Available online at www.pharmacie-globale.info ISSN 0976-8157

Research Article

PHARMACIE GLOBALE

INTERNATIONAL JOURNAL OF COMPREHENSIVE PHARMACY

EFFECTS OF ADALIMUMAB ON BONES DESTRUCTION/REPAIRING MARKER (CTX-I & PREPTIN) IN IRAQI PATIENTS WITH RHEUMATOID ARTHRITIS

Asaad H Alzaidy*[1], Intesar T Numan[2] and Nizar A Jassim[3]

[1]College of Pharmacy, University of Al-Qadisiyah, Aldiwanyah, Iraq.
[2]Department of Pharmacology and Toxicology, College of Pharmacy, University of Baghdad, Iraq.
[3]Department of Medicine, College of Medicine, University of Baghdad, Iraq.

Received: 30 Jan 2015; Revised: 28 Feb 2016; Accepted: 8 March 2016; Available online: 27 March 2016

ABSTRACT

Rheumatoid arthritis (RA) is a chronic inflammatory disease; with variable degrees of bones, joints damage diagnosed by biomarkers and imaging changes. RA treated with methotrexate (MTX) alone or combination with biological agents: adalimumab (ADA); produce changes in progression and severity of RA, affecting level of bone formation and regeneration of biomarkers; such as CTX-I and Preptin. Objective of the current study was to assess treatment effect of MTX and MTX combined with adalimumab on level of bone resorption marker CTX-I and bone regeneration biomarker preptin. The study was open, prospective involving three study groups of 100 Iraqi patients with RA in addition to 50 apparently healthy subjects. Patients were allocated to take MTX and ADA plus MTX. CTX-I level in MTX and MTX + ADA groups; initially were significantly higher ($p<0.05$) than value of normal subjects. These values were significantly reduced after 3 months of treatment. The greater significant reduction in CTX-I was occurred in Adalimumab plus MTX group as compared with control group. Level of preptin in MTX and ADA+MTX groups; initially were significantly lower than the values of the normal subjects. After 3 months of treatment; significant elevation in preptin occurred in ADA+MTX group as compared with control and MTX alone group. Use of adalimumab in combination with MTX may enhance bone regeneration in RA patients resulted in an increase in bone regeneration biomarker preptin. But; this combination may have no additive effect to MTX on bone resorption at least on short term use (3months).

Keywords: Rheumatoid arthritis; adalimumab; MTX; preptin; CTX-I.

INTRODUCTION

Rheumatoid arthritis (RA) is a chronic inflammatory disease of poly articular arthritis in association with serological evidence of auto-reactivity affecting approximately 1% of adults worldwide. It typically leads to deformity and destruction of the joints and systemic disorders throughout the body as well.[1,2]

The advent of the biological agents has had a major impact on the treatment of RA, with TNF-α blockers achieving widespread use frequently in combination with MTX. The TNF-α blockers include monoclonal antibody adalimumab (ADA). Well-performed clinical trials indicate that TNF-α blockers can improve the signs and symptoms of RA and can retard radiographic progression. Biomarkers play pivotal roles in disease diagnosis and interventions at early stage and are also helpful in knowing the state of treatment and how body is acting or responding to the medication.[3] Therefore, exploring and measuring biologic markers in blood or in joint fluids may serve as indicators of diagnosis and also indicators of prognosis and the subsequent response to therapy.

Preptin, a peptide that corresponds to Asp69–Leu102 of pro- insulin-like growth factor-2 (pro-IGF-2), was identified as an additional molecule stored in secretory vesicles of pancreatic beta cells, and co-secreted with insulin and amylin. Similar to the other beta-pancreatic hormones, preptin has anabolic activity in bone.[4]

In bone physiology, the C-terminal telopeptide (or more formally, carboxy-terminal collagen crosslinks, and known by the acronym CTX) is a telopeptide that can be used as a biomarker in the serum to measure the rate of bone turnover. It can be useful in assisting clinicians to determine a patient's nonsurgical treatment response as well as evaluate a patient's risk of developing complications during healing following surgical intervention.[5]

Since the introduction of TNF-α blocking agents, their efficacy and safety have been studied in many randomized controlled clinical trials as well as observational studies of RA patients.[6,7] However, data regarding predictors of good clinical response of anti–TNF therapy are still sparse. The aim of this study is to assess the predictors of response to adalimumab in treatment of Iraqi patients with active RA.

SUBJECTS AND MATERIALS

Study design

This is an open label three group prospective study that was conducted over 11-month period.

***Corresponding Author:**
Asaad H Alzaidy
College of Pharmacy, University of Al-Qadisiyah,
Aldiwanyah, Iraq.
Email: alzaidy_asaad@yahoo.com

Patients

The study was conducted on Iraqi patients with rheumatoid arthritis (RA) who visited the Rheumatology Clinic in Baghdad Teaching Hospital from December 2014 to November 2015. To be included in this study, the patient should meet the 1987 American College of Rheumatology criteria for the classification of RA[8], also he should have history of failed adequate response to conventional DMARDs and his disease activity score 28 (DAS28)[9] should be equal to or greater than 5.1 (severe disease activity).

The exclusion criteria include patients less than 18 years old, patients with a previous history of biologic agent intake and those with other connective tissue diseases overlapping with RA.

125 patients with rheumatoid arthritis participated in this study; only 100 patients completed the follow up. Patients were allocated to take either methotrexate (n=63) and adalimumab plus MTX (n=62); only 50 patients completed the follow up for each groups. In addition to 58 apparently healthy subjects participated in this study as a control group, only 50 subjects completed the follow up.

All patients included in the study signed the informed consent form. Ethical Approval was obtained from the scientific Ethics Committee of Baghdad University, College of Pharmacy, and from Baghdad Teaching Hospital (Medical City, Iraq), Rheumatology Medical Department.

Sample collection

5 ml of venous blood was obtained from each patient by vein puncture before and 12 weeks after initiation of adalimumab therapy. The blood was transferred to plane tube, and then left at room temperature for at least 30 minute to coagulate, and then centrifuged for 10 min at 4000 rpm to obtain serum. The resultant serum was kept frozen until the time of analysis. After that the serum was used by using ELIZA technique for preptin, CTX-I.

Statistical analysis

The results have been expressed as Mean ± Standard Deviation (SD). Paired T test was used to test the significance of difference in means of pre and post treatment, One-way analysis of variance (ANOVA) was used to examine the degree of difference among studied groups, Chi Square test was applied for the analysis of some parameters in the study. The results of analysis with (P) values less than 0.05 were considered significant.

RESULTS AND DISCUSSION

Effect on bone destruction biomarker (CTX-I)

Table **1**, figures (**1** and **2**) shows CTX-I serum level in MTX and MTX plus adalimumab groups; initially were significantly higher ($p<0.05$) than the value of the normal subjects. These values were significantly reduced after 3 months of treatment. The greater significant reduction in CTX-I was occurred in Adalimumab Plus MTX group as compared with control group.

Table 1. Effect treatment on bones destruction/repairing marker CTX-I & preptin for study groups

Variables	Control N=50	MTX Alone N=50	MTX plus Adalimumab N=50	P value
Serum CTX				
Initial values	23.14 ±12.2	43.04 ±24.0 [a]	41.1 ±31.7 [a]	<0.001
After 3 months	26.1±11.9	22.1 ±14.7 *	18.3±13.3[a]*	0.016
% change	12.7	-48.7	-55.6	0.036
Serum preptin				
Initial values	132.3 ±44.5	52.65 ±5.1 [a]	47.1±34.0 [a]	<0.001
After 3 months	127.8 ±34.54	68.2 ±40.3 [a]*	86.4 ±39.5[ab]*	<0.001
% change	-3.4	29.7	83.6	<0.001

**Significant (p value<0.05) as compared with initial values; a Significant (p value<0.05) as compared with control values; b Significant (p value<0.05) as compared with MTX alone values*

Figure 1. Effect of treatments on CTX-I in RA patients, treated with MTX alone for 12 weeks compared with ADA +MTX and control.

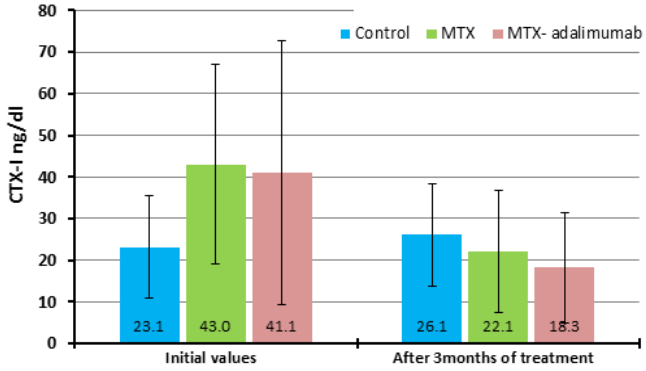
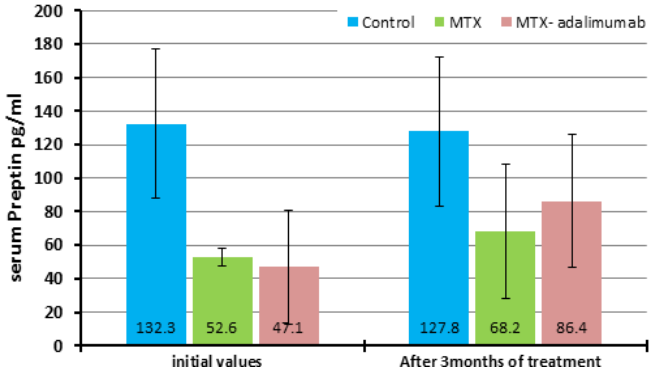


Figure 2. Effect of treatments on Preptin in RA patients, treated with MTX alone for 12 weeks compared with ADA +MTX and control.



Effect on bone repairing biomarker (Preptin)

Serum level of preptin in MTX and ADA+MTX groups; initially were significantly lower than the values of the normal subjects. These values were significantly elevated ($p<0.05$) after 3 months of treatment. The greater significant elevation in preptin occurred in ADA+MTX group as compared with control and MTX alone group.

Biochemical markers of bone turnover have been used as standard practice to measure the effects of therapy in slowly progressing bone and joints condition such as rheumatoid arthritis.[10] Because biochemical markers are sensitive and dynamic indicators of tissue turnover, they have the potential to provide information on treatment efficacy more rapidly than a variety of imaging methods.[11] Randomized clinical trials have clearly demonstrated that biological agents are able to prevent partial or even total articular erosions in RA patients. So these agents like adalimumab may positively change in the level of Bone formation and or destruction biomarkers.[12]

Bone formation rather than resorption markers better showed the bone response to anti-TNF- α.[13] This agree with our finding where greater changes occurred in level of preptin; but not CTX. This increment of preptin dose dependently stimulate the proliferation and activity of

osteoblasts and hence bone mineral density (BMD); and reduce bone resorption.[14] This effect was noticeable after treatment with ADA+MTX. Although addition of anti TNF α agent; such as dalimumab; may produce rapid decline CTX-I level.[15]

But the magnitude of change in CTX-I was not greatly different from that induced by MTX alone. Since MTX also improved bone resorption and has a bone protective effect in patient with RA.[16] That's mean, probably; use of adalimumab may did not provide significant more protective effect against bone resorption; during 12 weeks. or this effect may require more time to be obvious; where most of clinical trials showed modest changes in bonne resorption after more than 3months (probably one year or more).[17,18]

CONCLUSION

Use of adalimumab in combination with MTX may rapidly enhance bone regeneration in RA patients revealed as an increase in bone regeneration biomarker a preptin. But; this combination may have no additive effect to MTX on bone resorption at least on short term use (3months).

ACKNOWLEDGEMENTS

The present work was abstracted from PhD. theses submitted to the Department of Pharmacology and Toxicology, College of Pharmacy, Baghdad University. The authors gratefully thank Consultant Rheumatology Medical Department in Baghdad Teaching Hospital/ Medical city/ Baghdad for supporting the project. Authors confirm that there is no potential conflict of interest.

REFERENCES

1. Feldmann M, Brennan F M and R N Maini; Rheumatoid arthritis. Cell. 1996; 85(3):307-310.
2. Scott D L, Wolfe F and Huizinga T W J; Rheumatoid arthritis. The Lancet. 2010; 376(9746):1094–1108.
3. Xiaoyin Niu, Guangjie Chen; Clinical Biomarkers and Pathogenic-Related Cytokines in Rheumatoid Arthritis. *J Immunol Res*. 2014; 698192
4. Buchanan C M, Phillips A R, Cooper G J; Preptin derived from proinsulin- like growth factor II (proIGF-II) is secreted from pancreatic islet beta-cells and enhances insulin secretion. *Biochem J*. 2001; 360:431-439.
5. Marx R E et al. Oral Bisphosphonate-Induced Osteonecrosis: Risk Factors, Prediction of Risk Using Serum CTX Testing, Prevention, and Treatment. *J Oral Maxillofac Surg*. 2007; 65:2397-2410.
6. Maini R, St Clair EW, Breedveld F, Furst D, Kalden J, Weisman M, Smolen J, Emery P, Harriman G, Feldmann M, et al.: Infliximab (chimeric anti-tumor necrosis factor alpha monoclonal antibody) versus placebo in rheumatoid arthritis patients receiving concomitant methotrexate: a randomized phase III trial. ATTRACT Study Group. Lancet. 1999; 354:1932-1939.
7. Zink A, Listing J D, Kary S, Ramlau P, Stoyanova-Scholz M, babinsky K, von Hineuber U, Gromnica-Ihle E, Wassenberg S, Antoni C et al. Treatment continuation in patients receiving biologics or conventional DMARD therapy. *Ann Rheum Dis*. 2005; 64:1274-1279.
8. Arnett F C, Edworthy S M, Bloch D A et al. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. *Arthritis Rheum*. 1988; 31(3):315-24.
9. Van Der Heijde D M, Van't Hof M, Van Riel P L and Van De Putte L B; Development of a disease activity score based on judgment in clinical practice by rheumatologists. *J Rheumatol*. 1993; 20:579-81.
10. Schaller S, Henriksen K, Hoegh-Andersen P, Sondergaard B C, Sumer E U, Tanko L B, Qvist P, Karsdal MA; *In-vitro*, *ex-vivo* and *in-vivo* methodological approaches for studying therapeutic targets of osteoporosis and degenerative joint diseases: how biomarkers can assist? Assay Drug Dev Technol. 2005; 3:553–580.
11. Karsdal M A, Henriksen K, Leeming D J, Woodworth T, Vassiliadis E, Bay-Jensen A C; Novel combinations of Post-Translational Modification (PTM) neoepitopes provide tissue-specific biochemical markers - are they the cause or the consequence of the disease? *Clin Biochem*. 2010; 43:793–-04.
12. Patrice Fardellone, Alice Séjourné, Julien Paccou, Vincent Goëb. Mediators of Inflammation. 2014; Article ID 484280, 5 pages http://dx.doi.org/10.1155/2014/484280 Review Article Bone Remodelling Markers in Rheumatoid Arthritis.
13. Szentpetery M J McKenna, Murray B F et al. Periarticular bone gain at proximal interphalangeal joints and changes in bone turnover markers in response to tumor necrosis factor inhibitors in rheumatoid and psoriatic arthritis. The Journal of Rheumatology. 2013; 40(5):653-662.
14. Cornish J, Callon K E, Bava U, Watson M, Xu X, Lin J M, Chan V A, Grey A B, Naot D, Buchanan C M, Cooper G J, Reid I R; Preptin, another peptide product of the pancreatic beta-cell, is osteogenic *in-vitro* and *in-vivo*. *Am J Physiol Endocrinol Metab*. 2007; 292(1):E117-22. Epub 2006 Aug 15.
15. Chopin F, Garnero P, Henanff A le, Debiais F, Daragon A, Roux C; Extended report Long-term effects of infliximab on bone and cartilage turnover markers in patients with rheumatoid arthritis. *Ann Rheum Dis*. 2008; 67:353-357. doi:10.1136/ard.2007.076604
16. El Miedany Y M, Abubakr I H, El Baddini M; Effect of low dose methotrexate on markers of bone metabolism in patients with rheumatoid arthritis. *J Rheumatol*. 1998; 25(11):2083-7.
17. Cheryl Barnabe, David A Hanley; Seminars in rheumatology and rheumatoidlogy. Effect of Tumor Necrosis Factor Alpha Inhibition on Bone Density and Turnover Markers in Patients with Rheumatoid Arthritis and Spondyloarthropathy. 2009; 39(2):116-122
18. Karsdal Morten, Woodworth Thasia G, Kenwright, Andy, Byrjalsesn Inger, Platt Adam, Schett Georg; Biochemical Markers of Bone Balance (CTX-I/Osteocalcin) Improve with Tocilizumab Treatment in A Population of RA Patients with an Inadequate Response to Anti-TNF Therapy. Arthritis & Rheumatism. 2010; 62. Abstract Supplement.