Oxidative stress in secondary osteoarthritis: from cartilage destruction to clinical presentation?

Christoph Ziskoven,1,2 Marcus Jäger,1 Christoph Zilkens,1 Wilhelm Bloch,2 Klara Brixius,2 Rüdiger Krause3

1Orthopedic Department, Heinrich-Heine University Medical School, Düsseldorf, Germany
2Department of Molecular and Cellular Sport Medicine, German Sport University, Cologne, Germany

Abstract

Due to an increasing life expectancy, osteoarthritis (OA) is one of the most common chronic diseases. Although strong efforts have been made to regenerate degenerated joint cartilage, OA is a progressive and irreversible disease up to date. Among other factors the dysbalance between free radical burden and cellular scavenging mechanisms defined as oxidative stress is a relevant part of OA pathogenesis. Here, only little data are available about the mediation and interaction between different joint compartments. The article provides a review of the current literature regarding the influence of oxidative stress on cellular aging, senescence and apoptosis in different joint compartments (cartilage, synovial tissue and subchondral bone). Free radical exposure is known to promote cellular senescence and apoptosis. Radical oxygen species (ROS) involvement in inflammation, fibrosis control and pain nociception has been proven. The data from literature indicates a link between free radical burden and OA pathogenesis mediating local tissue reactions between the joint compartments. Hence, oxidative stress is likely not only to promote cartilage destruction but also to be involved in inflammmatory transformation, promoting the transition from clinically silent cartilage destruction to apparent OA. ROS induced by exogenous factors such as overload, trauma, local intraarticular lesion and consecutive synovial inflammation cause cartilage degradation. In the affected joint, free radicals mediate disease progression. The interrelationship between oxidative stress and OA etiology might provide a novel approach to the comprehension and therefore modification of disease progression and symptom control.

Introduction

Osteoarthritis (OA) is a common and progressive chronic disease leading to impaired joint function and can result in immobility mostly in elderly people.1 The prevalence of OA increases significantly in advanced age. Radiographic evaluation of knee x-ray images taken from the Framingham osteoarthritis study showed alterations in K/L (Kellgren/Lawrence Grade) > or =2 in 44% of parents (mean age 72 years) and 22% of offspring (mean age 54 years).2 Although several risk factors like overweight or the family history of OA are well known, the mechanisms on the cellular level leading from the presence of risk factors to cartilage degeneration are not completely understood in detail. Besides other agents, recent data focus on oxidative and nitrosative stress as one aspect involved in the pathogenesis of OA.3 In the course of the growing knowledge about the relevance of free radicals for cellular ageing, their involvement in degenerative diseases of the joint came into focus of more detailed investigations.4

Method of data selection

We present a review of the current literature over the relevance of free radicals in OA pathogenesis. Here, all relevant original publications in this topic documented in the USA National Library of Medicine and the library of the National Institutes of Health, USA were considered. Reviews and single case reports were excluded. The online search was conducted by use of the terms oxidative stress, oxidative damage and radical oxygen species, each of them combined with the search term osteoarthritis. This yielded a cumulative number of 203 studies. Of these results, 33 were identified as review or case report, leaving 170 relevant studies for this review. The relevance of the studies cited herein was considered by actuality, included sample size and methods as well as by the number of being cited evaluated by use of ISI Web of Science®. Finally, the selected studies are discussed critically and new insights were given to this innovative research field.

Lessons from actual studies

The silent way of osteoarthritis

OA is not limited to articular cartilage only, but also affects the subchondral bone, as well as the adjacent connective tissue and the synovial membrane resulting in pain, swelling, progressive deformity and instability. Although OA occurs in many joints, the knee, hip, hand and facet joints are mostly affected.5 Yet, patients suffering from OA often show a discrepancy between objective findings gathered in x-ray or magnetic resonance imaging (MRI) examinations and patient-reported pain and functional restrictions.6 The sudden onset of disabling joint pain and articular effusion is one typical clinical feature of decompensation indicating the term activated OA. On a cellular and subcellular level, proinflammatory agents such as nitric oxide, interleukin 1 (IL-1), and tumor necrosis factor (TNF) α are overexpressed in chondrocytes and joint stromal cells in OA.7,8

The influence of free radicals on osteoarthritis development

Many studies focused on damaging effects of oxidative and nitrosative agents,8,9 whereas in the following the differential role of Radical oxygen species (ROS) and radical nitric species (RNS) in osteoarthritis came into focus. A study comparing the effects of nitric oxide and oxidative dysbalance in OA and rheumatoid arthritis postulated a major role of nitric oxide (NO) to modulate chondrocyte function in OA.10 In contrast to the uncoupling effect of ROS on synovium inflammation,11 ROS have been shown to downregulate the expression of pro-inflammatory genes in

Correspondence: Christoph Ziskoven, MD Orthopedic Department, Heinrich-Heine-University Medical School, Moorenstr. 5, D-40225 Düsseldorf, Germany. E-mail: christoph.ziskoven@med.uni-duesseldorf.de

Key words: osteoarthritis, free radicals, oxidative damage.

Contributions: all work performed in the writing and research process of this paper was done by scientific assistants (CZ, KB, MJ) and the heads of the departments of the Heinrich-Heine University Medical School (RK) and the German Sport University (WB). These institutions are financed by public funds; no additional private funding was required.

Conflict of interest: the authors report no conflicts of interest.

Received for publication: 21 October 2010. Accepted for publication: 29 November 2010.

This work is licensed under a Creative Commons Attribution 3.0 License (by-nc 3.0).

©Copyright C. Ziskoven et al., 2010 License PAGEPress, Italy Orthopedic Reviews 2010; 2:e23 doi:10.4081/or.2010.e23
The role of nitrosative and oxidative stress in the three compartments of joint

There is evidence that alterations of subchondral bone\textsuperscript{39} and synovial cells/fluid are crucial for OA pathogenesis, which stands in contrast to the classic focus on cartilage degradation in OA development. The joint is hence to be considered as a dynamic system consisting of three discrete but permanently interacting compartments.\textsuperscript{22}

Cartilage tissue is classified with conjunctive tissue, deriving from mesenchymal stem cells during embryonic development. Oxidative dysbalance in cartilage is an effect of two major mechanisms. First, it occurs when antioxidative capacity is depleted. A more oxidative intracellular state is a cause for pathologic alterations in osteoarthritic cartilage.\textsuperscript{21}

Second, nutrition of articular cartilage as avascular tissue containing chondrocytes embedded into a large amount of extracellular matrix is dependent on diffusion from synovial- and subchondral bone compartment.\textsuperscript{23} Oxygen and metabolic end products have to diffuse over relative long distances, resulting in low O\textsubscript{2} tension in cartilage tissue.\textsuperscript{24} A rise of O\textsubscript{2} tension in cartilage tissue as is typical for inflammation in turn enhances free radical formation, an interesting finding in the context of cartilage degeneration in chronically irritated joints.\textsuperscript{25} The upregulation of iNOS related NO production is a well known feature of IL-1.\textsuperscript{26} Nitric oxide was suspected to play a major role in the influence of mitochondrial function in OA (Figure 1). Furthermore, NO dependent intracellular signaling in OA nociception is subject of recent investigation.\textsuperscript{27}

In contrast, excessive repetitive load was proven to induce chondrocyte apoptosis, a fact which points out the relevance of injurious cartilage compression for OA pathogenesis.\textsuperscript{1,27} Mechanical loading that exceeds the tolerance of the articular surface was indirectly identified to cause oxidative stress by the decrease of cell death after submission of antioxidants.\textsuperscript{3,4} A study on cartilage explants showed an increase of ROS synthesis after admission of mechanical load.\textsuperscript{1,4} Furthermore, some data indicate that radical nitrogen species act as mediators of cell death after mechanical overuse and injury to cartilage.\textsuperscript{4} These findings are in accordance to the fact that also excessive shear stress is able to induce chondrocyte death mediated by ROS.\textsuperscript{39}
The synovial compartment consists of the synovial membrane and the synovial fluid. The implications of this joint compartment for OA development, also in relation with ROS, were extensively investigated during the last decade. The synovial cells are involved in the synthesis of the synovial fluid, an ultrafiltrate of the serum enriched with products of the cells. It forms a capillary layer in the healthy joint and provides the minimization of friction. Here, a large amount of cross linked hyaluronan, an anionic non-sulfated glycosaminoglycan, contributes substantially to the rheologic properties of joint fluid. Oxidative stress, namely peroxynitrit, has been shown in vitro to depolymerise glycosaminoglycans, whereas hyaluronan degradation can be prevented by antioxidative agents. In conclusion, hyaluronan acts as a ROS scavenger in the joint by a competition with other substrates suitable for the reaction with radical oxygen molecules.

A reduced antioxidative capacity in synovial fluid of osteoarthritic joints has been found, investigating mainly extracellular superoxide dismutase (EC-SOD or SOD3) in irritated versus late stage OA joints. Here, the reduced antioxidative capacity was described as a disability of the joint to adapt to increased oxidative stress in late stage OA. Finally, the question if oxidative stress is an initial cause or secondary consequence of OA remains unanswered.

The inflammatory transformation of synovial membrane in OA is observed frequently in arthroscopic examination. Distinct synovitis involving infiltration of activated B cells and T lymphocytes and overexpression of proinflammatory mediators is a common finding in histological and arthroscopic examination of patients suffering from OA. Increased levels of IL-1α, IL-1β, and TNF-α could be found in synovial membranes from patients with OA.

Radical oxygen species play a major role in inflammatory intracellular signaling mechanisms of synovia. The activation of nuclear transcription factor (NF)-κB that is implicated in the inflammatory response in vascular endothelium and type A synovial lining cells is a feature of synovial tissue from both RA and OA patients. Furthermore, synovitis is secondary to a local increase of the pro-inflammatory enzymes cyclooxygenase-2 and iNOS. NO mediates the effects of a number of proinflammatory cytokines, including interleukin-1 (IL-1) and tumor necrosis factor (TNF)-α. A canine in vivo model demonstrated a NO dependent up-regulation of IL-1-converting enzyme in chondrocytes, providing another interrelationship between synovial inflammation and cartilage degradation by paracrine free radical signaling.

One possible mechanism leading from inflammatory transition of the synovial membrane to joint degeneration is ROS/RNS production by synovial cells (Figure 2). Synovial inflammation was discussed to impair the ability of chondrocytes to balance anabolic and catabolic activities in extracellular matrix formation. A transgenic mouse model yielded evidence that increased ROS production is a relevant cause for chondrocytic death and activation of matrix metalloproteinases in an inflammatory joint disease.

At third the subchondral bone is involved in ROS mediated OA pathogenesis. In advanced stages of OA, first hypertrophic alterations of the bone presented by subchondral sclerosis and osteophytes occur; later on osteoclast activation leads to local bone resorption (bone cysts) affecting the peri-articular bone. The subchondral bone together with articular cartilage is considered as a unit, playing an important role in OA disease development particularly on the bounding surface between bone and cartilage. Only few studies have focused on the action of radical oxygen species in the context of subchondral bone remodeling. One study demonstrates the effect of total fraction of avocado/soybean unsaponifiables on bone remodeling in experimental canine osteoarthritus. Here, after drug admission a significantly reduced expression of inducible nitric oxide synthase in cartilage together with decreased bone remodeling is reported. The exact mechanisms of bone remodeling in relationship with ROS action are only partially understood.

The implication of cellular senescence, apoptosis and ageing in osteoarthritis

The heterogeneity of OA leads to a huge variety of known etiopathogenic factors. Yet, in literature review there is a common thread regarding the uniform presentation of end stage OA. There are mainly three pathologic alterations in joint that represent OA disease development on the cellular level. Cellular senescence, apoptosis and the effects of ageing show a relationship with OA disease progression in all joint compartments. Here, ROS/RNS have been proven to be involved in the regulation of each of these factors.

Preterm cellular senescence has been addressed to play an important role in OA. The phenomenon of cellular senescence has been described for the first time by Hayflick in the 60’s of the last century as the inability of matured cells to divide indefinitely. This Hayflick limit is mainly an effect of telomere shortening. Linear chromosomes are capped by repetitive nucleoprotein structures, called telomeres. Each cell division results in a progressive shortening of telomeres that, below a certain threshold, promotes genome instability, senescence, and apoptosis. Telomeres are highly sensitive against ROS induced instability due to their high content of guanines. Therefore, telomere length is reducing at a faster rate during oxidative stress. There is evidence that senescence can be induced by extrinsic factors in chondrocytes by so called free radical induced stress senescence. Searching a specific marker for chronic exposure to oxidative stress, irreversible telomere length shortening was discussed. These facts imply a connection between oxidative stress and OA development. Yet, to date the amount of data on the impact of telomere length shortening under in vivo conditions in tissues other than the hematopoietic system is limited.

There are two clinical presentations of OA that are possibly associated with free radical
induced cellular senescence:

First, there is few data on the exact mechanisms that promote joint capsule thickening by fibrosis and consecutive stiffness. The cellular mechanism of fibrosis control is regulated by cellular senescence in at least one tissue. Since fibrosis is a common feature of OA, intraarticular paracrine ROS/NOS messaging might be beneficial for limitation of capsule fibrosis by promoting senescence of proliferating fibroid cells.

Second, joint inflammation is another characteristic finding in OA with often acute onset, leading to pain and swelling of the affected joint. Beside immunogenic cells a huge number of other cell types are involved in inflammation, in the context of OA pathogenesis particularly fibroblasts. The first study showing an inflammatory response by mesenchymal cells was performed on skin fibroblasts. The inflammation involved the transcriptional upregulation of cytokines, such as interleukins (IL-1, IL-15), their receptors and chemotactic secreted factors. This coordinat- ed secretion of proinflammatory molecules is a result of cellular senescence. It is likely that ROS induced senescence of synovial and cartilage cells can promote the inflammatory transition of the osteoarthritic joint.

Free radical damage induced cell apoptosis provides a strategy to ensure the survival of the organism by scavenging of damaged cells. The process of apoptosis is initiated by activation of caspases (cysteine-aspartic-acid-pro- teases), a process that what long time considered to be is irreversible once started. Free radical species have been shown to promote apoptosis by activation of c-Jun N-terminal kinases (JNK) and p38 Mitogen-activated protein (MAP) kinases via apoptosis signal-regulating kinase (ASK) 1. Interestingly, within this pathway free radicals are formed as a second messenger amplifying ASK1 activation leading to cell death within ROS concentrations that are not sufficient to initiate apoptosis.

Today, chondrocytic death in OA has been examined in various studies, most of which report an increased amount of apoptotic cell death. Besides other stimuli, exogenous and endogenous NO has been proven to induce chondrocyte apoptosis by intracellular induction of ROS. The cytotoxic effect is a result of increased ROS load and formation of cytotoxic peroxynitrit by direct reaction of NO and ROS. Visc versa, chondrocyte apoptosis induced by toxins can be suppressed by hypoxia shifting chondrocytes to a less oxidative intracellular state. An accumulation of cartilage matrix proteins in the endoplasmic reticulum and Golgi apparatus of chondrocytes modified by oxidant stress during aging has been discussed as a cause for decreased synthesis of cartilage matrix proteins and eventu- al chondrocyte apoptosis.

Most studies on apoptosis and OA are focused on cartilage tissue, only few data exist about the relevance of cell death in the other joint compartments. Furthermore, most studies are performed in advanced OA stages on cartilage explants obtained during total joint replacement surgery. In contrast, little is known about the function of apoptosis in early stage OA.

Aging leads to impaired extracellular matrix composition. A dysbalance of chondrocyte function in favor of catabolic action against extracellular matrix synthesis increases within aging. Studies measuring changes in joint cartilage using MRI gathered evidence that its thickness decreases with significant age dependency. Also hydration, decline of cellularularity and a gradual loss of cartilage matrix are reported in aged cartilage leading to a weakening of tissue.

Interestingly, the joint is capable of adapting to oxidative stress induced by extrinsic factors if it is not subject to pathologic alterations. Repetitive and cyclic mechanical stress, e.g. in long distance running does not induce progressive knee OA. The crucial biomechanical cause seems to be excessive load within overweight or joint malalignment, or the combination of such factors.

As a result of the slow metabolism and low proliferation rate of cartilage tissue chondrocytes and cartilage extracellular matrix collagens lack the turnover most tissues show. A theory of aging with relevance for cartilage degeneration is the assumption that cells gather damage by free radicals produced by the electron transport chain in mitochondria. Cartilage tissue is highly sensitive against cumulative effects of extrinsic factors like oxidative stress. There is evidence of the accumulation of nonenzymatic glycated and oxidized proteins, so called advanced glycation endproducts (AGEs) in joint cartilage. The production of AGEs is increased in diabetic individuals. The influence of AGE accumulation on chondrocytes is mediated by specific receptors for advanced glycation endproducts (RAGE) located in the membrane of chondrocytes. They show an increased expression in aging and during OA development. Stimulation of these receptors leads to an increased expression of matrix metalloproteinases followed by increased catabolic function of chondrocytes. The intracellular messaging of RAGE has early been proven to be ROS dependent. RAGE activation in turn induces oxidative stress, showing one more involvement of oxidative stress in pathologic alterations leading to joint degradation.

Resulting from the clearly demonstrated age dependency of disease development, biochemical factors influencing chondrocyte function and survival may play a central role in the pathophysiological cascade leading to OA and provide therefore therapeutic options on long-term.

Discussion

The progression of OA from silent cartilage destruction to painful clinical presentation is an important subject to further investigation. The novel approach to OA considers the joint as a dynamic system of three different tissues interacting by fluid diffusion and paracrine factors. Because of their chemical properties, free radicals meet the requirements to mediate and amplify the characteristic sequence of joint degeneration in all tissues affected, making them as well a crucial factor for the involvement of all three joint compartments in disease development as for the sudden breakdown of compensation leading to inflammatory transformation of osteoarthritic joints. Therapeutic modifications of these mechanisms may be promising area of further research to achieve comprehension and therefore approach to modification of disease progression.

References

8. Ghiz S, Nietfeld JJ, Duits AJ, et al. Elevated synovial fluid levels of inter-


90. Harman D. The biologic clock: the mito-
94. Dettoraki A, Gil AP, Spiliotis BE. Association between serum levels of the soluble receptor (sRAGE) for advanced glycation endproducts (AGEs) and their receptor (RAGE) in peripheral blood mononuclear cells of children with type 1 diabetes mellitus. J Pediatr Endocrinol Metab 2009;22:895-904.