Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 7, Issue, 11, pp.23063-23066, November, 2015

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

SERUM CATHEPSIN K, ITS CORRELATION WITH RADIOLOGICAL DESTRUCTION OF JOINTS IN ESTABLISHED RHEUMATOID ARTHRITIS

***Sudeshna Sil, Vibha, C. and Vishwanath, H. L.**

Department of Biochemistry, Bangalore Medical College & Research Institute, Bangalore

ARTICLE INFO

Article History:
Received 24th August, 2015
Received in revised form
05th September, 2015
Accepted 07th October, 2015
Published online 30th November, 2015

Key words:

Rheumatoid arthritis;
Synovitis; Cathepsin K;
Cysteine protease; Osteoclast;
Collagen type I;
Collagen type II;
Osteonectin; X-rays.

ABSTRACT

Introduction: Rheumatoid arthritis is an autoimmune disease of unknown etiology characterised by persistent inflammatory synovitis involving multiple peripheral joints in a symmetrical distribution. Cathepsin K is a cysteine protease that plays an essential role in osteoclast function and in the degradation of protein components of bone matrix. It is synthesized by osteoclast and activated synovial macrophages and it cleaves collagen type I, collagen type II and osteonectin, therefore having a role in bone remodelling and resorption. Larsen score is a method of radiological scoring of X-rays in Rheumatoid arthritis.

Aim: To measure the serum levels of cathepsin K as a marker of bone resorption in rheumatoid arthritis and its correlation with the Larsen score.

Method: Case control study involving 30 patients of rheumatoid arthritis as cases and 30 age and sex matched healthy individuals as controls. Serum cathepsin K was determined and X-rays were scored by Larsen scoring method in cases.

Results: Mean serum cathepsin K (p moles/litre) was significantly increased in cases (116.18 ± 78.18) than controls (12.86 ± 3.04) with $P < 0.001$ and a significant positive correlation of serum cathepsin K with Larsen score (r value 0.903 and $P < 0.001$) was seen in cases.

Conclusion: The upregulation of serum Cathepsin K in cases and its significant positive correlation with the Larsen score mirrors the destruction of bone structures in established Rheumatoid arthritis. Cathepsin K seems to be a valuable parameter for assessment of bone metabolism in rheumatoid arthritis patients. Its measurement will contribute to the development of targeted therapies for the prevention of further bone destruction.

***Citation*:** Sudeshna Sil, Vibha. C and Vishwanath, 2015. "Serum cathepsin k, its correlation with radiological destruction of joints in established rheumatoid arthritis", *International Journal of Current Research*, 7, (11), 23063-23066.

INTRODUCTION

Rheumatoid arthritis is the paradigm of a systemic autoimmune disease characterized by inflammatory polyarthritis. The hallmark of RA is symmetric synovial inflammation causing cartilage damage and bone erosion with tenderness of multiple joints, particularly the small joints of hands and feet. Its course is extremely variable and it is frequently associated with extra-articular features (Hochberg *et al*., 2007 and Fauci *et al*., 2012). Cathepsin K is a cysteine protease that plays an essential role in osteoclast function and in the degradation of protein components of bone matrix. It is synthesized by osteoclast and activated synovial macrophages and it cleaves collagen type I, collagen type II and osteonectin, therefore having a role in bone remodelling and resorption (Batmaz, 2014; Skoumal, 2005).

****Corresponding author: Sudeshna Sil***
Department of Biochemistry, Bangalore Medical College & Research Institute, Bangalore

Cathepsin K is a tissue-specific protease associated with pycnodysostosis, a rare genetic disorder that manifests itself as bone abnormalities such as short stature, acro-osteolysis of distal phalanges and skull deformities due to mutation in the cathepsin K gene (Singh *et al*., 2004). Cathepsin K knockout mice develop an osteopetrosis. Inhibition of cathepsin K may therefore prevent bone resorption, as could be demonstrated in bone metastasis from breast cancer (Ishikawa *et al*., 2001). Diagnostic imaging provides an important tool to aid in the early diagnosis of RA and can also be used to evaluate the extent and severity of the disease. It provides an objective measure of the anatomic damage that defines the course of the disease and the long term effects of treatment. They also provide a permanent record with which the disease can be serially evaluated. An additional advantage of radiographs is that they can be randomized and blinded for standardised scoring (Hochberg *et al*., 2002). Initially it was scored by Steinbrocker method. But now the most widely used are Sharp and Larsen Method (Sokka, 2008). The upregulation of serum Cathepsin K and its correlation with Larsen score (Batmaz

et al., 2014), as a parameter for radiological changes, mirrors the destruction of bone structures in established Rheumatoid arthritis (Skoumal *et al*., 2005). Cathepsin K inhibitors can be used for treatment of Rheumatoid arthritis (Bromme *et al*., 2009).

MATERIALS AND METHODS

Source of data

The study comprised of outpatients and inpatients of rheumatoid arthritis from the department of Medicine and Orthopaedic, Victoria Hospital and Bowring and Lady Curzon Hospital, Bangalore. (Approved by Ethical Committee)

Methods of collection of data

Study design: Comparative Case control study
Study period: November 2012 – October 2014
Sample size: 30 cases of rheumatoid arthritis and 30 healthy age and sex matched individuals as controls.

Inclusion criteria

1. Patients of established Rheumatoid arthritis ≥ 18 years diagnosed as per 2010 American College of Rheumatology and the European League Against Rheumatism Classification criteria on treatment with DMARDs
2. Patients not on corticosteroids for atleast 2 months.

Exclusion criteria

a. Patients with osteoarthritis.
b. Patients with neoplastic diseases and osteolytic bone metastasis.
c. Patients with cardiovascular diseases.
d. Patients of rheumatoid arthritis on biological response modifiers.
e. Obese patients (Body Mass Index ≥ 30kg/m^2)
f. Pregnant and lactating mothers.

Parameters

1. Serum Cathepsin K by Enzyme-Linked Immunosorbent Assay

2. X-ray of both hands including wrist joint and feet in cases scored by Larsen scoring method.

X-ray of both hands including wrist joints and feet of all 30 patients were taken and Larsen scoring was done by trained radiologist. The Larsen Grading Scale is applied to standard radiographs of the hands, wrists, and feet. There are 16 sites in both hands, 8 sites in both wrists, and 8 sites in both feet. The joints considered are PIP 2 to 5 and MCP 2 to 5 in each hand, four quadrants in the wrist, and MTP 2 to 5 in each foot (Larsen, 1995 and Larsen *et al*., 1977). Total 32 sites are graded and the score ranged from o to 160 (Fig. 1)

Statistical analysis

Descriptive and inferential statistical analysis has been carried out in the present study. Results on continuous measurements are presented on Mean ± Standard Deviation (SD) (Min-Max) and results on categorical measurements are presented in Number (%). Significance is assessed at 5 % level of significance.

- Analysis of variance (ANOVA) has been used to find the significance of study parameters between three or more groups of patients.
- Pearson Correlation test is used to compare between two variables.
- Student's t test (two tailed, independent) has been used to find the significance of study parameters on continuous scale between two groups (Inter group analysis) on metric parameters.
- Chi-square/ Fisher Exact test has been used to find the significance of study parameters on categorical scale between two or more groups.

Statistical software: The Statistical software namely SAS 9.2, SPSS 15.0, Stata 10.1, MedCalc 9.0.1, Systat 12.0 and R environment ver.2.11.1 were used for the analysis of the data and Microsoft word and Excel have been used to generate graphs, tables etc.

RESULTS

Age Distribution

Table 1. Age distribution of cases and controls

Age in years	Cases		Controls	
	No	%	No	%
31-40	7	23.3	8	26.7
41-50	11	36.7	11	36.7
51-60	9	30.0	8	26.7
61-70	3	10.0	3	10.0
Total	30	100.0	30	100.0
Mean ± SD	48.70 ± 8.33		48.30 ± 8.68	

P value:
** Strongly significant P ≤ 0.01

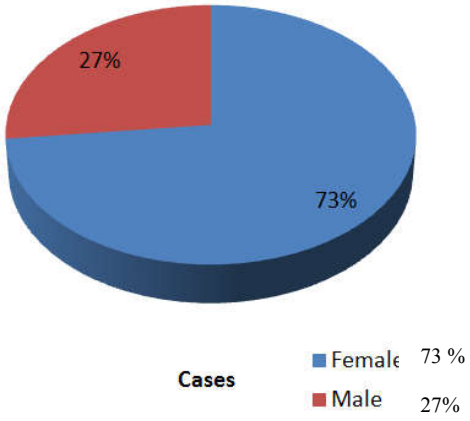
Table 2. Comparison of age in two groups studied

	Cases	Controls	P value
Age in years	48.70 ± 8.33	48.30 ± 8.68	0.856

Age of the cases and controls are thus matched with **P = 0.856**

Gender Distribution

Gender distribution is cases



Gender distribution in controls

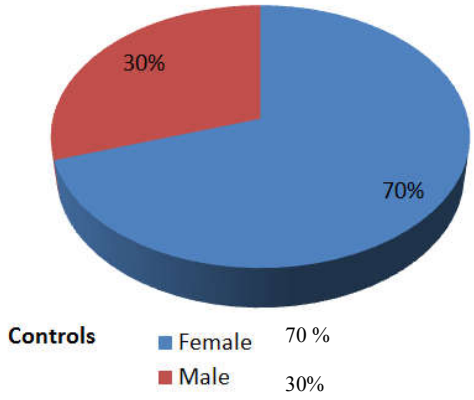


Table 3. Serum CATHEPSIN K (pmoles/litre)

	Cases	Controls	P value
CATHEPSIN K (pmoles/litre)	116.18 ± 78.18	12.86 ± 3.04	P <0.001**

Table 4. Distribution of LARSEN SCORE in Cases

Larsen score	No. of patients	%
<30	8	26.7
30-70	12	40.0
>70	10	33.3
Total	30	100.0

Table 5. Pearson correlation of CATHEPSIN K with LARSEN SCORE in cases

Pair	Cases	
	r value	P value
CATHEPSIN K vs LARSEN SCORE	0.903	<0.001**

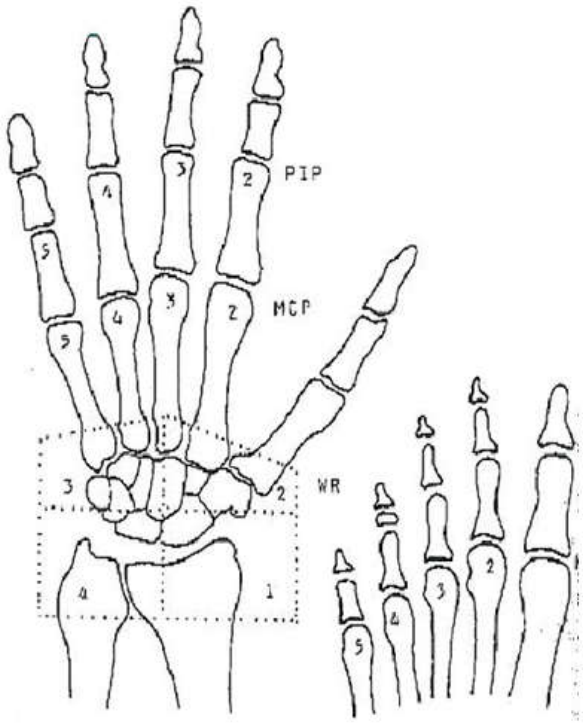


Fig. 1. Sites scored by the Larsen Grading Scale

There is a significant positive correlation of serum Cathepsin K in cases with the Larsen Score of cases with a r value of 0.903 and P < 0.001 which is strongly significant.

DISCUSSION

Serum Cathepsin K (pmoles/litre) of the cases were significantly increased with Mean ± SD (116.18 ± 78.18) as compared to the controls with Mean ± SD of 12.86 ± 3.04. Skoumal *et al*. (2005) showed a significant increase of serum cathepsin K level in the patients of RA (median first-third quartile range 54.8 pmol/l) as compared to controls (median first-third quartile range 12.7 pmoles/l) with P = 0.0003 Batmaz *et al*. (2014) showed that the levels of Cathepsin K were elevated in the serum of the patients with postmenopausal RA when compared with that of the healthy postmenopausal women as controls (P<0.05).

The pathogenesis of bone erosion and the role of osteoclasts in RA is regulated by the cytokine system. RANKL which is essential for osteoclast differentiation stimulates cathepsin K mRNA in human osteoclasts. Inflammatory cytokines such as TNF-α and IL-1β also enhances the production of the cysteine proteinase like cathepsin K at the pannus cartilage junction (Huet *et al*., 1993), making it a valuable marker of bone resorption in RA as shown by (Hou *et al*., 2001). Cathepsin K is not only expressed by osteoclasts but also by synovial fibroblasts, and suggest that cathepsin K contributes to bone destruction mediated by RA synovial cells.

The expression of cathepsin K around lymphocytic infiltrates suggests further to facilitate the movement of mononuclear cells through the perivascular interstitial matrix and thereby contribute to interstitial matrix turnover. Pearson correlation was done with Cathepsin K of cases vs Larsen score in cases which showed a strongly significant correlation with r value of 0.903 and P < 0.001. Skoumal *et al.* also showed a statistically significant correlation between serum cathepsin K and Larsen score with a P value of 0.004 in RA patients. In the study done by Batmaz *et al*. cathepsin K was positively correlated with Larsen scores (hands, feet and total) (P < 0.05) (Batmaz, 2014) in RA patients. So, this study indicated that increased Cathepsin K is associated with increased bone destruction in RA patients.

Conclusion

In conclusion, levels of serum cathepsin K was compared between 30 cases of established rheumatoid arthritis and 30 healthy individuals as controls. Serum cathepsin K was significantly increased in cases than controls. Larsen score is a method of scoring X-rays for assessing joint destruction in RA. Serum cathepsin K had a significant positive correlation with the Larsen score. So, serum cathepsin K can be a valuable parameter for assessing bone destruction as well as inhibition of cathepsin K can be a new target for preventing further bone erosion and joint destruction in established rheumatoid arthritis.

REFERENCES

Batmaz, I., Cakirca, G., Sariyildiz, M.A., Dilek, B., Mete, N., Hamidi, C., *et al*. 2014. Serum osteocalcin, bone alkaline

phosphatase and cathepsin K levels of patients with post menopausal RA. Acta Medica Mediterranea. 30: 397.

Bromme, D., Lecaille, F. Cathepsin, K. 2009. inhibitors for osteoporosis and potential off target effects. Expert Opin Investig Drugs. May; 18(5): 585–600.

Fauci, A.S., Loscalzo, J., Kasper, D.L., Hauser, S.L., Longo, D.L., Jameson, J.L., editors. 2012. Harrison's Principles of Internal Medicine. 18th edition. Vol 2. New York: McGraw-Hill; p.2738

Hochberg, M.C., Silman, A.J., Smolen, J.S., Weinblatt, M.E., Weisman, M.H. editors. 2007. Rheumatology. 4th edition. Vol 1. *Philadelphia: Mosby Elsevier*; p. 856-58.

Hou, W.S., Li, Z., Gordon, R.E., Chan, K., Klein, M.J., Levy, R, *et al*. 2001. Cathepsin, K is a Critical Protease in Synovial Fibroblast-Mediated Collagen Degradation. *American Journal of Pathology*. December; 159(6):2167-2177.

Huet, G., Filpo, R.M., Colin, C. *et al*. 1993. Stimulation of the secretion of latent cysteine proteinase activity by tumor necrosis factor alpha and interleukin-1. Arthritis Rheum. 1993; 36: 772-80.

Ishikawa, T., Kamiyama, M., Tani-Ishii, N., Suzuki, H., Ichikawa, Y., Hamaguchi, Y., *et al*. 2001. Inhibition of osteoclast differentiation and bone resorption by cathepsin K antisense oligonucleotides. Mol Carcinog ; 32:84-91

Larsen, A. 1995. How to apply Larsen score in evaluating radiographs of rheumatoid arthritis in long-term studies. *J Rheumatol* ; 22:1974–5.

Larsen, A., Dale, K., Eek, M. 1977. Radiographic evaluation of rheumatoid arthritis and related conditions by standard reference films. *Acta Radiologica Diagnosis*. 18:481-491

Singh, A.R., Kaur, A., Anand, N.K., Singh, J.R., 2004. Pyknodysostosis visceral manifestations and simian crease. *Indian J Pediatr*. 71:453-455.

Skoumal, M., Haberhauer, G., Kolarz, G., Hawa, G., Woloszczuk, W., Klingler, A. 2005. Serum cathepsin K levels of patients with longstanding rheumatoid arthritis: correlation with radiological destruction. Arthritis Res Ther. Nov; 7: R65-R70.

Sokka, T. 2008. Bulletin of the NYU Hospital for Joint Diseases. 66(2):166-68.

****