ANTIOXIDANT VITAMINS AND LIPID PEROXIDATION ABNORMALITIES IN THAI PATIENTS WITH CHRONIC RENAL FAILURE

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ABSTRACT

Oxidative stress has been shown from other publications to play an importance role in pathogenesis of renal disease and the progress of the disease up to end stage. The present study was performed in 54 Thai chronic renal failure (CRF) patients with oxidative stress status and was compared to 32 normal subjects. The results revealed that CRF patients were deficient in red blood cell α-tocopherol(vitamin E). These patients had higher rates of lipid peroxidation and were highly susceptible to red cell hemolysis. The results thus confirmed the existence of impaired antioxidant system. Our study also suggested that oral supplementation of α-tocopherol in chronic renal failure patients should be further study.

Keywords: antioxidant vitamins, lipid peroxidation, chronic renal failure.

INTRODUCTION

It has been conceived that reactive oxygen species(ROS) play a major role as mediators of tissue injury in both immune-inflammatory process and ischemia[1]. More recently, the reactive oxygen species were considered to involved in the pathogenesis of renal injury and glomerulonephritis as well[2]. ROS can be generated from activated infiltrating blood cells and from renal cells themselves[3].They invade the renal tissue and induce renal cell injury. The injury is not only directed from the serious effect of ROS but also from the work load of remnant kidney[4]. ROS react with the membrane lipids and may contribute in the kidney cell injury. This results in the increase of whole-kidney MDA (Malondialdehyde) content. The MDA represents an index of peroxidation of polyunsaturated fatty acid by ROS ,which is considered to be quantitative indicators of the peroxidation reaction of ROS[5].

In Thailand, the CRF remains a major medical problem because of its increasing incidence and high costs of long term dialysis program and transplantation. However, very little information on oxidative stress and the importance of antioxidant vitamins in Thai chronic renal failure patients is available. For these reasons, the present work was investigated on the oxidative stress, lipid peroxidation products and the antioxidant capacity such as antioxidant vitamins, in Thai chronic renal disease patients with varying degrees of renal dysfunction. Thai chronic renal failure patients from both out-and in-patients of Medicine Department, Siriraj Hospital were enrolled in this study with informed consents. For the purpose of comparison, healthy volunteers from hospital staff and personnels were constituted as normal control group.

MATERIALS AND METHODS

The experiment was studied in 54 chronic renal failure patients (19 males and 35 females),age range from 20 to 72 years (average 45.77 ± 14.26) recruited with informed consents. All subjects were free from diabetes mellitus, chronic respiratory insufficiency, intercurrent infection, hepatic disorder, alcohol ingestion and cigarette smoking. None of them had either blood or plasma transfusion during the last 3 months preceding the study. Iron and vitamins supplements were stopped 2 weeks before blood and urine determinations. The subjects were divided into 5 groups according to severity of the renal failure, represented by serum creatinine levels. : Group I = serum creatinine ≤ 2 mg/dl. ; Group II = serum creatinine >2 up to 4 mg/dl ; Group III = serum creatinine >4 up to 8 mg/dl; Group IV = serum creatinine >8 up to 12 mg/dl and Group V = serum creatinine > 12 mg/dl.

There were 33 cases of glomerulonephritis, 11 cases of nephrosclerosis, 7 cases of chronic tubulointerstitial nephritis, 1 case of polycystic kidney disease and 2 cases of unclassified etiology. Thirty-two healthy volunteers (16 males and 16 females), aged from 18 to 50 years(average 30.69 ± 8.63) were as controls. Fresh and heparinized blood samples were obtained for the plasma and urine lipid peroxide were determined by malondialdehyde (MDA) formation[6,7]. The erythrocyte and plasma antioxidant vitamins were performed by using high performance liquid chromatography[8,9]. All results are expressed as the mean ± SD. Statistical

analysis was analyzed using unpaired two tailed Student's t-test. Statistical significance level was defined as $P<0.05$

RESULTS

The clinical, chemical and hematological data of the 54 CRF patients and 32 normal control are observed. The typical laboratory alterations of chronic renal failure patients were apparent. Compared to the normal group, CRF patients has elevated serum creatinine, blood urea nitrogen and urine protein whereas low hemoglobin concentration and hematocrit were observed. These differences are highly statistically significant.

The plasma cholesterol, triglyceride, low-density lipoprotein, low-density lipoprotein concentrations were significantly higher in CRF patients. CRF patients contained significantly lower concentration of high-density lipoprotein. Average concentrations of plasma lipid peroxidation product (MDA) and urine MDA in CRF patients were significantly higher than control (table 1). In addition, the elevated plasma MDA in group 1 and 2 of the CRF patients had a trend to increase comparing to the other group.

Antioxidant capacity of CRF patients showed a clear cut decrease in red blood cell vitamin E. The red blood cell vitamin E in CRF patients and normal subjects are shown in table 1. However, the mean plasma levels of vitamin E in CRF patients were not lower than that of the normal control group. The result also showed that other antioxidant vitamins such as vitamin A and β-carotene were not difference between two groups.

Table 1 Plasma ; urine lipid peroxidation product (MDA); erythrocyte vitamin E; plasma antioxidant vitamins (vitamin A, E and β-carotene) at different state of renal dysfunction in patients and in normal subjects.

Data	Control (n=32)	Patients					
		All (n=54)	group 1 (n=12)	group 2 (n=8)	group 3 (n=13)	group 4 (n=13)	group 5 (n=8)
Plasma MDA(μmol/l)	7.39 ±3.39	92.49 ±61.66	123.50 ±54.47*	146.43 ± 84.44*	83.85 ±61.38*	64.23 ±35.70*	69.38 ±45.55*
Urine MDA (nmol/Ccr.)	32.08 ±24.0	246.14 ±325.16*	63.49 ±67.17*	129.98 ±85.11*	200.66 ±239.79*	429.47 ±363.59*	548.77 ±621.92*
RBC vitamin E (μg/ml PRC)	3.38 ±0.45	2.23 ±0.52	2.17 ±0.41*	2.32 ±0.79*	2.36 ±0.36*	2.25 ±0.53*	1.97 ±0.56*
Plasma vit-E (mg/dl)	1.82 ±0.53	1.88 ± 0.59(NS)	2.22 ± 0.91(NS)	2.09 ± 0.73(NS)	1.73 ± 0.59(NS)	1.73 ± 0.55(NS)	1.68 ± 0.62(NS)
Plasma vit-A (mg/dl)	0.23 ±0.01	0.23 ± 0.08(NS)	0.22 ± 0.07(NS)	0.25 ± 0.01(NS)	0.22 ± 0.01(NS)	0.24 ± 0.01(NS)	0.21 ± 0.01(NS)
Plasma β-carotene (mg/dl)	0.04 ±0.04	0.041 ± 0.04(NS)	0.035 ± 0.05(NS)	0.043 ± 0.04(NS)	0.049± 0.05(NS)	0.044 ± 0.04(NS)	0.03 ± 0.03(NS)

value are mean ± SD , * $P < 0.05$, NS = non significance

DISCUSSION

The present study implied the existence of impaired antioxidant systems in Thai chronic renal failure patients as the results of their were compared to that of the normal subjects. The reduced red blood cell vitamin E levels corresponded with prior results[10] . Our data indicated that CRF patients has a higher rate of lipid peroxidation.The lipid peroxidation product : MDA per nephron was increased significantly in both plasma and urine.

Plasma α-tocopherol (vitamin E) is readily influenced by the plasma lipid concentration[11]. Our results agreed with previous publication indicating that the plasma levels of α-tocopherol are in normal range in Thai chronic renal failure. Our results also confirmed the assumption that the content of α-tocopherol in red blood cells is significantly lower than in the normal level. Dietary deficiency in α-tocopherol leads to decrease GFR, nephron enlargement and tubulointerstitial disease[12]. There are several works focused on the increased usage of α-tocopherol by red blood cell membrane in such patient[13]. Studies on the depletion of α-tocopherol on the hematopoietic system of several species has showed the changes in red blood cell mass, red cell size , and the sensitivity of the red cell to hyperoxia and peroxide[4].

α-Tocopherol status has been traditionally determined by measuring plasma α-tocopherol levels but only a few investigation of α-tocopherol levels in red cells was obtained owing to technical difficulties of the assay[13]. Erythrocytes α-tocopherol may be good indicators of α-tocopherol nutritional status and refect the content of α-tocopherol in membranes[14].

CONCLUSION

The result of the present study should be concluded that Thai chronic renal failure patients were subjected to oxidative stress as indicated by deficiency in α-tocopherol in red blood cell and increasing of lipid peroxidation. Suffering from α-tocopherol deficiency or under prooxidant stress has been claimed to biochemical and subcellular damage by lipid peroxidation[15]. Oxidative damage to polyunsaturated lipids in tissue membranes (lipid peroxidation), a free radical process, is a widely accepted mechanism for cellular injury. Malondialdehyde is one of the products of lipid peroxidation which appears to be produced in relatively constant proportion to lipid peroxidation. It is therefore a good indicator of the rate of lipid peroxidation[16]. Our study also suggested that oral supplementation of α-tocopherol in chronic renal failure patients should be further study.

REFERENCES

1. Hayslett JP. Functional adaptation to reduction in renal mass.*Physiological reviews.*1979;59 (1) :137-164.
2. Johnson RJ,Lovett D,Lehrer RI,Couser EG,Klebanoff SJ.Role of oxidants and proteases in glomerular injury. *Kidney Int.*1994;45:352-359.
3. Nath KA,Fischereder M, Hostetter TH.The role of oxidants in progressive renal injury. *Kidney Int.*1994;45 (Suppl 45):111-115.
4. Kayden HJ, Bjornson L. The dynamics of vitamin E transport in the human erythrocyte. *Ann NY Acad Sci* 1972; 203 :127-139.
5. Vanella A et al. Superoxide dismutase activity and reduced glutathione content in erythrocytes of uremic

patients on chronic dialysis. *Acta Haematol*.1983;70:312-315.
6. Nath KA,Hostetter MK, Hostetter TH.Pathophysiology of chronic tubulo-interstitial disease in rat : interactions of dietary acid load, ammonia and complement component C_3. *J Clin Invest* .1985 ; 76 : 667-675.
7. Laurent B,Ardaillou R. Reactive oxygen species : production and role in the kidney. *Am J Physio. 1986*; 251: 765-776.
8. Miller KW, Yang CS. An Isocratic high performance liquid chromatography method for the simultaneous analysis of plasma retinol α-tocopherol, and various carotenoids. *Anal Biochem* .1985 ; 145 : 21-26.
9. Sierra C, Pastor Mc, Ramon M de. Liquid chromatography determination of α-tocopherol in erythrocytes. *Clin Chim Acta* .1992 ; 208 : 119-126.
10. Paul JL,Man NK,Moatti N,Raichvarg, D. Membrane phospholipid peroxidation in renal insufficiency and chronic renal hemodialysis. *Nephron*.1991;12(1) : 4 -7.
11. Horwitt HK,Harvey CC,Dahm CH,Searcy MT.Relationship between tocopherol and serum lipid levels for determination of nutritional adequacy. *Ann NY Acad Sci* .1972;203: 233-236.
12. Nath KA,Salahudeen AK,Clark EC, Hostetter MK, Hostetter TH. Role of cellular metabolites in progressive renal injury. *Kidney Inter.* 1992 ; 42 (Suppl 38):103-113.
13. Ono K. Effects of large dose vitamin E supplementation on anemia in hemodialysis patients.*Nephron.* 1985; 40(4): 440-445.
14. Lehmann J. Comparative sensitivities of tocopherol levels of platelets, red blood cells and plasma for estimating vitamin E nutritional status in rats. *Am J Clin Nutr* .1981 ; 34 : 2104-2110.
15. Tudhope GR, Hopkins J. Lipid peroxidation in human erythrocytes in tocopherol deficiency. *Acta haematol.* 1975 ; 53 : 98-104.
16. Tappel Al. Vitamin E and free radical peroxidation of lipids. *Ann NY Acad Sci. 1972 ; 203 : 12-28.*