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An introduction to the pathophysiology of osteoarthritis

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TABLE OF CONTENTS

1. ABSTRACT

Osteoarthritis involves the degeneration of articular cartilage together with changes in subchondral bone and limited intra-articular inflammation. In this chapter these changes are reviewed at the tissue, cell and molecular levels to reveal the complexity of a process which involves multiple changes in joint structure and turnover.

2. INTRODUCTION

Osteoarthritis (OA) represents a clinical classification of pathological conditions involving a progressive degeneration of articular cartilage, a remodelling of sub-chondral bone and a synovitis which is usually limited.

The condition is variously described as a part of a process of age-related change or a disease. It is twice as prevalent in women than men and increases in incidence with age, there being a major rise after 60 years (1). It is believed that the changes that lead to the development of OA are slow (insidious). That in idiopathic OA clinical presentation may result from changes over 15-20 years. The disease may involve primarily one or two large joints or may be generalized. Following joint trauma there is an increased incidence of OA (1) which probably results from accelerated degeneration over a period of about 10-15 years. In contrast, familial OA presents very early, often following cessation of growth, as a consequence of alterations in cartilage matrix structure which leads to the manifestation of joint degeneration following the cessation of growth. An example is provided by patients with a mutation in the type II collagen gene (2).

3. GENERAL CHANGES IN BONE AND CARTILAGE IN OSTEOARTHRITIS

3.1. Bone

OA involves not only the degeneration of articular cartilage leading to eburnation of bone but also a synovitis that is usually limited. There is extensive re-modelling of sub-chondral bone resulting in the so-called sclerosis of this tissue observed radiographically. These bone changes are often accompanied by the formation of sub-chondral cysts as a result of focal resorption. These are particularly noticeable by magnetic resonance imaging (see below).

The bone changes may also be systemic in nature. The work of Dequeker and his colleagues (3) has produced evidence of changes in bone metabolism in sites such as the iliac crest which are suggestive of systematic changes. Analyses of the molecular composition of OA bone have provided indications of fundamental changes in bone metabolism (4). Deoxypyridinoline cross-links, resulting from bone resorption, are elevated in urine (5) as is osteocalcin elevated in serum (6).

There is evidence from scintigraphy to indicate that changes in bone metabolism are identifiable several years prior to evidence for clinical onset of the disease (7-9). Whether these changes precede those in cartilage

remains to be determined once comparable analyses can be made of metabolic changes in cartilage metabolism.

The dynamic interplay between bone and cartilage is reflected in how changes in one tissue may influence the other and thus determine the development of OA. Bone cells from OA patients can alter chondrocyte metabolism (10). This is most strikingly observed in osteoporosis, where bone density is reduced by excessive osteoclastic resorption of bone. Patients with osteoporosis usually show little or no evidence of OA. Moreover, patients with OA do not usually develop osteoporosis (11-13). These observations may be explainable in part at the level of biomechanical interactions, indicating that reduced bone density may protect against degeneration leading to OA.

A principle anatomical feature of OA is the development of osteophytes. The osteophytes, which have a cap of articular cartilage, and an actively remodelling bone base, may serve to reintroduce some stability into an otherwise unstable joint.
These form from an endochondral process in sites at the edges of the damaged articular cartilage. Addition to periosteum in culture or injection in vivo of transforming growth factor-beta results in expression of the hypertrophic phenotype by newly formed chondrocytes in periosteal tissue and subsequent mineralization of extracellular matrix (14, 15). This is an essential requirement for endochondral ossification. Intra-articular injection of TGF-beta$_1$ induces osteophyte formation (16). Thus TGF-beta, and probably other bone morphogenetic proteins, may play an important role in osteophyte formation. These bone morphogenetic proteins are also excessively produced in joint inflammation.

3.2 Cartilage

The classic loss of articular cartilage observed in OA may be initiated as a focal process as is observed, for example, in early experimentally induced OA in rabbits following section of the anterior cruciate ligament and/or partial medial meniscectomy. Focal lesions may progressively enlarge to involve specific joint compartments, inducing alterations in articulating surfaces by producing changes in loading. Degenerative changes may involve the whole articular cartilage in post-traumatic OA where alterations in cartilage matrix turnover are detectable within days or weeks following joint injury such as damage to the anterior cruciate ligament or meniscus (17).

In idiopathic OA, degeneration is first observed at the articular surface in the form of fibrillation. This initially involves splits more or less parallel to the articular surface. Later splits penetrate the damaged more superficial cartilage. Cell cloning is observed early on but again is confined to more superficial sites. Progressive loss of cartilage then occurs. Apoptosis is enhanced in OA, particularly at and close to the articular surface (18, 19).

These degenerative changes ordinarily represent a very slow process which may take as long as 15-20 years. But it is accelerated in cases of joint injury and inflammation (synovitis) or it may present clinically on cessation of growth as in familial OA. For reasons that remain unresolved degeneration is often more pronounced in the tibial cartilage particularly in the medial-compartment. This is observed in human and experimental studies of OA and also in experimental inflammatory (rheumatoid-like) arthritis (20).

4. GENERAL MOLECULAR CHANGES IN CARTILAGE MATRIX AND ALTERATIONS IN GENE EXPRESSION

4.1. Cartilage Degradation

Superficial fibrillation at and close to the articular surface is associated with increased denaturation and loss of type II collagen of collagen fibrils (21, 22). This leads to a loss of tensile properties, particularly in OA prone joints such as the hip (24) whose mechanical properties are determined by these fibrils (23-25). These tensile properties are normally much higher at the articular surface than elsewhere in the cartilage (23). It is here that they are first lost in OA development (23, 25). Damage to the fibrils leads to a loss of the small proteoglycans decorin and biglycan which are usually closely associated with the fibrils at the articular surface (26). In addition there is loss of the large proteoglycan aggrecan (22).

The loss of these molecules is associated with increased cleavage of type II collagen by collagenase (27) and aggrecan cleavage (28-30) and the degradation of small proteoglycans (30). This usually correlates with increased expression and content of various metalloproteinases (MMP) at the articular surface early in the degenerative process (20, 31). These include stromelysin-1 (MMP-3), gelatinases A (MMP-2), and B (MMP-9) and collagenases-1 (MMP-1), collagenase-2 (neutrophil collagenase or MMP-8), (32), collagenase-3 (MMP-13) (32-34) and MT1-MMP (membrane type 1- MMP or collagenase 4 or MMP-14) (35, 36). Matrilysin expression is also increased (37). These proteinases are very much involved in the excessive matrix degradation that characterizes cartilage degeneration in OA (20, 31). Activators of prometalloproteinases which are increased in OA include MT1-MMP which activates gelatinase A and collagenase-3 and gelatinase A which can superactivate collaenase-3 (38), stromelysin-1, which can superactivate collagenases (20), and plasminogen activator (39) that activates plasminogen to produce plasmin which is a general MMP activator (20). Also the cysteine proteinase cathepsin B is upregulated in OA (40, 41) and may be an important activator of MMPs (42).

4.2. Cartilage synthesis and changes in matrix structure.

The early damage to and loss of these molecules in OA is accompanied by an increased content of biglycan and decorin (26) and aggrecan (A.R. Poole, A. Reiner, unpublished) in the mid and deep zones presumably to compensate for the increased loading on the chondrocytes with the loss of the more superficial cartilage. There is also a marked increase in the synthesis of type II collagen in these sites (43), mainly type IIB as revealed by experimental studies (44) but also some type IIA (45),

normally only observed prior to chondroblast differentiation early in development (46). There is induction of limited expressions and of synthesis of type III collagen (47). Type VI content is increased (48-50).

Aggrecan synthesis is upregulated (20, 30) and altered chondroitin sulfate structure is demonstrable by immunoassay or immunohistochemistry (29, 51, 52) in response to the increased damage. Larger aggrecan molecules appear with more advanced degeneration reflective of new synthesis (29). There is increased expression and synthesis of the proteoglycans versican, decorin, biglycan, fibromodulin and lumican as well as of link protein (53, 54). This synthetic increase is associated with an enhanced content of insulin growth factor-1 (55) which is a potent stimulant for aggrecan synthesis. This increased synthesis is seen in the same sites where degradation is enhanced.

There are many changes in the contents of other matrix molecules in OA. Cartilage oligomeric protein, is altered in distribution (56), contents of cartilage matrix protein (57), tenascin (58), osteonectin (59) and fibronectin (60) are all increased.

Ordinarily mature articular chondrocytes do not express type X collagen, which is normally synthesized by hypertrophic chondrocytes in the growth plate in association with cartilage matrix resorption. But in OA type X expression and synthesis occurs (61). This is observed in degenerate cartilage and in association with increased expression of the cell surface type II collagen receptor annexin V (62). These molecules are normally highly expressed by early hypertrophic chondrocytes. Parathyroid hormone related peptide, which is expressed by pre-hypertrophic cells in the growth plate and suppresses hypertrophy, is also upregulated in OA cartilage (63, 64). This expression of hypertrophy is accompanied by apoptosis, a feature of terminal hypertrophic cells (18, 19). These changes are observed in the more superficial and mid zones and may represent a chondrocyte response to a damaged extracellular matrix with the reversion to a more fetal phenotype.

5. REGULATION AND MECHANISMS OF CARTILAGE MATRIX TURNOVER

As damage to the cartilage progresses one can recognize increased denaturation (21, 22) and increased cleavage of type II collagen by collagenase (27). Denaturation is seen especially around chondrocytes but it extends into interterritorial sites remote from these cells, in OA, unlike what is seen in ageing. This clearly implicates the chondrocyte as a mediator of the damage to these molecules within the matrix which were synthesized by these same cells. Studies with an inhibitor that blocks the activities of collagenases, except collagenase-1, have revealed that collagenase cleavage of type II collagen in most human OA cartilages and mature bovine articular cartilages can be frequently inhibited in human cartilages (Dahlberg, L., Billinghurst, R.C., Manner, P., Nelson, F., Webb, G., Ionescu, M., Reiner, A., Tanzer, M., and Zukor, D., Chen, J., Van Wart, H.E., and Poole, A.R., submitted) and always in the case of bovine articular cartilages (Billinghurst, R.C., Ionescu, M., Reiner, A., Wu, W., Dahlberg, L., Chen, J., Van Wart, H., and Poole, A.R., submitted) implicating collagenases such as collagenase-3 in this process.

What is responsible for the increased synthesis and activation of these MMPs at the cellular level is still unclear. There is evidence that some fragments of fibronectin can stimulate chondrocyte-mediated cartilage resorption via cell surface receptor activation (65) just as in fibroblasts MMP-1 is upregulated through an RGD-integrin receptor activation (66). Since fibronectin is produced in increased amounts in OA cartilage (60) this activation of chondrocyte degradation by matrix degradation products may play an important role in establishing positive feedback generation of proteolysis. Such cellular responses involve the production of cytokines such as IL-1 which no doubt plays an autocrine/paracrine-role (67).

There is increased expression on OA chondrocytes of the receptors for interleukin-1 (68) and of IL-1 itself, even more than in rheumatoid arthritis (RA) (69). Tumor necrosis factor alpha, is also upregulated in OA even more so than in RA (69) and the receptor for TNFalpha shows increased expression when compared to normal cartilage (70). The presence of the TNFalpha p55 receptor (but not the p75) on OA chondrocytes correlates with the susceptibility of cartilage explants to TNFα-induced proteoglycan loss (70).

IL-1 and TNFalpha are both potent activators of cartilage degradation *in vitro* (20, 31). In combination with oncostatin M, IL-1 is even more potent in causing cartilage resorption but levels of this cytokine, a member of the IL-6 family, are not usually elevated in OA SF (71). Thus oncostatin may be of more importance when synovitis is more pronounced in the OA joint. Other cytokines that are upregulated in OA include macrophage inflammatory protein-1beta (72).

Nitric oxide (NO) synthase (iNOS) is upregulated in OA chondrocytes compared to normal (73) and to RA cartilage (69). This is associated with increased generation of NO. Il-1 and TNFalpha are potent stimulators of NO production in cartilage (74). The expression of iNOS with TNFalpha and IL-1beta is correlated in chondrocytes from arthritis patients (69). NO inhibits aggrecan synthesis, when induced by IL-1 (75). However, protease activity and proteoglycan degradation are enhanced when NO production is blocked (76). Thus it may also play a protective role. NO can also induce apoptosis in chondrocytes (77) and this may account, in part, for the increased apoptosis seen in OA.

Changes in extracellular matrix (ECM) loading can also induce ECM cleavage as well as changing the synthesis of ECM macromolecules (31). The pathological changes in cartilage ECM in OA are likely to result in a disturbance of the normal balance between mechanical loading and direct cytokine/growth factor signalling changing gene expression.

Analyses of the proteoglycan aggrecan have revealed that excessive cleavage occurs in OA cartilage in the core protein, particularly in the interglobular domain between the G1 and G2 domains. There are two principle sites of cleavage, the MMP site where multiple MMPs including stromelysin-1 can cleave, and the aggrecanase site where cleavage can also be produced by a number of proteinases (20), including membrane proteinases with characteristics of the ADAMTS family (78). Both these cleavages are much enhanced in OA cartilage (28). It is still unclear as to the relative contributions of different proteinases in aggrecan degradation since quantitative assays to measure both cleavage sites have not been used.

A deficiency of the tissue inhibitors of MMP (TIMPs) in OA (79, 80) clearly favours the excessive proteolysis that is observed in the diseased articular cartilage.

6. INFLAMMATION

There is no question that in established OA there is some synovitis (inflammation of the synovium) albeit much less than is ordinarily observed in diseases such as RA (81, 82). Thus it is more appropriate to speak of OA rather than osteoarthrosis when referring to established disease. Inflammation may be of earlier onset in post-traumatic OA in view of observations of early marked increases in metalloproteinases in synovial fluid, such as stromelysin-1 which are more likely to be derived mainly from activated synovial cells than cartilage. Synovial cells actively synthesize and secrete hyaluronic acid (HA, hyaluronan) as well as many other cells in the body. However, in studies of RA close direct correlations have been observed between serum HA and joint inflammation and diseases progression (joint damage) (83, 84). The latter has also been observed in OA for persistent elevation of serum HA where accelerated progression in joint damage has been reported (85). Moreover, HA levels correlate with knee joint space in OA (85, 86). Similarly, cartilage oligomeric protein which is synthesized by synovial cells and chondrocytes when expressed to TGFbeta (87), is increased in patients which exhibit accelerated large joint degeneration (88, 89). These associations suggest that joint inflammation may accelerate joint damage. This would likely result from local proinflammatory cytokine generation (see below).

C-reactive protein is also elevated in serum in patients with OA (90) as is eosinophil cationic protein and myeloperoxidase (A.R. Poole, M. Ionescu, unpublished). Together these changes provide evidence for a clearly definable systemic and local inflammatory component in OA. Recently, evidence for T cell immunity to the cartilage proteoglycan aggrecan and link protein has been reported. This is also observed in OA (91). T-cell immunity to type III collagen has previously been seen in experimental OA induced by partial meniscectomy (92).

7. CHANGES IN MOLECULAR MARKERS IN BODY FLUIDS REFLECT SKELETAL CHANGE AND INFLAMMATION

Cartilage degradation can now be detected using antibodies to the collagenase generated cleavage site in type II collagen. This is increased in joint fluids of rabbits and dog following section of the anterior cruciate ligament (A.R. Poole, C. Billinghurst et al., in preparation). Other assays to detect type II collagen degradation products involving the carboxy telopeptide-cross-link complex are also under development (D. Eyre and L. Atley, unpublished; I. Otterness, personal communication). Synthesis of type II procollagen can be detected by measurement of the c-propeptide of this molecule (43). The content is increased in OA synovial fluid and following traumatic injury that can lead to OA. However, serum c-propeptide content is decreased in idiopathic OA (43). In familial OA serum c-propeptide content is increased in most patients (93) reflecting the degeneration of the articular cartilage.

Proteoglycan degradation products bearing an antigenic KS epitope may be increased (94) or decreased (84) in serum in OA patients. An epitope (termed 846) present on the proteoglycan aggrecan, on some of the chondroitin sulfate chains of molecules, is released from cartilage and correlates with synthesis of aggrecan. This is markedly increased in content in OA synovial fluid (95, 96).

Progression of OA from knee joint pain to joint space narrowing is reflected by an increase in serum cartilage oligomeric protein (88, 89). Since this molecule is also secreted by synovial cells activated by TGFbeta (87, 88) it may also reflect a synovitis. Hyaluronic acid (hyaluronan) has also been shown to be increased in serum in knee OA patients (97) particularly in those with more rapidly undergo joint degeneration (85). As in studies of patients with rheumatoid arthritis (83), this may signify a synovitis that accelerates cartilage damage since there is a correlation between serum HA and joint space (85, 86).

Increased bone turnover in OA is reflected by increases in the bone specific deoxypyridinoline cross links in urine (5). Serum osteocalcin (6) and bone sialoprotein (89) may also be increased.

These markers are proving useful in assessing disease progression and activity and response to therapy designed to control inflammation, arrest cartilage degradation and promote synthesis and tissue repair. They are being introduced into clinical trials to both investigate chondroprotection and determine whether they may be prognostic of longer term outcome of therapy. Such a careful double-blinded validation process is essential if their usefulness is to be clearly determined and critically assessed.

6. CONCLUDING REMARKS

Much progress has resulted from increased research in OA in recent years. The application of technical advances in analytical techniques to study cells and tissues has provided important new insights into the pathophysiology of OA and identified new therapeutic targets for intervention and regulation of this degenerative process.

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