammatory disease, joint overload or trauma, developmental defects, genetic and chondrocyte injury.

On the other hand, in human OA there is a strong correlation between the biomechanical properties of cartilage, the tissue biochemical composition and the stage of the disease[40,41]. Thus it would appear that, although mechanical factors might be most prominent in inducing and promoting OA, changes in chondrocyte metabolism and alterations in matrix composition might be more prominent in the perpetuation of OA and in influencing predisposing conditions for OA development. Since alterations of the material properties and changes in the biochemical composition of the matrix are an integral part of the OA disease process it may be argued that the changes in the macromolecular organization of PGs and the loss of matrix molecules are key events at some times in the development of OA[29-34].

In the early stages of canine experimental OA, the cartilage from operated knees becomes thicker owing to an increase in its water and PG content. This phenomenon, termed hypertrophic repair has also been observed in other animal models of OA and in the early stages of human OA as well[42,43]. However, despite its enhanced PG content, OA cartilage does not withstand mechanical stress as well as normal hyaline cartilage and full-thickness loss of cartilage tissue occurs with time post-surgery[17,28]. Because PG aggregation increases dramatically the rheological properties of PG molecules[44], these observations might be related, at least in part, to the reduction in cartilage content of hyaluronan (HA), the backbone upon which aggrecan molecules align to form aggregates. Accordingly, the reduction in HA content seen in OA cartilage is likely to contribute to the apparent irreversibility of the OA disease process, a contention strengthened by the observation that the content of HA in articular cartilage remains normal in disuse atrophy, a condition in which the chondrocytes are able to overcome the loss of PGs that ensues as the result of increased enzyme activities[31,34,45].

The stimulation of PG synthesis, which is sustained for at least 64 weeks after ACLT and which has also been noted until the late stages of human OA, usually overcompensates for increased proteoglycan degradation secondary to the chondrocyte release of proteinases including MMPs and ADAMTs[20,46]. On the other hand, the increase in the rate of collagen synthesis is less pronounced than that of PGs and does not seem to be able to (over) compensate the rate of collagen breakdown, which is believed to be initiated by MMP-13 or collagenase-3[47]. The release of MMPs by chondrocytes is known to be affected by a wide range of conditions and processes, many of which may play a role in OA. In humans, increased weight and preceding joint injury, both of which alter the normal biomechanics of the joint, have been shown to increase the risk of developing OA[48]. Attempting to mimic these effects *in vitro* by subjecting cartilage explants to mechanical stimuli results in not only an increase in proteoglycan synthesis but also an increase in release of degradative enzyme activity[22]. A number of cytokines and cell-signaling molecules, including interleukin-1 and nitric oxide, have also been shown to play an important role in chondrocyte response, upstream to the release of the various degradative enzymes[49].

The heightened metabolic activity, both anabolic and catabolic, exhibited by chondrocytes in canine experimental OA is reflected by the wide range of fragments of type II collagen, proteoglycans and other matrix molecules that can be found in the synovial fluid, serum and urine of animals thus affected[18,47]. These fragments serve as potential biomarkers for the disease process, and it is likely that over the next decade biomarkers in OA will play as large a role in this disease, and possibly larger than biomarkers do in osteoporosis, another disease involving extracellular matrix.

Possible role of subchondral bone in the OA process

Obviously, the OA disease process is associated with changes in periarticular bone including sclerosis of the subchondral bone, the formation of so-called cysts within the bone, and the development of osteophytes. Whether bone changes are secondary or play an active role in the initiation and/or progression of OA still remains a matter of debate.

Radin and Rose[50] proposed that, by enhancing shear and tensile stresses in the overlying cartilage, alterations in underlying subchondral bone stiffness may be an important

etiologic factor in OA. Although the authors did not provide any direct experimental data to support the shear hypothesis, it should be noted that cartilage damage does not always lead to full-thickness cartilage loss, and thus to OA[51]. Further, normal subchondral bone attenuates loads through the joint to a greater degree than either articular cartilage or periarticular soft tissues and hence protects the overlying cartilage from damage caused by excessive loads[52,53]. Therefore, the health and integrity of the overlying cartilage depend on the mechanical properties of its bony bed.

According to Wolff's law, bone remodeling or changes in the density and distribution of bone, reflects the adaptation of the bone tissue to variations in mechanical load. Therefore, the articulating ends of bone maintain a joint shape that is dependent on mechanical stresses in the joint. In patients with OA, radiography reveals the bone anatomical changes that have occurred in the past whereas bone scintigraphy reflects the level of bone remodeling activity at the time of the scan, the degree of radioisotope uptake being related to the extent of mineral surface exposed at sites of bone resorption or formation.

The following sequence of subchondral bone changes can be deducted from studies conducted in animal OA models and in human OA as well. The earliest bone changes appear to be a marked decrease in the apparent volume and density of the trabecular subchondral bone whereas the subchondral plate thickness remains normal[54-56]. In canine experimental OA, this osteopenia of periarticular cancellous subchondral bone is associated with a marked increase in the urinary levels of deoxypyridinoline cross-links, which occurs as early as at 6 weeks after ACLT and which is sustained for at least 14 weeks after surgery[35]. An increase in thickness of both the subchondral cortical plate and the subjacent horizontal trabeculae only occurs at later stages of the disease process but prior to joint space narrowing, and thus cartilage loss[57-59]. When joint space narrows, and thus cartilage starts to be lost, the subchondral cortical plate and the trabeculae become thicker. Further, as cartilage loss progresses, the articular surface of opposing bones flattens and acquires greater articular congruence. Ultimately, the cancellous bone in the sub-articular region collapses leading to altered limb alignment and deformity.

The new bone formed in OA is far from being normal. It is laid down rapidly as a coarse fibrous or woven bone as observed in areas of bone repair[53]. Further, when compared to normal joints, the subchondral cortical plate and trabecular bone from OA joints are less stiff and dense, showing a greater porosity, a reduced mineral content as well as mechanical weakness[60-62]. Thus, a distinction must be made between, on the one hand, bone's apparent density that is defined as bone mass/total volume, and, on the other hand, bone's material density that is defined as bone mass/bone volume[53]. The so-called subchondral bone sclerosis detected on radiographs of OA joints reflects the increase in apparent density whereas, in fact, the subchondral trabecular bone exhibits a reduction in mineral content and material density. It is believed that this hypomineralization of OA trabeculae results from enhanced bone turnover as detected by changes in alkaline phosphatase, osteocalcin, or other biochemical markers[53,63].

Although a cross-sectional evaluation of biochemical markers of bone metabolism in patients with knee OA suggests a decrease in bone turnover[64], the vast majority of data in the literature point out to the contrary. Indeed, OA femoral heads exhibit enhanced levels of immature keto-imine cross-linked collagen[63] and, in patients with OA, the urinary levels of deoxypyridinoline, a marker of bone resorption, are increased and correlate with disease severity according to radiographic grading[65,66]. Further, increased subchondral uptake of technetium labeled bisphosphonate predicts cartilage destruction and correlates with biochemical markers of bone and cartilage turnover[67,68]. It is worth stressing that in the early stages of canine experimental OA and in humans suffering ACL injuries, an increase in the scintigraphic uptake is specific for the unstable knee and not seen on the contralateral, stable knee[69,70]. Interestingly, after surgery to repair the ACL injury, the radionuclide uptake declines, suggesting that restoration of more normal loading may be able to mitigate the bone remodeling effects observed.

Increased radionuclide bone uptake has been associated with the "bone marrow edema" that is detected by T2-weighted MRI imaging. These high intensity MRI lesions, which in fact include bone marrow fibrosis, bone marrow necrosis, and abnormal trabeculae[71], exhibit a close relationship with progression of the OA disease process as assessed by the rate of narrowing of the overlying joint space[72-74]. As similar changes have been reported in patients with avascular necrosis and stress fractures, it is hypothesized that such lesions may either be causally related to the progression of OA or be the consequence of increased loading, and again a marker for further cartilage damage.

Bone remodeling and cartilage degradation

Although the above data support bone remodeling being a marker of the disease process, they nevertheless suggest that there is a causal relationship between bone changes and cartilage OA changes, a contention further strengthened by the observation that, in canine experimental OA, calcitonin, a potent marker of osteoclastic bone resorption, reduces not only the urinary levels of deoxypyridinoline cross-links and the periarticular bone loss but also, and most importantly, the extent of cartilage lesions[35,56].

The finding of increased bone metabolic activity in anatomic association with the involved joint cartilage in OA suggests a close coupling of biologic processes, perhaps both driven by similar mechanisms. The formation of a coarse fiber woven bone that exhibits enhanced lysine hydroxylation and decreased mineralization in the region of subchondral sclerosis might be seen as an attempt at repair since this type of bone is similar to callous bone formation at sites of fracture repair and at regions of rapid bone growth. Further, the finding that OA subchondral trabeculae contain type I collagen homotrimer in addition to the predominant type I colla-

gen heterotrimer found in normal bone might suggest a changed phenotypic expression of osteoblasts[75]. Whatever, such biochemical changes in collagen would be expected to alter the biomechanical properties of bone, rendering it weaker[61,62]. The resulting increase in deformation of the articular bone is likely to enhance the shear and tensile stresses generated in the overlying articular cartilage and, thereby, to contribute to the biomechanical attrition of the cartilage tissue, a contention strengthened by the observation that, in animals treated with calcitonin, the lack of reduction in subchondral bone mass was associated with a marked reduction in the severity of cartilage OA lesions in the cruciate-deficient joint[35,56]. Further, in the population-based Chingford study[76], women with progressive knee OA had higher urinary levels of type I collagen cross-links than those without progressive disease whereas the Framingham prospective study[77] has shown that, among women who already have knee OA, those with low bone mineral density, and those who are losing bone faster exhibit more rapid progression of radiographic changes than those with high bone mineral density who are losing bone more slowly.

On the other hand, molecules produced by bone cells could alter the metabolism of chondrocytes and vice versa. Thus, a yet unidentified factor released from OA subchondral bone stimulates the degradation of proteoglycans in cartilage explants culture[78]. Further, mechanical loading enhances cytokine production by osteoblasts and pressure can alter chondrocyte cytokine receptor expression[79]. Moreover, synovial fluid from OA patients, which contains soluble products of cartilage metabolism as well as a variety of anabolic and catabolic molecules produced by cells in the surrounding tissues, can significantly stimulate proliferation of osteoblast-like cells derived from trabecular bone obtained at knee surgery whereas bone cell migration contributes to synovial hyperplasia and osteophyte formation[80,81].

The overall findings described above support the concept that OA, or at least a major subset of this heterogeneous group of disorders, is a disorder of bone. Therefore, therapeutic strategies should preserve the normal healthy bone, prevent the osteoporotic changes in the sub-articular region, and slow down the deposition within the subchondral region of a weaker woven repair bone. By maintaining the normal contour and shape of the articular surfaces, such approach would preserve the patient's mobility for longer time periods than therapies directed at preserving articular cartilage alone since the latter is unambiguously affected by the alterations in the subchondral bone described above.

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