Hydroxyproline as a New Biomarker to Differentiate Osteoarthritis from Rheumatoid Arthritis in the Early Stages

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Article

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Abstract

Background: Osteoarthritis (OA) and rheumatoid arthritis (RA) are the most common inflammatory disorders of the musculoskeletal system which are defined by articular cartilage degradation and impairment of joint. When criteria and radiography are insufficient to diagnose early-stages of RA and OA to control disease course, biomarkers become necessary to early diagnosis these diseases.

Objective: We searched the levels of (MMP3) and (HP) in the blood of early RA and OA patients in order to see if they can be utilized to distinguish the two diseases at an early stage.

Patients and Methods: We recruited 1) forty patients who were diagnosed with early knee osteoarthritis using EULAR and Kellgren-Lawrence radiographic grading criteria, 2) forty patients with early RA (disease duration less than three years), and 3) forty control group consisted of normal volunteers who were age and sex matched. All subjects underwent a thorough medical history, clinical musculoskeletal examination, and laboratory tests, including the determination of blood MMP3 and HP. We did also a simple x-ray of their hands and knee joints.

Results: Comparing groups, we found a highly significant increase in serum MMP3 in OA patients than RA patients and control groups. MMP3 was also significantly higher in RA patients than in the control group. While, OA patients had significantly higher HP than RA patients and control groups, whereas RA patients and control groups had no significant difference in HP.

Conclusion: Hydroxyproline rather than MMP3 may be employed as a potential biomarker for early differentiation between osteoarthritis and rheumatoid arthritis.

Key Messages

1. Early RA presents with symptoms that are similar to those seen in other inflammatory diseases.
2. Patients with early RA are frequently characterized as undifferentiated arthritis, which is difficult to distinguish from other inflammatory arthritis until a definitive diagnosis.
3. Both Rheumatoid arthritis (RA) and osteoarthritis are marked by the deterioration of articular cartilage and the impairment of joint function (OA).
4. When diagnostic criteria are not met, blood levels of Hydroxyproline (HP) rather than MMP3 could be employed as a possible biomarker for early differentiation between osteoarthritis (OA) and rheumatoid arthritis (RA).

Introduction

The most common disease of the musculoskeletal (MSK) system is osteoarthritis (OA). Every year, millions of people are affected with OA. Symptomatic knee OA has been documented in 6–10% of the adult population, making it one of the most prevalent joints involved in OA (1).

Because clinical and radiographic evidence are insufficient to diagnose early-stage OA and predict disease progression, biomarkers that assist doctors in early diagnosis, assessing disease activity, predicting prognosis, and monitoring response to medication are critical (2).

Rheumatoid arthritis (RA) is an autoimmune disease that causes a chronic, systemic inflammatory disorder that can affect many tissues and organs, but primarily affects synovial joints, causing synovial hypertrophy and chronic joint inflammation, as well as extra-articular manifestations, and is thought to affect genetically susceptible people (3)

Early RA presents with symptoms that are similar to those seen in other inflammatory diseases. Patients with early RA are frequently characterized as undifferentiated arthritis, which is difficult to distinguish from other inflammatory arthritis until a definitive diagnosis. Early RA was previously defined as patients with a disease duration of less than two years. (4) Both Rheumatoid arthritis (RA) and osteoarthritis are marked by the deterioration of articular cartilage and the impairment of joint function (OA). (5)

Chondrocytes and extracellular matrix make up articular cartilage (ECM). Normal cartilage's ECM is in a state of dynamic balance between collagen synthesis and breakdown. This equilibrium is tipped in favour of proteolysis in RA and OA disorders, which is linked to pathologic cartilage loss. (6)

The major enzymes responsible for this destruction and collagen degradation are matrix metalloproteinases (MMPs), which are secreted by chondrocytes (7, 8)

Because MMP-3 is produced by synovial fibroblasts of rheumatoid joints and not by normal synovial cells, it has been claimed that it is a sign of synovitis coming from the joint. (9, 10)

Hydroxyproline is one of the particular amino acids found in collagens, which serve as MMP substrates. The measurement of hydroxyproline can help with the diagnosis and prognosis of diseases caused by problems with collagen metabolism (11).

The goal of this study is to determine MMP3 and HP levels in early knee osteoarthritis and early RA patients in the hope of discovering biomarkers that may aid doctors in early diagnosis, disease activity assessment, prognosis prediction, and therapy response monitoring.

Patients And Methods

This research looked at:

- Forty patients who were diagnosed with early knee osteoarthritis using EULAR and Kellgren-Lawrence radiographic grading criteria.

    1. Forty patients who were diagnosed with early knee osteoarthritis using EULAR and Kellgren-Lawrence radiographic grading criteria.
    2. Forty patients with early RA (disease duration less than two years):

3. Forty normal volunteers control group who were age and sex matched.

All patients and controls were recruited from Al-Zahra University Hospital's Rheumatology and Rehabilitation outpatient department.

This study was carried out according to regulations and approval of Ethics Committee of Faculty of Medicine for Girls, Al-Azhar University, Nasr City, Cairo, Egypt, Registered at Central Administration of Research & Development; Egyptian Ministry of Health: Reg No. RHBIRB2018122001.

We obtained informed ethical consents from all patients to participate in this study, as well as its publication according to the recommendations of the above-mentioned committee.

All subjects underwent a thorough medical history, clinical musculoskeletal examination, and laboratory tests, including the determination of blood MMP3 and HP using the Sandwich ELISA technique. They were also given a simple plain x-ray of their hands and knee joints.

All OA and RA patients were chosen based on the disease's inclusion and exclusion criteria.

Results

We discovered a highly significant increase in serum MMP3 in OA patients when compared to RA patients and control groups,. MMP3 was also significantly higher in RA patients than in the control group ($P < 0.001$). Meanwhile, we discovered that OA patients had significantly higher HP than RA patients and control groups ($P < 0.001$), whereas RA patients and control groups had no significant difference in HP ($P > 0.05$).

Discussion

Osteoarthritis (OA) is a chronic, degenerative osteoarthropathy characterised by joint pain, soreness, stiffness, swelling, function limitations, and deformity. Excessive degradation occurs in OA, resulting in the progressive loss of matrix proteins and joint integrity (12).

Rheumatoid arthritis, on the other hand, is a chronic inflammatory joint condition that can lead to cartilage and bone loss, as well as disability. Early diagnosis is critical for maximum therapy effectiveness, especially in individuals who have well-defined risk factors for poor outcomes, such as high disease activity, autoantibodies, and early joint injury. (3).

Many individuals with early disease symptoms do not meet diagnostic criteria for any of the diseases, posing a difficulty in disease prescribing and management. As a result, we set out to find a biomarker that could help identify both diseases in their early stages.

Many studies have shown that serum MMP3 levels are higher in OA patients than in healthy people (13–17), and we observed that serum MMP3 levels were considerably higher in knee OA patients than in

healthy people.

MMP3 levels were also observed to be significantly higher in RA patients than in healthy people.

As a result, MMP3 was unable to distinguish between the two diseases in the early stages, despite being considerably higher in OA patients than in RA patients.

We detected a highly significant increase in serum hydroxyproline (HP) in OA patients compared to both RA and normal controls, which is consistent with earlier studies. (18, 19)

In the meantime, there was no discernible change in HP serum levels between RA patients and healthy controls.

Others have observed that plasma hydroxyproline levels were elevated 44 percent in early OA and 58 percent in other non-RA conditions, but not in early RA as compared to healthy controls.

(20)

Conclusion

When diagnostic criteria are not met, blood levels of Hydroxyproline (HP) rather than MMP3 could be employed as a possible biomarker for early differentiation between osteoarthritis (OA) and rheumatoid arthritis (RA).

Declarations

Competing interests:

- The author confirms that there are no any potential conflicts of interest.

Contributorship:

1-Adel Elbeialy (AE)

2-Hemmat Elabd (HE)

3-Amira Shahin (AS)

4-Radwa Ibrahim (RI)

The codes, supervised the research.

AE, HE, AS, and RI performed the experiments.

AE, HE, AS, RI wrote and revised the manuscript.

AE is the corresponding author

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- I confirm also that there are not any financial support or other benefits from commercial sources for the work reported on in the manuscript, or any other financial interests that I may have, which could create a potential conflict of interest or the appearance of a conflict of interest with regard to the work.

Ethical approval information:

- This study was carried out according to regulations and approval of Ethics Committee of Faculty of Medicine for Girls, Al-Azhar University, Nasr City, Cairo, Egypt, Registered at Central Administration of Research & Development; Egyptian Ministry of Health: Reg No. RHBIRB2018122001
- Informed ethical consents were obtained from all patients to participate in this study, as well as its publication according to the recommendations of the above-mentioned committee.

Data sharing statement:

- I confirm hereby that the manuscript has not been submitted or is not simultaneously being submitted elsewhere, is not at the time of submission under consideration by another journal or other publication, and that no portion of the data has been or will be published elsewhere while the manuscript is under review by the journal, unless rejected or withdrawn by the author. Also we confirm that no portion of the data has been or will be published elsewhere while the manuscript is under review by the journal.

Data Availability:

- The datasets used and/or analyzed during the current study available from the corresponding author on reasonable request.

Patients and Public involvement

- I confirm here that all patients are fully anonymized and unidentifiable in any way, and they gave us informed written consent that they agree to publish this research in its present form.

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Table 1

Table (1): Serum levels of MMP3 and HP in OA and RA groups.

"Enzyme"	OA *(n=40)*	RA *(n=40)*	Control *(n=40)*	p-value
MMP3 pg/mL	559.92±112.84	153.25±162.05		**<0.001**
MMP3 pg/mL		153.25±162.05	59.79±63.54	**<0.001**
HP µg/mL	12.87±18.75	4.81±6.89		**<0.001**
HP µg/mL		4.81±6.89	4.52±1.55	**> 0.05**