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Original Article

Biochemistry Section

Oxidative Stress and Total Antioxidant Capacity in Renal Failure and Renal Calculi Patients-A Cross Sectional Study

SHIVPRASAD S, SMITA SONOLI, ANURADHA B PATIL

ABSTRACT

Background: Oxidative stress has been the centre of focus in most of the research, and in the field of medicine it takes a special place especially in diseases associated with kidneys. Stimulation of renal tissue by factors like infection, proteinuria, food allergens, drugs, high glucose levels, and increased blood pressure reactive oxygen species are released resulting in tissue injury and progression of disease.

Aims: To determine the level of oxidative stress and the antioxidant defense in patients with CRF, ARF and renal calculi.

Methods & Materials: Patients between age group of 30 to 50 years were selected as participants of the study. 50 cases each of acute renal failure, chronic renal failure and renal calculi were taken along with 50 control samples. The blood was collected after obtaining written informed consent. 5 ml of blood was collected under aseptic precautions and all the parameters were evaluated by standard biochemical methods. Oxidative stress was measured in terms of MDA, a reliable indicator of lipid peroxidation product.

Statistical Analysis: Student "t" test (unpaired) – was used to calculate the statistical significance between cases and controls and also by Analysis of variance (ANOVA).

Results & Conclusion: Malondialdehyde representing the oxidative stress in patients of Chronic Renal Failure, Acute Renal Failure and renal calculi was high indicating the stress among those patients. Total antioxidant capacity was low among ARF and CRF in comparison with controls. Reactive oxygen species (ROS) formation is part of the nonspecific defense system of an organism. ROS may also affect almost all cells of the host organism, in particular at sites where there is active inflammation.

Key Words: Oxidative stress, Chronic renal failure, Acute renal failure, Renal calculi

INTRODUCTION

Oxidative stress is defined as an imbalance between the over whelming production of free radicals or failure of antioxidant defense mechanism. Reactive Oxygen Species (ROS; Free radicals) are highly reactive species generated by mitochondria during oxidative phosphorylation; lipoxygenase and cyclooxygenase; cytochrome p450 ; NADPH oxidase enzyme present in neutrophils, macrophages and phagocytes by procress called respiratory brust [1]. ROS include superoxide anion radical, hydrogen peroxy radical,hydrogen peroxide, nitric oxide [2].They produce tissue injury by lipid peroxidation, enzyme inactivation, damage to DNA and structural proteins [3] suggesting that ROS are involved in biological phenomenon and are very toxic. Human body has developed defense mechanisms against these radicals, which include SOD, calalase , Glutathione peroxidase(enzymatic antioxidants) and Vitamin A,E, and C (non enzymatic antioxidants).

No disease is left unassociated with oxidative stress so are renal disorders, on stimulation of renal tissue by factors like infection, proteinuria, food allergens, drugs, high glucose levels, and increased blood pressure ROS are released resulting in tissue injury and progression of disease.

Renal sources of ROS are cells of renal tissue like vascular (endothelial and smooth muscle cells), glomerular (endothelial and mesangial cells), tubular(proximal, distal, and convoluted tubular cells) [4] along with infiltrating blood cells and infection controlling cells like neutrophils, macrophages and phago-

cytes. Hyper production of ROS is associated with shortened cell survival, deformed cells leading to lipid peroxidation.

Traditionally the main reason for the formation and progression of renal diseases in urban society is affluent life style, usage of higher antibiotics to combat infections, diabetes mellitus, hypertension, variety of food stuffs contaminated with insecticides. These increase the magnitude of the renal disease. Global incidence per year is 8,50,000 deaths and 115,010,107 disability and In India: 1,50,000 new cases of CRF occur for every one billion population [5]. However the reliable data for the overall incidence and prevalence of renal disorders is lacking because of non-maintenance of national register. In this part of our country, (North Karnataka, India) incidence of acute glomerular nephritis is 25, nephritic syndrome 60, acute renal failure 80, chronic renal failure 50, and renal calculi 1000 cases/year.

The magnitude of disease has compelled researchers to understand the disease in depth; In modern medicine more and more emphasis are laid on biochemical changes like oxidative stress and antioxidant defence. ROS lead to cellular damage causing biochemical and pathological changes in the renal tissue which is reflected by serum and urinary changes of urea, uric acid, creatinine, total protein, albumin, cholesterol etc. These biochemical parameters are reliable and time tested diagnostic and prognostic indicators of renal disorders.

Oxidative stress is measured in terms of MDA, a reliable indicator of lipid peroxidation product along with total antioxidant capacity in renal disorder patients like acute renal failure, chronic renal failure and renal calculi. These assays will be accomplished by estimations of serum urea, uric acid, creatinine, total protein, albumin and cholesterol which will help in diagnosis and prognosis of disease. This study may help us to optimize antioxidant supplementation as adjuvant therapy in treatment of renal disorders which will help in preventing the impeding complications caused by progression of the disease.

MATERIALS AND METHODS

Source of data

The data was collected from patients who attended Urology, Nephrology, Medicine, and Outpatient department of KLES Dr Prabhakar Kore Hospital, Belgaum, India.

Method of collection of data

Patients between age group of 30 to 50 years diagnosed by senior consultant, based on clinical examination and relevant biochemical findings served as cases. Age and sex matched healthy individuals served as controls. The blood was collected after obtaining written informed consent. 5ml of blood was collected under aseptic precautions and all the parameters were evaluated by standard biochemical methods.

Estimation of MDA-Chemical method known as TBARS method and analyses of the reaction was done by spectrophotometer [6].

Estimation of TAC-Chemical method-D Koracevic et al.,-spectrophotometer [7].

Study Design: Cros-sectional study.

Ethical Clearance: Ethical clearance was taken prior to the conduct of the study from the institutional ethical committee.

Sample Size: 50 cases each of acute renal failure, chronic renal failure and renal calculi were taken along with 50 control samples.

Inclusion Criteria: Normal healthy persons – as controls and clinically diagnosed patients of ARF, CRF and RC – as cases were enrolled.

Exclusion Criteria: Includes Patients on dialysis and on treatment, cerebrovascular disorders, Coronary artery disease, Malignancy, Hepatic disorders, Respiratory disorders.

STATISTICAL ANALYSIS

Student t test (unpaired) – was used to calculate the statistical significance between cases and controls and also by Analysis of variance (ANOVA).

RESULTS

Oxidative stress measured in terms of MDA was high among ARF, CRF and renal calculi as compared with controls (P=0.001). Total antioxidant capacity was low among ARF and CRF in comparison with controls. The renal calculi patients showed no significant difference in total antioxidant capacity as compared with controls [Table/Fig-1].

Parameter	Control	ARF (n=50)	CRF (n=50)	Renal Calculi(n=50)
MDA(nmol/ml) (Mean ± SD)	1.59± 1.14	6.93± 1.77	5.77± 1.35	3.62±1.22
TAC(µmol/l) (Mean ± SD)	1009± 138.02	620± 172.20	545± 111.04	1058±141.07

[Table/Fig-1]: Results of MDA & TCA in ARF, CRF & Renal calculi including controls

DISCUSSION AND CONCLUSION

The malondialdehyde representing the oxidative stress in patients of CRF, ARF and renal calculi was high indicating the stress among those patients. The ROS are released resulting in tissue injury and progression of disease. Renal sources of ROS are cells of renal tissue like vascular (endothelial and smooth muscle cells), glomerular (endothelial and mesangial

cells), tubular (proximal, distal, and convoluted tubular cells [4] along with infiltrating blood cells and infection controlling cells like neutrophils, macrophages and phagocytes. Hyper production of ROS is associated with shortened cell survival, deformed cells leading to lipid peroxidation and increased malondialdehyde level.

Reactive oxygen species (ROS) formation is part of the non-specific defense system of an organism. ROS may also affect almost all cells of the host organism, in particular at sites where there is active inflammation. The latter plays a role in a variety of renal diseases, such as glomerulonephritis, acute or progressive renal failure, or tubulointerstitial nephritis [8,9].

Chronic renal failure was characterized by a pro-oxidant state in plasma, demonstrated both by a depletion of antioxidants and by the presence of the products of free radical actions. Our results of the study were in accordance with some studies done previously [10, 11]. ROS are also considered as a contributing factor in the development of ischaemia-reperfusion injury [12]. Moreover, due to their impact on cell cycle regulation [13], oxygen radicals may contribute to hypertrophy of tubular cells [14].

In the vascular system, the interaction of O_2– with NO seems to be of major importance, particularly in the setting of hypercholesterolaemia, atherosclerosis, and in hypertension. The important endothelial vasodilator autacoid NO is inactivated by O_2– [15], and the reaction product peroxynitrite (ONOO–) further strongly helps in formation of even more ROS such as OH• [16]. Atherogenic lipoproteins like LDL and Lp(a) not only undergo oxidative modification in the vascular wall [17], but are also capable of enhancing generation of ROS via stimulation of NADH/ NADPH-dependent oxidases in endothelial cells, smooth muscle cells, juxtaglomerular cells, and mesangial cells [18,19].

Enhanced O_2–formation results in either cell proliferation or apoptotic death of endothelial cells, depending on the concentration and the time of exposure [19,20]. It is worthwhile to supplement the patients with antioxidants so as to combat the oxidative load in the body, and also to replenish the antioxidants in the body.

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AUTHOR(S):

1. Dr. Shivprasad S
2. Dr. Smita Sonoli
3. Dr. Anuradha B Patil

PARTICULARS OF CONTRIBUTORS:

1. Assistant Professor, Department of Biochemistry, Jawaharlal Nehru Medical College, Belgaum, Karnataka, India.
2. Associate Professor, Department of Biochemistry, Jawaharlal Nehru Medical College, Belgaum, Karnataka, India.
3. Professor & HOD, Department of Biochemistry, Jawaharlal Nehru Medical College, Belgaum, Karnataka, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:

Dr. Shivprasad S,
Assistant Professor, Department of Biochemistry,
Jawaharlal Nehru Medical College,
Belgaum, Karnataka, India.
Email: Shivprasad273@yahoo.com
Ph: 9916895566

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