Lipid-peroxidation and Antioxidant Status in Osteoarthritis and Rheumatoid Arthritis Patients

Shweta Dwivedi[1], Shreesh Singh[2], Geeta Jaiswal[2]

ABSTRACT

Introduction: Rheumatoid arthritis (RA) and Osteoarthritis is a chronic multisystem disease of unknown cause. The pathogenesis of this disease is due to the generation of ROS and RNS at the site of inflammation. Increased oxidative stress and decreased antioxidant status are the hallmark of these diseases. The objectives of the study was to evaluate the lipid-peroxidation level by measuring malondialdehyde, superoxide dismutase, cerulloplasmin and non - enzymatic antioxidant (Vitamin E and Vitamin C) status in patients of RA and OA and compare with normal individuals.

Material and Methods: In the present study we took 100 rheumatoid arthritis (RA), 100 osteoarthritis (OA) patients and 100 healthy individuals. Serum Lipid-peroxidation were measured by blood hemolysate by Utley's method, Superoxide dismutase measured by Marklund's and Marklund's method, ascorbic acid by Carl A Burtis method and vitamin E by Emer Engle method and Cerulloplasmin by Ravin's method.

Result: In the present study we found increased lipid-peroxidation (MDA), Superoxide dismutase and Cerulloplasmin levels in both the study group as compared to control. Significantly decreased Vitamin E and Vitamin C levels found in both the study groups as compared to control.

Conclusion: The result shows increased oxidative stress in osteoarthritis and rheumatoid arthritis patient as compared to control. Present study indicates that osteoarthritis patients have higher oxidative stress which might be the result of increased extent of lipid peroxidation or due to decreased level of antioxidants.

Keywords: Lipid-peroxidation, Superoxide-dismutase, Cerulloplasmin, Oxidative Stress, Antioxidant

INTRODUCTION

Arthritis**,** the joint inflammation, refers to a group of diseases that cause pain, swelling, stiffness and loss of motion in the joints.[1] Osteoarthritis (OA) is one of the most prevalent and disabling chronic disease affecting the elderly. The current concept holds that Osteoarthritis involves the entire joint organ, including the subchondral bone, menisci, ligaments, periarticular muscle, capsule, and synovium[2] The etiology of knee OA is multifactorial. Excessive musculoskeletal loading, high body mass index, previous knee injury, female gender and muscle weakness are well- known factors.[3] The imbalance between pro-oxidants and antioxidants gives rise to cellular oxidative stress, which plays an important role in the progression of OA.[4]

Free radicals and oxidants play a dual role, since they can be either harmful or helpful to the body. The process of free radicle formation plays a major part in the development of chronic and degenerative illness such as cancer, autoimmune disorders, aging, cataract, arthritis, cardiovascular and neurodegenerative diseases (Lien et al., 2008).[5]

Lipid- peroxidation mediated by free radical is considered to be the major mechanism of cell membrane destruction and cell damage. Free radicals are formed in both physiological conditions in mammalian tissues.[6] the uncontrolled production of free radicals is considered as an important factor in the tissue damage induced by several pathophysiologies. Alteration in the oxidant- antioxidant profile known to occur in rheumatic diseases.

In the present study we evaluate the status of lipid-peroxidation, superoxide dismutase, Cerulloplasmin and non – enzymatic antioxidant (vitamin E and Vitamin C)

MATERIAL AND METHODS

The study was undertaken in the Department of Biochemistry, M.L.N. Medical College, Allahabad after taking the ethical approval from the institutional ethical board. All patients were clinically evaluated. Subjects were selected from the urban area of Allahabad after taking the written informed consent. Detailed history was taken including age, sex and presence of any risk factor.

The subjects were categorized into three groups.

Control group (age: 30-70 years): Normal healthy individuals they are free from any diseases and not any infection.

Study Group I (age: 30-70 years): In this category patient suffering osteoarthritis.

Study Group II (age: 30-70 years): In this category patient suffering rheumatoid arthritis.

Inclusion Criteria: The study population consisted of 100 osteoarthritis patients and 100 rheumatoid arthritis patients. The information on measurement like height, weight was collected from each subject. These subjects, having normal dietary habits without any supplements of vitamins to last six month. They were non alcoholic subjects.

Exclusion Criteria: In the present study the subjects were non alcoholics, non smokers and not suffering any diseases. These subjects were not taking any supplements. All patients were diagnosed as having RA and OA according to the American Rheumatism Association criteria of 1987[7]

The study was further proceeded by collecting blood samples of subjects of the above said groups and availing them for the determination of given parameters.

[1]Assistant Professor, Department of Biochemistry, [2]Department of Pharmacology [3]Professor, Department of Biochemistry, M.L.N. Medical College Allahabad, India

Corresponding author: Dr. Shweta Dwivedi, 22A, Pura Dalel, Tilak Nagar, Allahpur, Allahabad, India

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The parameters mainly observed were oxidative stress marker (Lipid peroxidation), Ceruloplasmin, antioxidant enzyme (SOD), non-enzymatic antioxidants (Vitamin C and Vitamin E). Blood samples approximately 6ml were collected from the subjects and mixed with anticoagulant to avoid the clotting of blood samples. An aliquots (6ml) of blood sample was transferred to a centrifuge tube and kept at room temperature, plasma and packed cells were separated by centrifugation at 3000 rpm for 15 minutes. For the preparation of hemolysate the whole volume of packed cells was washed three times with cold saline water. One volume of packed cells was demolished with 1.8 volumes of cold distilled water and 0.2 volumes of cold saline water. If required, the plasma and hemolysate was refrigerated at 4°C. EDTA vials were used for the estimation of non-enzymatic antioxidant (Vitamin E and Vitamin C) by Emer-Engle method[8] and Carl A Burtis[9] method. Lipid-peroxidation was determined by Utley's method[10] with the help of red cell hemolysate and Superoxide –dismutase by Marklund and Maklund[11] method and Cerulloplasmin by Ravin's Methods.[12]

STATISTICAL ANALYSIS

Mean ± SD were used to express the data. Student t test was used for Statistical comparisons. The null hypothesis was rejected by P< 0.05.

RESULT

In the present study, Serum Lipid-peroxidation (MDA), Superoxide dismutase, Cerulloplasmin and non-enzymatic antioxidant (Vitamin E and C) levels were estimated (table-1, figure-1).

The serum MDA levels in the rheumatoid arthritis and osteoarthritis patient was 4.64±0.229 and 2.19±1.21 increased than that of controls (1.68±0.994 nmol/ml, P<0.001). Serum SOD levels in the rheumatoid arthritis patients and osteoarthritis patient was (2450± 40.1 and 2377± 38.5 unit/ml) which was increased in RA and OA patients than controls (2166±145, P< 0.001).

Serum Cerulloplasmin levels in the rheumatoid arthritis patients and osteoarthritis patient was (56.75± 19.5 and 43.65± 16.05 mg/dl) which was increased in RA and OA patients than controls (40.81±11.35, P< 0.001). Serum Vitamin E levels in the rheumatoid arthritis patients and osteoarthritis patient was (0.33± 0.02 and 0.45± 0.03 mg/dl) which was increased in RA and OA patients than controls (0.907±0.25, P< 0.001).

Serum Vitamin C levels in the rheumatoid arthritis patients and osteoarthritis patient was (0.64± 0.06 and 0.53± 0.07 mg/dl) which was increased in RA and OA patients than controls (0.87±0.25, P< 0.001). The levels of MDA, Superoxide-dismutase and Cerulloplasmin levels were significantly higher in both the study groups as compared to control subjects (P<0.001). While non- enzymatic antioxidant (Vitamin E and Vitamin C) were significantly lower in RA patients than in control subjects (P<0.001).

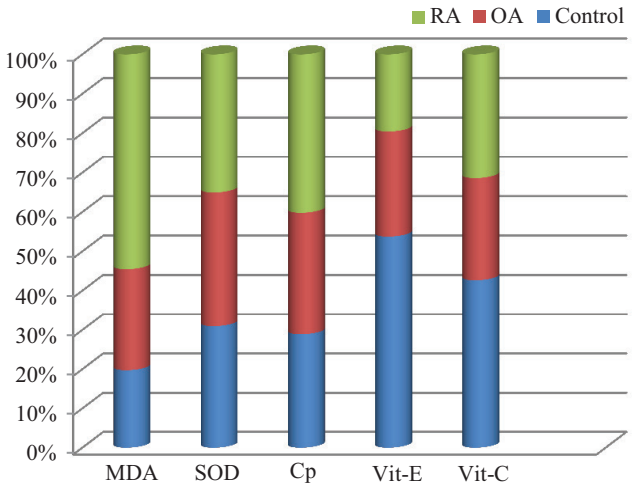

Figure-1: Levels of Lipid-peroxidation (MDA), Superoxide-dismutase (SOD), Cerulloplasmin (Cp), Vitamin E and Vitamin C in RA and OA patient compared to Control

DISCUSSION

Rheumatoid arthritis is one of the most common inflammatory diseases worldwide. In the present study elevated levels of MDA and multidirectional antioxidant were native in both arthritic patients as compared to control. Oxygen free radicals have been implicated as mediators of tissue damage in patients of rheumatoid arthritis (RA).[13] Osteoarthritis is an inflammatory disorder of the joint.[14]

Free radicals can be produced from non-enzymatic reactions of oxygen with organic compounds as well as those initiated by ionizing radiations. The non-enzymatic process can also occur during oxidative phosphorylation in the mitochondria.In the present study serum MDA was found significantly higher in RA and OA patient as compared to control. Our findings for lipid-peroxidation in both the study groups are similar to the study of Akyol et al.[15] They reported elevated MDA levels in their study. In contrast of our study, Kajanachumpol et al[16] reported no significant changes in MDA levels in rheumatoid arthritis patients as compared to control.

The elevated MDA level in our OA patients coincide with result of Surapaneni et al,[17] Maneesh et al,[18] and Rubyk et al,[19] both are reported significantly elevated lipid peroxidation levels in their study these results are inconcordance of our study. This result shows elevated lipid-peroxidation is a important cause for arthritis.

According to Mezes et al[20] and Ciemen MY et al,[21] SOD is the important antioxidant enzyme having an antitoxic effect against

S.N.	Particulars	Control (n=50)	RA Patients(n=50)	OA Patients (n=50)
1	Lipid-peroxidation	1.68±0.994	4.64±0.229	2.19±1.21
2	Superoxide-dismutase	2166±145	2450±40.1	2377±38.5
3	Cerulloplasmin	40.81±11.35	56.75±19.5	43.65±16.05
4	Vitamin E	0.907±0.25	0.33±0.02	0. 45±0.03
5	Vitamin C	0.87±0.25	0.64±0.06	0.53±0.07

Table-1: levels of Lipid-peroxidation, Superoxide-dismutase, Cerulloplasmin and Non-enzymatic antioxidant status in both the study groups compared to control groups

superoxide anion. The over expression of SOD might be an adaptive response and it result in increased in dismutation of superoxide to hydrogen peroxide.

Ostalowska et al[22] have reported increased activities of superoxide dismutase in synovial fluid of patients with primary and secondary osteoarthritis of the knee joint.

Recklies et al[23] mentioned that SOD is the first line of defense against ROS; it catalytes thedismutation of the superoxide anion into hydrogen peroxide. Ceruloplasmin, Cu containing protein, has been found to be increased in RA patients as compared to control. Increased levels of Ceruloplasmin may be related to its scavenging action of superoxide radicals that are generated during the inflammatory process of RA.[24]

Cerulloplasmin (Cp) is an acute phase protein that is primarily synthesized in the liver and secreted into the blood. It is a prominent antioxidant that can scavenge ROS.[25] We observed a significant increase in plasma Cp in OA patients was significantly higher than in control. In agreement with our findings, many authors[26] observed increased plasma Cp level in RA. On the other hand, Ashour et al.[27] stated that the raised levels of Cp are significantly increased in RA group but not in OA group. This outstanding agreement about Cp level in RA was emphasized by Nagler et al.[28] as Cp is considered the principal plasma and synovial antioxidant in RA, being responsible for up to 70% of the protective capacity against superoxide free radicals. Nevertheless, Louro et al.[29] stated that although the increase in the concentration of Cp might offer an additional safeguard against oxidative stress.

Significantly lower levels of Vitamin E and Vitamin C were found in RA patients as compared to control group. Vitamin E helps to trap free radicals and interrupt the chain reaction that damages the cells. As there is an increased oxidative stress in RA there may be increased consumption of Vitamin C and Vitamin E. This reduction in ascorbate levels suggests its role in combating oxidative stress.[30]

Our findings inconcordance with the findings of Surapaneni KM et al.[17] They observed significantly increased levels of erythrocyte SOD and significantly decreased Vitamin E and ascorbic acid levels in osteoarthritis patient as compared to control. The result suggest higher oxygen free radical production, evidence by increased SOD, increased MDA and decreased Vitamin E and Vitamin C activity, support to the oxidative stress in osteoarthritis. The increased activities of antioxidant enzymes may be a compensatory regulation in response to increased oxidative stress.

CONCLUSION

In the present study we observed elevated levels of lipid-peroxidation, superoxide-dismutase and extracellular antioxidant Cerulloplasmin in patient with osteoarthritis and rheumatoid arthritis as compared to control. Oxidative stress may be involved in rheumatoid arthritis and osteoarthritis.

REFERENCE

1. Deborah PM. Epidemiology of rheumatoid arthritios: determinants of onset, persistence and outcome. Clin Rheum. 2002;111:172-7.
2. Nuki G. Osteoarthritis: a problem of joint failure. Z Rheumatol. 1999;58:142-7.
3. Takeda H., Nakagawa T., Nakamura K., Engebretsen L. Prevention and management of knee osteoarthritis and knee cartilage injury in sports. Br J Sports Med. 2011;45:304-309.
4. Martin J. A., Buckwalter J A. Aging, articular cartilage chondrocyte senescence and osteoarthritis. Biogerontology. 2002;3:257-264.
5. Lien Ai Pham – Huy, Hua Hc, Chong Phy- Huy, Free Radicals, Antioxidants in Disease and Health, Review Article, Int. J Biomed. 2008;4:89-96.
6. Plaa, G.L., and Witschi. H. Chemicals, drugs and lipid peroxidation. Ann. Rev. Pharmacol, Toxicol. 1976;16:125-141.
7. American College of Rheumatology and HDC Committee on Clinical Guidelines 1996.
8. Hasim, S.A. and Schuttranger, G. Am. J. Clin. Nutrl August. 1996;19;137-141.
9. Burtis, C.A. 1994 Tietz Text book of Clinical Chemistry. 2 WEI.
10. Utley, H.G. Bernhein, F and Hochstein, P. Arch. Biochem, Biophys. 1967;118-28.
11. Marklund S, Markuland G. A simple assay for superoxide dismutase using autooxidation of pyrogallol. European Journal Biochem. 1974:469-72.
12. Ravins, H.A. J. Lab.Clin.Med. 1961;58,161.
13. De Bandt M, Grossin M, Driss F, Pincemail J, Babinchevaye C, Pasquier C. Vitamin E uncouples joint destruction and clinical inflammation in a transgenic mouse model of rheumatoid arthritis. Arthritis Rheum. 2002;46:522-28.
14. Gotia, S.,Popovici, I. and Hermeziu, B. Antioxidant enzymes levels in children with juvenile rheumatoid arthritis. Southeast Asian J Prop Med Public Health. 2001; 31:335-8.
15. Akyol O, Isci N, Temel I, Ozgocmen S, Uz E. The relationships between plasma and erythrocyte antioxidant enzymes and lipid peroxidation in patients with rheumatoid arthritis. Joint Bone Spine. 2001;68:311-7.
16. Surapaneni KM, Venkataraman G. Status of lipid peroxidation, glutathione, ascorbic acid, vitamin E and antioxidant enzymes in patients with osteoarthritis. Indian J Med Sci. 2007;61:93-14.
17. Krishna Mohan Superaneni, G. Venkataraman, 2007. Status of lipid peroxidation, Glutathione Ascorbic acid, Vitamin E and Antioxidant enzymes in patients with osteoarthritis.
18. M Maneesh, H Jayalekshmi, T Suma Chatterjee, A Chakrabarti and TA Singh. Indian Journal of Clinical Biochemistry. 2005;20:129-130.
19. Rubyk BI, Fil chagin NM, Sabadyshin RA. Change in lipid peroxidation in patient with primary osteoarthosis deformans. Ter Arkh. 1988;60:110–113.
20. Mezes M, Bartosiewiez G. Investigation on vitamin E and lipid-peroxide status in rheumatic disease. Clin Rheumatol. 1983;2:259-63.
21. Cimen MY, Cimen OB, Kaemaz M, Ozturk HS, Yorgancioglu R, Durak I. Oxidant/ antioxidant status of the erythrocytes from patients with rheumatoid arthritis. Clin Rheumatol. 2000;19:275-7.
22. Ostrakhovitch, E.A. and Afanas'ev, I.B. Oxidative stress in rheumatoid arthritis leukocyte: suppression by rutin and other Antioxidants and chelators. Biochem. Pharmacol. 2001;62:743-746.
23. Recklies AD, Poole AR, Banerjee S, Bogoch E, Dibattista J, Evans CH, et.al. Pathophysiologic aspects of inflammation in diarthroidal joints. In: Buckwalter JA, Einhorn TA, Simon SR, editors. Orthopaedic basic science: biology and biomechanics of the musculoskeletal system, 2nd ed.

Rosemont, IL: AAOS; 2000.p.489-530.
24. RoshanAlam, Saba Khan and Khushtar Anwar Salman. Evaluation of antioxidants and lipid peroxidation status in rheumatoid arthritis patients. NJIRM 2013;4:1-5.
25. Gambhir JK, Lali P, Jain AK. Correlation between blood antioxidant levels and lipid peroxidation in rheumatoid arthritis. Clin Biochem 1997;30:351–5. 2003;42:1234–41.
26. Kamanl A, Naziroglu M, Aydilek N, Hacievliyagil C. Plasma lipid peroxidation and antioxidant levels in patients with RA. Cell Biochem Funct. 2004;22:53–7.
27. Ashour M, Salem S, Hassaneen H, El-Gadban H, Elwan N, Awad A, et al. Antioxidant status in children with juvenile rheumatoid arthritis living in Cairo, Egypt. Int J Food Sci Nutr. 2000;51:85–90.
28. Nagler RM, Salameh F, Reznick AZ, Livshits V, Nahir AM. Salivary gland involvement in rheumatoid arthritis and its relationship toinduced oxidative stress. Rheumatology (Oxford). 2003;42:1234-41.
29. Louro MO, Cocho JA, Mera A, Tutor JC. Immunochemical and enzymatic study of ceruloplasmin in rheumatoid arthritis. J Trace Elem Med Biol. 2000;14:174–8.
30. Mahajan M, Kaur S, Mahajan S, Kant R. Uric acid a better scavenger of free radicals than vitamin C in rheumatoid arthritis. Indian. J. Clin. Biochem. 2009;24:205-207.

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