INTRODUCTION

Osteoarthritis is the most common disease of joints. It is a group of overlapping distinct diseases, which may have different etiologies but with similar biological, morphological and clinical outcomes. The disease process not only affects the articular cartilage, but also involves the entire joint including the subchondral bone, ligaments, capsule, synovial membrane and peri-articular muscles. Ultimately, the articular cartilage degenerates with fibrillation, fissures, ulceration and full- thickness loss of the joint surface with abnormal new bone formation ***(Carr, 2013).***

Osteoarthritis is the most common form of arthritis and the leading cause of chronic disability ***(CDC, 2001)***. The estimated population prevalence varies from 4% to 30% depending on age, gender, and disease definition. Overall osteoarthritis affects 13.9 % of adults aged 25 and older and 33.6 % of those 65 and older ***(Lawrence et al., 2008).***

Osteoarthritis is a complex disease of the musculoskeletal system with both genetic and environmental risk factors ***(Valdes and Spector, 2011)***. From the results of heritability studies in twins, sibling pairs, and families, genetic factors are estimated to account for about 50% of the risk of developing osteoarthritis in the hip or knee, although precise estimates vary according to sex, affected site, and severity of disease ***(Valdes et al., 2010).***

Novel targets in OA include genes that are involved in OA pathophysiology, which have been discovered using gene network, epigenetic and microRNA (miRNA) approaches. Further insights into the molecular mechanisms involved in OA initiation and progression may lead to the development of new therapies that can control joint destruction and stimulate repair ***(Alcaraz et al., 2010).***

Molecular mechanisms that regulate the levels of proteins relevant to OA are potential targets for rational therapeutic intervention. Thus, there is a continuing need to deepen our understanding of the molecular mechanisms in OA and to investigate potential molecular targets for its treatment ***(YU et al., 2011)***.

Several genes have been associated with osteoarthritis susceptibility including GDF5, chromosome 7q22, and MCF2L. A recently identified compelling association is with the Nucleostemin-encoding gene which has been linked to osteoarthritis at a genome wide significance levels in European populations ***(Zeggini et al., 2012).***

The Nucleostemin gene (*NS*, also known as *GNL3*) was discovered during a search for genes that are preferentially expressed in neural stem cells ***(Tsai and McKay, 2002)***. It encodes a GTP binding protein (NS) that resides principally in the nucleolus, but can apparently shuttle to and from the nucleoplasm in response to various cues ***(Meng et al., 2008)***.

Nucleostemin (NS) has been shown to regulate cell cycle progression. NS possesses two GTP-binding domains and can be readily detected in neural stem cells, embryonic stem cells, hematopoietic primitive cells, and tumor cell lines ***(Tsai and McKay, 2002).*** However, it has been demonstrated that *Ns* is in fact widely expressed in many types of normal proliferating cells at levels similar to those in malignant cells ***(Fan et al., 2006).*** *GNL3* is expressed in mesenchymal stem cells, from which chondrocytes are derived, and regulates the G1–S phase transition in stem cells ***(Zeggini et al., 2012).***

AIM OF THE WORK

To quantify RNA expression of the recently identified Nucleostemin gene in the synovium and/or synovial fluid of patients with primary osteoarthritis and to correlate the level of RNA expression to the radiographic grading of the disease, thus identifying a new path that could be amenable to future therapeutic intervention.

OSTEOARTHRITIS

Background

Osteoarthritis is the most common type of joint disease. It represents a heterogeneous group of conditions resulting in common histopathological and radiological changes. It can be thought of as a degenerative disorder arising from biochemical breakdown of articular (hyaline) cartilage in the synovial joints. However, the current view holds that osteoarthritis involves not only the articular cartilage but also the entire joint organ, including the subchondral bone and synovium ***(Carr, 2013).***

Osteoarthritis predominantly involves the weight-bearing joints, including the knees, hips, cervical and lumbosacral spine. Other commonly affected joints include the distal interphalangeal (DIP), proximal interphalangeal (PIP), and carpometacarpal (CMC) joints ***(Sharma, 2001).***

Although osteoarthritis was previously thought to be caused largely by excessive wear and tear, increasing evidence points to the contributions of abnormal mechanics and inflammation. Therefore, the term degenerative joint disease is no longer appropriate in referring to osteoarthritis ***(Poole, 1999).***

Osteoarthritis has been classified into primary and secondary forms. Secondary osteoarthritis is conceptually easier to understand: It refers to disease of the synovial joints that results from some predisposing condition that has adversely altered the joint tissues (e.g., trauma to articular cartilage or subchondral bone). Secondary osteoarthritis can occur in relatively young individuals ***(Valderrabano et al., 2009).***

The definition of primary osteoarthritis is more nebulous. Although this form of osteoarthritis is related to the aging process and typically occurs in older individuals, it is, in the broadest sense of the term, an idiopathic phenomenon, occurring in previously intact joints and having no apparent initiating factor ***(Loughlin, 2005).***

No specific laboratory abnormalities are associated with osteoarthritis. Rather, it is typically diagnosed on the basis of clinical findings, with or without radiographic studies ***(Felson, 2009).***

The goals of osteoarthritis treatment include pain alleviation and improvement of functional status. Nonpharmacologic interventions are the cornerstones of osteoarthritis therapy including patient education, weight loss, exercise and physical therapy. Pharmacological therapy whether oral or intra-articular can provide pain relief and have an anti-inflammatory effect on the affected joint. If all other modalities are ineffective and osteotomy is not viable, or if a patient cannot perform his or her daily activities despite maximal therapy, arthroplasty is indicated ***(Kirkley et al., 2008).***

The high prevalence of osteoarthritis entails significant costs to society. Direct costs include clinician visits, medications, and surgical intervention. Indirect costs include such items as time lost from work. Costs associated with osteoarthritis can be particularly significant for elderly persons, who face potential loss of independence and who may need help with daily living activities. As the populations of developed nations age over the coming decades, the need for better understanding of osteoarthritis and for improved therapeutic alternatives will continue to grow ***(Losina et al., 2011).***

Anatomical considerations

A structural classification would categorize joints as synovial, fibrous and cartilaginous. This classification is based on the type of connecting tissue and the presence or absence of a joint cavity ***(Drake et al., 2009).*** Reviewing the anatomy of the synovial joint is a must for thorough understanding of osteoarthritis.

Synovial joints possess a cavity and the articular ends of bones forming the joint are enclosed in a fibrous capsule. As a result they are separated by a marrow cavity. The articular cavity (joint cavity) is filled with a fluid called synovial fluid and hence the name synovial joint. The synovial joints are the most evolved and freely movable joints ***(Singh, 2008).***

The characteristic features of synovial joints are as follows ***(Singh, 2008)*****:**

1. The articular surfaces are covered by a thin plate of hyaline cartilage.
2. The cavity of the joint is lined everywhere by synovial membrane except over the articular cartilages.
3. The cavity is filled with synovial fluid secreted by synovial membrane.
4. Some additional structures are present within the joint cavity, e.g.: articular discs, tendons and fat pads.

<u>Components of synovial joints and their functional significance:</u>

1. Articular capsule:

It consists of outer fibrous capsule and inner synovial membrane.

Fibrous capsule:

It completely invests the joint like a sleeve and encloses the synovial cavity. It is attached by continuous lines to the bones forming the joint close to the articular cartilages. It consists of longitudinal and interlacing bundles of white connective tissue which are sensitive to changes in position of the joint ***(Rawlani and Rawlani, 2011).***

Function ***(Singh, 2008)***:

a) The fibrous capsule stabilizes the joint in such a way that it permits movement but resists dislocation.

b) Numerous sensory nerve endings ramify on the capsule. The stimulation of these nerve endings produce reflex contraction of muscles acting in the joint in such a way that the joint is brought in a position of maximum comfort to protect the joint.

Synovial membrane:

It is a thin highly vascular membrane of connective tissue lining the inside of the fibrous capsule. It is reflected from the fibrous capsule onto the bones lying within the capsule but ceases at the periphery of the articular cartilage, articular disc or meniscus ***(Drake et al., 2009).***

Function:

The synovial membrane produces synovial fluid in sufficient quantity to keep the surfaces properly lubricated. Synovial fluid is formed through a serum ultrafiltration process by cells that form the synovial membrane (synoviocytes). Synovial cells also manufacture hyaluronic acid (HA, also known as hyaluronate), a glycosaminoglycan that is the major noncellular component of synovial fluid ***(Tortora and Derrickson, 2009)***.

Function of synovial fluid ***(Rawlani and Rawlani, 2011):***

- Synovial fluid supplies nutrients to the avascular articular cartilage.
- It provides the viscosity needed to absorb shock from slow movements, as well as the elasticity required to absorb shock from rapid movements.
- Synovial fluid also removes particulate matters and worn out cartilage cells by the phagocytic activity.

2. Articular cartilage:

The articular cartilage of most joints is hyaline in structure with few exceptions in which it is composed of fibro cartilage .The cartilage contains no intrinsic blood vessels. It receives its nutrition from the synovial fluid ***(Drake et al., 2009).***

Function:

It provides a smooth gliding surface and reduces forces of compression during weight bearing and muscle action. The cartilage facilitates joint function and protects the underlying subchondral bone by distributing large loads, maintaining low contact stresses, and reducing friction at the joint ***(Rawlani and Rawlani, 2011).***

Articular cartilage is a highly specialized and uniquely designed biomaterial that is synthesized by the sparsely distributed resident cells-the chondrocytes. The greatest portion of articular cartilage mass is composed of a macromolecular extracellular matrix surrounding the chondrocytes. The cells represent less than 5% of the total volume of articular cartilage but are of obvious importance for the maintenance of the tissue ***(Ghadially, 1993).***

Normal mature articular cartilage is divided histologically into four distinct zones based on the arrangement and orientation of its cellularity. The most superficial zone, closest to the articulating surface, is the tangential zone. Here the flattened cells have their long axes parallel to the articular surface. Immediately beneath the tangential zone, the cells are more rounded and are randomly oriented. This is the transitional zone. The chondrocytes in this area are metabolically active, producing collagen and proteoglycan.

Beneath this region is the radial zone in which cells are arranged in short columns perpendicular to the articular surface. These cells are smaller than those immediately above and appear less active. The calcified zone, with its pyknotic cells surrounded by calcifying matrix, resides deep to the radial zone ***(Ghadially, 1993)***.

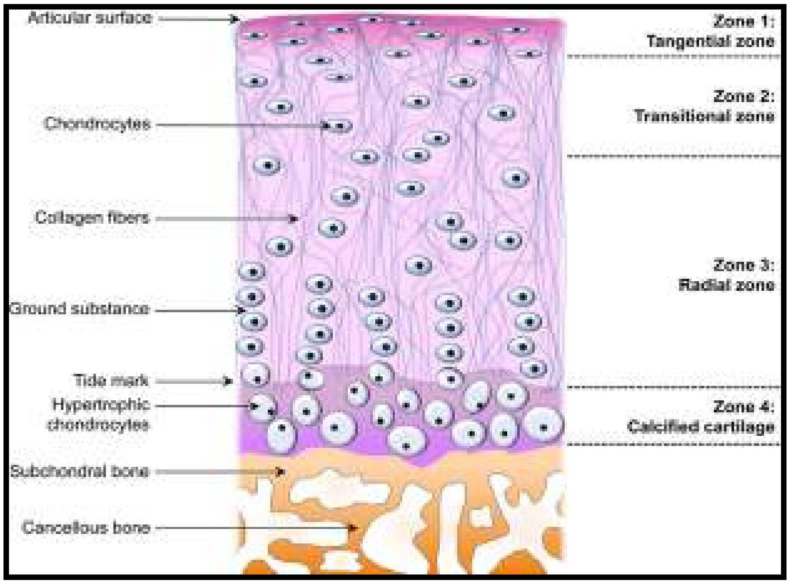

Figure (1): Histological structure of mature articular cartilage ***(Angele et al., 2004)***

Cartilage matrix consists of two basic components: a fibrillar and an extrafibrillar matrix .The fibrillar matrix is a network consisting mainly of collagen II together with other collagens, predominantly IX, XI, and XVI. The non-fibrillar component of hyaline articular cartilage consists predominantly of aggrecan-hyaluronan aggregates ***(Aigner and Schmitz, 2011)***.

Aggrecan is a high molecular weight proteoglycan. It exhibits a bottlebrush structure, in which chondroitin sulfate and keratan sulfate chains are attached to an extended protein core. In the matrix, aggrecan is arranged in high-molecular-weight aggregates formed by noncovalent association between proteoglycan subunits, hyaluronic acid and linkage protein ***(Nap and Szleifer, 2008).***

In terms of the physical properties of the cartilage matrix, tensile strength comes from the collagen network, which hinders expansion of the viscoelastic aggrecan component and, thus, provides compressive stiffness of the tissue. On the other hand, the aggrecan-hyaluronan aggregates bind high amounts of intercellular water owing to their extensive fixed charges and are responsible for the elasticity of the tissue. Thus, under compression, the cartilage matrix is compliant but rapidly regains its elasticity as water molecules are drawn back into the matrix on unloading by the strongly hydrophilic aggrecan aggregates ***(Aigner and Schmitz, 2011)***.

3. Articular disc or meniscus:

The articular discs are pads of fibrocartilage interposed between the articular surfaces of some joints, e.g. knee joint ***(Singh, 2008).***

Function ***(Singh, 2008):***

a) Helps in lubrication of a joint by maintaining an interval between the articular surfaces.

b) Divides the joints into compartments.

c) Acts as a ligament to modify certain movements.

d) Prevents wear and tear of articular cartilages by providing a cushioning effect**.**

4. Ligaments:

These are thickened bands of collagen fibers. They are of two types ***(Rogers, 2010)*****:**

a) True ligaments: they are local thickenings of parallel fiber bundles of capsular ligament, also called intrinsic ligaments e.g. medial and lateral collateral ligaments of knee joint.

b) Accessory ligaments which are separate from fibrous capsule and may be extracapsular or intracapsular. Anterior and posterior cruciate ligaments are examples of intracapsular accessory ligaments.

Function:

True ligaments permit the movements in one plane and prevent unwanted movements in another plane. They also stabilize the joint ***(Rogers, 2010).***

5. Bursae:

These are pouch-like sacs of connective tissue filled with synovial fluid, found near synovial tissues.

Function: The bursae cushion certain muscles and facilitate the movement of tendons or muscles over bony or ligamentous surfaces ***(Rawlani and Rawlani, 2011).***

6. Fat pads:

These are pads of fat placed between synovial membrane and fibrous capsule or between synovial membrane and bone e.g. infrapatellar fat-pad of knee joint.

Function: as fat is very pliant, the pads can accommodate themselves to changing condition of the joint during different movements ***(Singh, 2008).***

Pathophysiology

Traditionally, osteoarthritis was thought to affect primarily the articular cartilage of synovial joints; however, pathophysiologic changes are also known to occur in the