JOJ
Urology & Nephrology
ISSN: 2476-0552

Juniper Publishers
key to the Researchers

Research Article
Special Issue – January 2018
DOI: 10.19080/JOJUN.2018.04.555641

JOJ uro & nephron

Evaluation of Nephrotoxicity using Lysoenzymuria in Patients with Rheumathoid Arthritis Treated with Most used Slow acting Antirheumatic Drugs-Saards and Establishment to the Diagnostic Value as Sifted Test

Dejan Spasovski*

University Clinic of Rheumatology, Republic of Macedonia, Skopje, Europe

Submission: September 25, 2017; **Published:** January 11, 2018

***Corresponding author:** Dejan S, University Clinic of Rheumatology, Republic of Macedonia, Europe,Tel: +389023147-668, Email: drspasovski@yahoo.co.uk

Abstract

Aim: Aim of this research is to compare diagnostic values to the laboratory variable, to (appear) present quantitative works (evaluations) of diagnostic or sifted test with define to the sensitivity, the specification and the predictable value for positive and negative test and preciseness of the test , N-acetyl- β -D-glucosaminidase, (NAG), micro albumin, Rheumatoid factor (RF), C- reactive protein (CRP), DAS 28 index, at early found diagnosis, at not treated rheumatoid arthritis (RA). To determine whether changes in tubular enzymes excreted in urine depend on the disease evolution. Micro albuminuria is used as a marker for glomerular damage, and urine excretion of N-Acetyl- β -D-glucosaminidase (NAG) as indicator of proximal tubular damage.

Methods: Using colorimetric method for determination of NAG (ROCHE), as well as immune turbid metric method for detection of micro albuminuria, samples of 70 participants are examined (35 RA patients treated with Ketoprofen only, 35 RA patients treated with combined use of Methotrexate and Ketoprofen).

Results: It is determined that there is weak correlation between NAG and micro albuminuria (r=0.34) in the group of patients treated with Ketoprofen only, while poor correlation (r=0.21) in group of patients with combined use of Methotrexate and Ketoprofen.

Conclusion: NAG has higher sensitivity from micro albuminuria in detection of a simptomatic renal lesion at untreated RA. Methotrexate is more potent NAG inductor of Ketoprofen and provokes greater tubular enzymuria than Ketoprofen.

Keywords: N-acetyl- β -D-glucosaminidase; Nephrotoxicity; Lysoenzymuria; Rhumatoid artharitis

Abbrevations: **RA:** Rheumatoid Arthritis; RF: Rheumatoid Factor; NAG: N-Acetyl- β -D-Glucosaminidase; CRP: C- Reactive Protein; Anti-CarP: Antibodies against carbamylated protein antibodies; AAR: American Association for Rheumatism

Introduction

Antibodies against carbamylated protein antibodies (anti-CarP) were recently described in patients with early and established Rheumatoid Arthritis (RA). Anti-CarP target proteins are modified through a post-translational modification named carbamylation, mediated by cyanate which mainly modifies lysine residues to homocitrullin. Carbamylation of proteins has been implicated in the pathogenesis of rheumatoid arthritis in a manner similar to citrullination. The relationship of the anti-Car P antibodies to genetic risk factors, cigarette smoking and other antibodies was evaluated in two large cohorts of RA patients (Dutch EAC and Swedish EIRA). There were no significant associations among anti-Car P antibodies and HLA-DRB1 alleles, PTPN22 or smoking. There was an association between HLA-DRB1*03 and anti-Car P-FCS. This imply that the different subsets of RA such as Anti-CCP negative/anti-Car P positive patients might have different genetic and environmental risk factors which needs further investigations [1,2].

Being positive in patients with RF and Anti-CCP negative patients with arthralgia, anti-Car P antibodies are predicting the development of RA independently of anti-CCP positivity. Anti-Car P are present in both anti-cyclic citrullinated (anti-CCP) positive and negative RA. They are associated with more severe disease course in anti-CCP negative patients and have been found to associate with joint destruction, measured as radiological

progression in anti-CCP negative RA. Together with anti-CCP they are positively correlated with DAS28. The diagnostic value for RA in comparison with anti-CCP antibodies is still questionable and needs more investigations, but the concomitant presence of two to three antibodies highly increases the odds ratio of having RA in comparison with the presence of one or no antibodies [3-10].

Patient and Methods

Among the patient examined for this study, the diagnosis of the disease will be establish on the base of revised diagnostic criterion for classification of rheumatoid arthritis, suggested in 1987 American Association for Rheumatism (AAR) [11-15]. For classified purpose, the patient to be in the group of rheumatoid arthritis must fulfill at four from seven criterions. Criterions from one to four are present at least six weeks. In the study are involve 35 patients (female 28, male 7), who are ill from RA, and 35 patient (female 18, male 17) as control healthy group. Their average age is 56, 68 years (±6.79) (40-65 years) in the group RA, but 46,2 years (±12.49) (29-65 years) in control healthy group. The average time of the disease in month from the beginning is 43, 97 (±45.23), in interval from (1-168) months. One of the patients who are in research doesn't have medical record for past or present renal disease. Three patients previous are treated with oral corticosteroids medicine, while none of them has not used NSAIL. The rest of the patients refuse use of other medicines before taking the examinations (Table 1).

Table 1: Clinical characteristics of the patients who are in this research.

	RA Nº 35 Value (M ±SD)	Control Healthy Group Nº 35 Value (M± SD)
Male/female relation	28-Jul	17/18
Middle average age (years)	56.68 (± 6.79)(40-65)	46.20 (± 12.49) (29-65)
Middle duration (time) of disease (month)	43.97 (± 45.23)(1.0-168)	0.00 (± 0.00)(0.00-0.00)
Previous treatment with gold salts (Nº of patients)	0	0
Previous therapy with oral corticosteroids (Nº of patients)	3	0
Previous therapy with Metotrexat (Nº of patients)	0	0
Previous therapy with NSAIL (Nº of patients)	0	0

Criteria for inclusion

The study includes patients suffering from RA, age 18-65 years, till now not treated with NSAIDs and DMARDs.

Criteria for exception from the research

From the research were accepted all the patients with a disease or condition which could directly or indirectly influence a change in results:

- Patients with previous medical record for diseases of the spleen, thyroid gland, hepatal damage, renal, hematologic, cardiovascular, neurotic and lung damage, autoimmune disease, AIDS, aged <18 years.
- Patients with diabetes mellitus, acute infections, malignant neoplasm, febrile conditions.
- Patients treated with antibiotics and salycilate in the period of six months prior to the beginning of the study.
- Patients with hypertension arterialis, uric arthritis, uric infections, SLE, Sy Sjögren, mixed conjunction texture disease, vasculitis.
- Patients treated with anti hypertension, anti diabetic and cardiac therapy.
- Patients with anamnesis for transfusion of blood and overweight.
- Hypersensitive to some of the medicines or their components.
- Accepted patients who together with these medicines take medicines from basic line.
- Accepted patients whose results show that in 0 spot there is a glycemia, or increased level of degraded products: creatinine in serum and urine, urea in serum and disorder of the hematologic and enzymatic status. All patients took part in this study voluntarily, so the ethical criterion was not breached during our work.

Laboratory Assessment

Colorimetric assay for the determination of N-Acetyl-β-D-glucosaminidase (NAG) in urine (roche)

Principle of the assay: 3-Cresolsulfonphthaleinyl-N-acetyl-β-D-glucosaminide, sodium salt, is hydrolyzed by N-acetyl-β-D-glucosaminidase (NAG) with the release of 3-cresol-sulfonphthalein, sodium salt (3-cresol purple), which is measured photo metrically at 580nm. Turbid urines should be centrifuged and the supernatant decant. Reference value : NAG urine 0.27-1.18 U/mmol creatinine

Immuno turbidometric assay for the determination of urinary albumin

Principle of the assay: An undiluted sample is added to a buffer containing the antibody specific for human serum albumin. The absorbance (340nm) of the resulting turbid solution is proportional to the concentration of albumin in the sample urine. By constructing a standard curve from the absorbance of standards, the albumin concentration of the sample can be determined. The assay can be carried out manually (at room temperature) or with an automated analyzer using DAKO tests.

How to cite this article: Dejan S. Evaluation of Nephrotoxicity using Lysoenzymuria in Patients with Rheumathoid Arthritis Treated with Most used Slow acting Antirheumatic Drugs-Saards and Establishment to the Diagnostic Value as Sifted Test. JOJ uro & nephron. 2018; 4(4): 555641. DOI: 10.19080/JOJUN.2018.04.555641.

Sample collection and storage: For random urinary albumin measurement, use an early morning mid-stream specimen. Centrifuge cloudy samples before use and analyze clear supernatant in the assay. Reference value: 2.0-20.0mg/L .

Statistical Analysis

For testing about importance of difference between two arithmetic mean, respectively proportion is used Student's t-test, comparing the middle values of the certain numerous parameters between two groups, as and Wilcoxon-matched test for independent examples. Sensitivity and predictivity for positive and negative test of examined marks is defined with the test of sensitivity and specification. P value between 0.05 and 0.1 is taken as statistic significant. Data processing is done with statistics packet Statistical 7.0

Results

Testing the significance in differences in both groups in 0 spot, in the group of patients treated only with Ketoprofen the mean value of the urinary NAG induction ranges 0.93±0.48, in the group of patients treated with combined use of Methotrexate and Ketoprofen 1.13±0.54. It shows that Methotrexate is more potent NAG inductor compared with Ketoprofen, but during their combined use mean urinary NAG induction is increased considering size and time of appearance (Figure 1). In the group of patients treated with Ketoprofen, the distribution of patients according to the values of micro albuminuria in the five probes, one could conclude that elevated values of micro albuminuria are registered in 7(20%) of patients in the 4th week (2nd probe) when the level of micro albuminuria is highest (19.55±11.46).

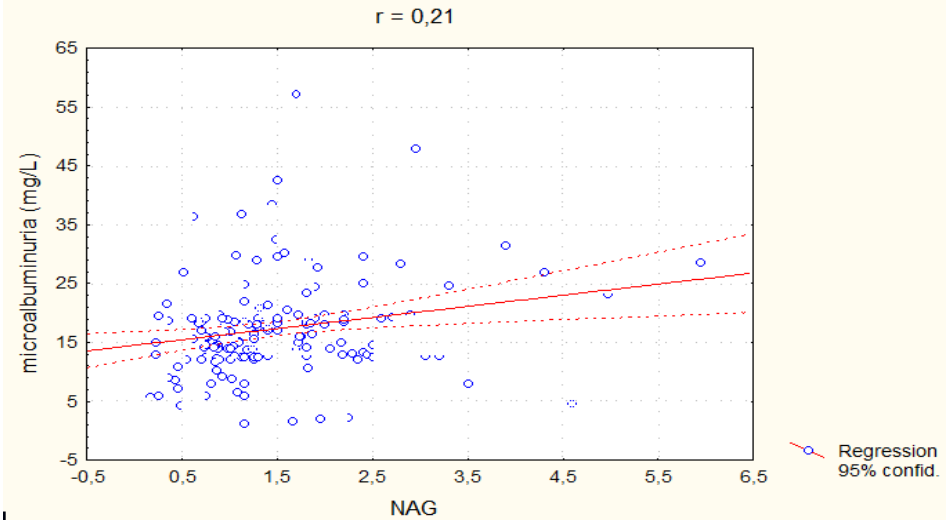

Figure 1: Pearson's coefficient of correlation (r) between the values of N-acetyl-β-D-glucosaminidase (NAG) and micro albuminuria in the group of patients with the combined use of Methotrexate and Ketoprofen.

Testing the significance of difference in both groups in 0 spot, in the group of patients treated with Ketoprofen only mean value of micro albuminuria is 17.91±11.17, while in the group treated with combined use of Methotrexate and Ketoprofen is 16.35±7.41. This explains why Methotrexate gives identical appearance of micro albuminuria compared with Ketoprofen, but with their combined use micro albuminuria is increased both in size and time of appearance.

Table 2: Diagnostic performance of NAG micro albumin and other laboratory variables at rheumatoid arthritis.

	Nag RA No35	Nag RA- No18	Nag RA+ No17	Micro Albumin RA No 35	Micro Albumin R A- No 18	Micro Albumin R A+ No 17
Sensitivity %	37.14	50	30.76	11.42	5.55	17.64
Specification%	77.14	77.14	77.14	94.28	94.28	94.28
Predictable Values for The Positive Test %	61.90	52.94	33.33	66.66	33.33	60
Predictable Values For The Negative Test %	44.89	25	32.5	48.43	34	29.78
Precision %	57.14	67.92	59.61	52.85	64.15	69.29

How to cite this article: Dejan S. Evaluation of Nephrotoxicity using Lysoenzymuria in Patients with Rheumathoid Arthritis Treated with Most used Slow acting Antirheumatic Drugs-Saards and Establishment to the Diagnostic Value as Sifted Test. JOJ uro & nephron. 2018; 4(4): 555641. DOI: 10.19080/JOJUN.2018.04.555641.

Diagnostic value of N-Acetyl- β -D-Glucosaminidase (nag) and micro albumin in urine in patients with rheumatoid arthritis (RA)

For NAG, micro albumin and for other laboratoric variables at rheumatoid arthritis, sensitivity, specification, predictable value for positive and negative test as the precision of them are show in Table 2. NAG has better diagnosed performances from micro albuminuria in relation with sensitivity (sensitivity 37,14% vs 11.42 %), but with lower specification (specification 77.14% vs 94.28%) in detection of renal tubular damage at untreated RA.

For NAG, micro albumin and for other laboratoric variables at rheumatoid arthritis, sensitivity, specification, predictable value for positive and negative test as the precision of them are show in Table 2. NAG has better diagnosed performances from micro albuminuria in relation with sensitivity (sensitivity 37,14% vs 11.42 %), but with lower specification (specification 77.14% vs 94.28%) in detection of renal tubular damage at untreated RA.

Discussion

There is no change of the clinical parameters of the renal function regarding degradation products of the nitric metabolism (creatinin in serum and urine, GFR) in the course of follow up. The least sensitive markers for early nephrotoxicity caused by Methotrexate and Ketoprofen are concentration of creatinin in serum and urine and urea in serum, as well as the level of CCC. These tests point at the changed, decreased glomerular filtration, but not at the changes in renal tubular function. We think that the use of these parameters can find application in the clinical practice in cases when there is much longer therapy with Methotrexate and Ketoprofen, combined with antibiotics, when they can indicate impairment of the glomerular filtration.

Does exist statements for working on RA under renal texture [15-18], expose NAG as the most relevant marker for assessment of asymptomtic renal disfunction. Sensitivity of NAG compare with the sensitivity of microalbumin is higher (37.14% Vs 11.42%). This sensitivity is close to the sensitivity of GFR calculated with calculated creatinin clirens after Cocroft@ Gault (40%), as mathematical score is composite from serum creatinine, age and body weight. NAG is isolated laboratory variable dominate from the rest its performance, in diagnose of asymptomatic renal tubular dysfunction. The rest standards working analyses are using for assessment of renal function, have show low sensitivity: creatinin serum, creatinin urine, urea serum (8.57% Vs 25.71% Vs 11.42%).

Seropositivity has influence of the appearance of NAG induction, which has show in our example that seropositive RF patients with DAS 28>3.2 have much higher NAG induction after circumference from seronegative RF with DAS 28 >3.2. Statistical connection of the duration of the disease in month and NAG enzymuria (p=0.000000) show that untreated RA works on the renal texture as one of visceral appearances of the disease. Untreated RA primary damages tubular, but in very small circumference (percentage) glomerular apparatus.

Conclusion

Determination of the urinary NAG together with the urinary creatinin excretion could serve as a more sensitive test for renal lesions in patients suffering from RA, as an additional diagnostic tool, and the information for the status of the disease. NAG has higher sensitivity from microalbuminuria. It's a relevant marker in assessment of asymptomatic renal damages at untreated RA. NAG can be used in everyday clinical practice.

References

1. Mellman I, Fuchs R, Helenius A (1986) Acidification of the endocytic and exocytic pathways. Annu Rev Biochem 55: 663-700.
2. Wellwood JM, Ellis BG, Price RG, Hammond K, Thompson AE, et al. (1975) Urinary N-acetyl-beta-D-glucosaminidase activities in patients with renal disease. Br Med J 16(3): 408-411.
3. Dos Santos AC, Colacciopo S, Dal Bo CM, dos Santos NA (1994) Occupational exposure to lead, kidney function tests and blood pressure. Am J Int Med 26(5): 635-643.
4. Shibasak T, Nakano H, Ohno I (1994) Effect of pentoxifylline on CdCl -induced nephrotoxici-2ty in the rat. Biol Trace Elem Res 41: 245-251.
5. Hong CY, Chia KS (1998) Markers of diabetic nephropathy. J Diabetes Complications 12(1): 43-60.
6. Sherman RL, Drayer ED, Leyland JBR (1983) N-Acetyl-beta-glucosaminidase and beta 2-microglobulin. Their urinary excretion in patients with renal parenchymal disease. Arch Intern Med 143(6): 1183-1185.
7. Hultberg B, Ravnskov U (1981) The excretion of N-acetyl-beta-glucosaminidase in glomerulonephritis. Clin Nephrol 15(1): 33-38.
8. Jung K, Mattenheimer H, Burchardt U (1992) Urinary Enzymes in Clinical and Experimental Medicine. Springer Verlag, Berlin, Germany.
9. Price RG (1992) Measurement of N-acetyl-beta-glucosaminidase and it's isoenzymes in urine, methods and clinical applications. Eur J Clin Chem Clin Biochem 30(10): 693-705.
10. Hofmann W, Guder WG (1989) A diagnostic programme for quantitative analysis of proteinuria. J Clin Chem Clin Biochem 27(9): 589-600.
11. Chiu JSP (1994) Models used to asses renal function. Drug Devel Res 32(4): 247-255.
12. Price RG (1992) The role of NAG (N-acetyl-D-beta-glucosaminidase) in the diagnosis of kidney disease including the monitoring of nephrotoxicity. Clin Nephrol 38(suppl 1): S14-S19.
13. Tucker SM, Pierce RJ, Price RG (1980) Characterization of human N-acetyl-beta-D-glucosaminidase isoenzymes as an indicator of tissue damage in disease. Clin Chim Acta 102(1): 29-40.
14. De Duve C (1964) Principles of tissue fractionation. J Theor Biol 6(1): 33-59.
15. Thuss S, Milano G, Kubar J, Namer M, Schneider M (1986) Clinical and pharmacokinetics evidence of a life-threatening interaction between methotrexate and ketoprofen. Lancet 1(8475): 256-258.
16. Brater DC (1986) Drug-drug and drug-disease interaction with non-steroidal anti-infamatory drugs. Am J Med 80(1A): 62-77.
17. Tracy TS, Krohn K, Jones DR, Bradley JD, Hall SD, et al. (1992) The effect of salicylate, ibuprofen and naproxen on the disposition of methotrexate in rheumathoid arthritis. Eur J Clin Pharmacol 42(2): 121-125.
18. Stewart CF, Fleming RA, Arkin CR, Evans WE (1990) Coadministration of naproxen and low-dose methotrexate in patient with rheumathoid arthritis. Clin Pharmacol Ther 47(4): 540-546.

How to cite this article: Dejan S. Evaluation of Nephrotoxicity using Lysoenzymuria in Patients with Rheumathoid Arthritis Treated with Most used Slow acting Antirheumatic Drugs-Saards and Establishment to the Diagnostic Value as Sifted Test. JOJ uro & nephron. 2018; 4(4): 555641. DOI: 10.19080/JOJUN.2018.04.555641.

DOI: 10.19080/JOJUN.2017.04.555641

How to cite this article: Dejan S. Evaluation of Nephrotoxicity using Lysoenzymuria in Patients with Rheumathoid Arthritis Treated with Most used Slow acting Antirheumatic Drugs-Saards and Establishment to the Diagnostic Value as Sifted Test. JOJ uro & nephron. 2018; 4(4): 555641. DOI: 10.19080/JOJUN.2018.04.555641.