IOSR Journal Of Pharmacywww.iosrphr.org
(e)-ISSN: 2250-3013, (p)-ISSN: 2319-4219
Volume 7, Issue 6 Version. 1 (June 2017), PP. 09-14

Investigating effect of vitamin C on oxidative stress in patients undergoing chronic ambulatory peritoneal dialysis

Shokouh Shayanpour[1], Heshmatollah Shahbazian[1], Farshid Padyab[1,*], Siamak Baghaei[2], Mehdi Zarei[3]

[1]*Chronic Renal Failure Research Center, Faculty of Medicine, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran.*
[2]*MD, Internist, The director of Continued Medical Education, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran.*
[3] *Assistant professor, Department of Food Hygiene, Faculty of Veterinary Medicine, Shahid Chamran University of Ahvaz, Ahvaz, Iran.*
***Corresponding author:** Farshid Padyab, Chronic Renal Failure Research Center, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran. Dr_padyab@yahoo.com*

Objective: *Oxidative stress increases in chronic renal failure patients and exacerbates during dialysis. The incidence of cardiovascular diseases are high in CRF patients and oxidative stress is a risk factor for CVD in CRF patients. The aim of this study was to assess the effect of antioxidant therapy including vitamin C on two markers of oxidative stress including MDA (Malondealdehyde) and TAC (Total Antioxidant Capacity) in patients undergoing peritoneal dialysis (PD).*
Materials and methods: *This is a prospective clinical trial conducted on 40 patients undergoing PD and were randomly divided into two groups of intervention (n=20) and placebo (n=20). The intervention group received 250 mg/day vitamin Corally for 8 weeks. Placebo group received placebo which was similar to intervention drug in terms of shape and color. The CRP, Albumin, TG, HDL, LDL, Uric acid were measured at baseline. The serum and peritoneal MDA and TAC levels were measured at baseline and at the end of the study.*
Findings: *The mean serum TAC and MDA decreased in compare to before study in intervention group, however they had not statistically difference. The mean peritoneal TAC significantly increased in compare to before study in both intervention and placebo groups, while the mean peritoneal MDA significantly decreased in compare to before study in both intervention and placebo groups.*
Conclusion: *Finally, we concluded that 8 weeks treatment with 250 mg/day vitamin C in combination with 250 mg B6, orally, could not change serum oxidative stress markers in serum, increased mean peritoneal TAC and decreased mean peritoneal MDA.*

Keywords: *Peritoneal dialysis, oxidative stress, TAC, MDA*

I. INTRODUCTION

Patients with end-stage renal disease (ESRD) need to undergo renal replacement therapy in the form of hemodialysis (HD), peritoneal dialysis (PD) or renal transplantation (1). PD is an established treatment in a limited time(2, 3). In patient with chronic renal failure (CRF). Long-term PD is associated with prolonged exposure of the peritoneal membrane with PD solution. PD solution contains high concentrations of glucose which increase cellular reactive oxygen species (ROS) (4). ESRD patients are characterized by increased OS (oxidative stress) and dialysis exacerbates OS (5, 6). The TAC is a useful marker of total defense system against free radicals for assessing oxidative stress status andis found in plasma which results from the cooperation of enzymatic antioxidants and scavenger antioxidants(7). The MDA is generated from lipid peroxidation in plasma due to hypertriglyceridemia in renal failure patients(8). The most important antioxidants that protect from oxidant damage include some high molecular weight such as erythrocyte superoxide (SOD) and gluthathione peroxidase (GPx); and vitamins A, C, and E as nonenzymatic antioxidants(1, 9, 10). Oxidative stress in dialysis patients leads to atherosclerosis and cardiovascular diseases. Moreover, it is common cause of CVD mortality.

The aim of this study was to assess the effect of antioxidant therapy including vitamin C and vitamin B6 on two markers of oxidative stress including MDA and TAC in PD patients.

II. MATERIALS AND METHODS

This study is a randomized, double blind, placebo-controlled trial carried out on 40 patients (18 females and 22 males) on continuous ambulatory PD from Hemodialysis Department at Imam Khomeini Hospital, after taking informed written consent.

The patients were not discriminated for the primary disease leading to CRF.

Inclusion criteria were age above 50 years, dialysis duration at least 6 months, the regular program for CAPD (Continuous Ambulatory Peritoneal Dialysis) for 3 months with 3-4 times exchange using two liter hypertonic or isotonic glucose solutions oricodextrin which added to peritoneal fluid solution.

Exclusion criteria were peritoneatitis or other infections, existence of inflammatory diseaseswithin weeks after blood sampling, chronic inflammatory diseases, patients who use NSAID,allopurinol or antioxidant medication, insufficient regulation of diabetes (HbA1c>7.5%).

The PD patients were randomly assigned into two groups of intervention (n=20) and placebo (n=20) using random list. Patients and the person who assessed the patients were unaware about the study protocols (double blind study). Firstly, subjects in two groups entered an antioxidant therapy washout period for 8 weeks. And then the intervention group received vitamin C in a dose of 250 mg/day and B6 in a dose of 250mg/day, orally, for two 8 weeks, while patients in placebo group received placebo for 8 weeks. The vitamin C and placebo were similar in regards of shape and taste.

The biochemical parameters have been investigated included albumin, CRP, HDL, LDL, total cholesterol, TG, uric acid at the baseline for two groups. Plasma MDA and TAC levels and also peritoneal fluid MDA and TAC levels were measured to assess the effect of vitamin C and B6, in combination, therapy on the OS status.

The venous blood sampling was drawn before starting the dialysis procedure. The samples were collected into empty sterile standard tubes then immediately centrifuged at 300 rpm for 15 minutes to separate plasma to maintain it at -80 0C for analysis time. The MDA and TAC were measured using spectrophotometric method.

Statistical analysis

Data are summarized as mean ± SD or number (percentage). Normality of the data was checked by Shapiro-Wilk test which showed that there were a normal distribution between data, except for uric acid. Compare mean analysis on biochemical characteristics was performed with student's t test, except for uric acid which tested by Mann-Witney test. The data were analyzed using SPSS software version 22.

Ethics issues

The study protocol was approved by ethical committee of Ahvaz Jundishapur University of Medical Sciences and received ethical code of IR.AJUMS.REC.1395.583. This research conducted as a thesis of Farshid Padyab, MD, for completing specialty certificate in internal medicine. The study protocol also was registered at Iran clinical trial registry which can be sited in irct.ir with registry number of IRCT2016050227675N2. All patients assigned written informed consent.

III. FINDINGS

The demographic information is shown on table 1.

As shown in Table 2, biochemical characteristics including LDL, HDL, TG, Alb, uric acid and CRP had not statistically significant difference between two groups.

The serum and peritoneal TAC and MDA at baseline and at end of the study are shown in Table 1.

The mean serum TAC were similar between two groups at baseline (P=0.1). The mean serum TAC was 359.9±87.85 at baseline in intervention group. This changed to 329 ± 55.02 after 8 weeks off the intervention. However, this reduction was not statistically significant (P=0.9). In contrast, the mean serum TAC in placebo group increased from 322.27 ± 51.14 at baseline to 346.2 ± 63.6 at end of the study, but it was not statistically meaningful (P=0.6). The mean serum TAC at the end of the study was similar between two groups (P=0.5) (Figure 1). There was no significant difference in mean MDA serum between intervention and placebo group at baseline (P=1.00). The serum MDA serum showed slight decrease for both intervention and placebo group from baseline to the end of the study (P=0.7 and P=0.2, respectively).

There was no statistically difference in mean MDA serum among intervention and placebo subjects at the end of the study(4.2 ± 1.3 and 4.69 ± 1.6, P=0.8) (Table 3).

The subjects' mean peritoneal TAC was similar at baseline between intervention and placebo groups (P=1.000). There was a sharp increase in mean peritoneal TAC from baseline to the end of the study in intervention group (124.4 ±51.28and 322.79 ± 38.98, P<0.001) and also in placebo group (116.99 ± 40.7and322.8 ± 37.98, P<0.001).

The mean peritoneal MDA decreased noticeably two months after intervention comparing to preintervention period (11.6± 2.17and4.2 ± 1.4, P<0.0001). This value showed similar reduced pattern in placebo group (P<0.0001) (Table 3).

IV. DISCUSSION

The present study demonstrate that 8 weeks treatment trial of daily oral antioxidant containing 250 mg vitamin C and 250 mg vitamin B6, in combination, could significantly increase the TAC and decrease the MDA, the markers of antioxidant stress, in peritoneal effluent, but the therapy did not significantly change TAC and MDA in the serum.

This no significant decrease the serum TAC and MDA levels after treatment by vitamin C may be explained by a flow of vitamin C from serum into a peritoneal cavity which results in increased bioavailability of vitamin in peritoneal effluent. Moreover, this phenomenon can explain why peritoneal MDA decreased significantly in intervention group after treatment in compare to before treatment (11). However, we cannot explain similar decrease in peritoneal MDA in placebo group.

Oxidative stress has a main role in pathogenesis of atherosclerosis and development of cardiovascular diseases. Oxidative stress occurs in CRF and exacerbates along with PD or HD (12).

The evidence for protective effects of antioxidants has been provided from some observational studies. However, large clinical trials include HOPE study and Heart Protection study have reported no effect on CVDs. It has been proposed that antioxidant are useful in patients with chronic oxidative stress rather than general CVD patients (13).

Oxidative stress defense system supported from endogenous and exogenous sources (nutrient and treatment). Nutritional antioxidant includes vitamin E, vitamin C and beta-carotene.

Several studies have investigated the effect of oral or parenteral antioxidant agents in hemodialysis (HD) patients, to reduce oxidative stress, only 4 out of 11 studies, using vitamin C orally, led to improved OS (14, 15). Even the studies using various antioxidants or using vitamin E –coated dialysis membranes have inconclusive results on oxidative stress (4).

To the best of our knowledge, current study is the first to assess vitamin C and B6 on two groups of PD. Most studies on oxidative therapy have been conducted on HD patients and also most investigated vitamin is vitamin E. Moreover, most these studies have single HD or PD patients (compared with healthy individuals). In addition, there are a lack of data on TAC and MDA levels in groups of vitamin C-treated and placebo treated HD or PD patients. However, a study by Samouilidouet al., and several other study which has compared TAC and MDA levels before and after HD and PD treatment. They confirmed that the serum TAC and MDA levels are similar between HD and PD patients.

Our study is more similar with previous study by Boudouris et al., (12)except that they did not consider placebo in PD group, while we used vitamin C-treated PD patients in compare to placebo-treated PD patients. We observed that intervention had no effect on serum TAC and MDA levels, while significantly increased peritoneal TAC and decreased MDA levels. These finding are consistent with study by Boudouris et al.(12).

The doses of 100-200 mg/day are necessary to maintain normal values of serum vitamin C. Higher doses are associated with development of hyperoxalemia. In addition, vitamin B6 deficiency, which also common in CRF patients, increase significantly the oxalate products (15). It is notable that Coombs et al., noted that studies with no change in oxidative stress used the time period ranged from 4 to 12 weeks and dose of 250 mg per day (16-18).

Our findings regarding the effect of placebo was interesting. The placebo had similar effect on serum and peritoneal TAC levels and peritoneal MDA levels. While placebo-treated PD patients showed increased serum MDA level.

Our study have some limitation. The low sample size and not measuring the serum and peritoneal vitamin C levels are the most important limitations. Moreover, the causes led to peritoneal dialysis was not registered by authors.

Conclusion: Finally, we concluded that 8 weeks treatment with 250 mg/day vitamin C in combination with 250 mg B6, orally, could not decrease serum oxidative stress markers and increase antioxidant capacity, while, although we observed significant decreasing in peritoneal MDA and increasing in TAC, placebo group had same finding too that should be evaluated in further studies.

V. CONCLUSION

Finally, we concluded that 8 weeks treatment with 250 mg/day vitamin C in combination with 250 mg B6, orally, could not decrease serum oxidative stress markers and increase antioxidant capacity, while, although we observed significant decreasing in peritoneal MDA and increasing in TAC, placebo group had same finding too that should be evaluated in further studies.

Financial Disclosure

No financial interest related to the material in the article.

Finding/support

This project was supported by Ahvaz Jundishapur University of of Medical Sciences.

Acknowledgement
This research conducted as a thesis of Farshid Padyab, MD, for completing specialty certificate in internal medicine and is supported by Ahvaz Jundishapur University of Medical Sciences. We would like to thank Miss Marjan Nour Ali Jolak and Miss Masoumeh Khani, the personnel of Peritoneal Dialysis Department at Imam Khomeini Hospital.

Table 1. Demographic and biochemical characteristics.

		Intervention	Control	P value
Gender	Male	11 (55%)	11 (55%)	1.000
	Female	9 (45%)	9 (45%)	
Age (yr)	Mean ± SD	60.2 ± 7.9	59.02 ± 5.29	0.4
	Median	61	60	
	Min-max	50-78	53-80	
Dialysis duration (years)		2 ± 1.9	3± 0.1	0.6
PET	Low	4 (20%)	3 (15%)	0.5
	Low average	4 (20%)	5 (25%)	
	High average	7 (35%)	9 (45%)	
	High	5 (25%)	3 (15%)	

Table 2. biochemical characteristics

CRP	Positive		5 (25%)	6 (30%)	1.000
	Negative		15 (75%)	14 (70%)	
LDL (mg/dl)		Mean ± SD	102 ± 19.42	115.25 ± 28.85	0.09
		Median	100	115	
		Min-max	70-160	70-170	
HDL (mg/dl)		Mean ± SD	39.4 ± 7.85	39.8 ± 6.81	0.3
		Median	37.5	40	
		Min-max	30-55	30-55	
TG (mg/dl)		Mean ± SD	192.25 ± 14.09	186.25 ± 16.29	0.2
		Median	195	182.5	
		Min-max	165-220	160-220	
Albumin (g/l)		Mean ± SD	3.83 ± 0.19	3.89 ± 0.19	0.3
		Median	3.85	3.95	
		Min-max	3.5-4.1	3.5-4.2	
Uric acid (mg/dl)		Mean ± SD	3.75 ± 0.96	3.75 ± 1.01	0.9
		Median	4	4	
		Min-max	3-4	2-6	

Table 3. TAC and MDA serum and peritoneal levels

		Before	After	P value [a]	P value [b]	P value [c]
Serum						
TAC (µmol/l)	Intervention	359.9 ± 87.85	329 ± 55.02	0.5	0.9	1.00
	Placebo	322.27 ± 51.14	346.2 ± 63.6		0.6	
MDA (µmol/l)	Intervention	5.05 ± 2.0	4.2 ± 1.3	0.8	0.7	1.00
	Placebo	6.04 ± 2.09	4.69 ± 1.6		0.2	
Peritoneal						
TAC (µmol/l)	Intervention	124.4 ± 51.28	322.79 ± 38.98	1.00	0.001	1.00
	Placebo	116.99 ± 40.7	322.8 ± 37.94		0.001	
MDA (µmol/l)	Intervention	11.6 ± 2.17	4.2 ± 1.4	1.00	0.001	1.00
	Placebo	11.1 ± 2.01	3.9 ± 1.1		0.001	

[a] Before treatment-before placebo.
[b] Before treatment-after treatment or before placebo-after placebo.
[c] After treatment- after placebo.
MDA:malondialdehyde; TAC: total antioxidant capacity.

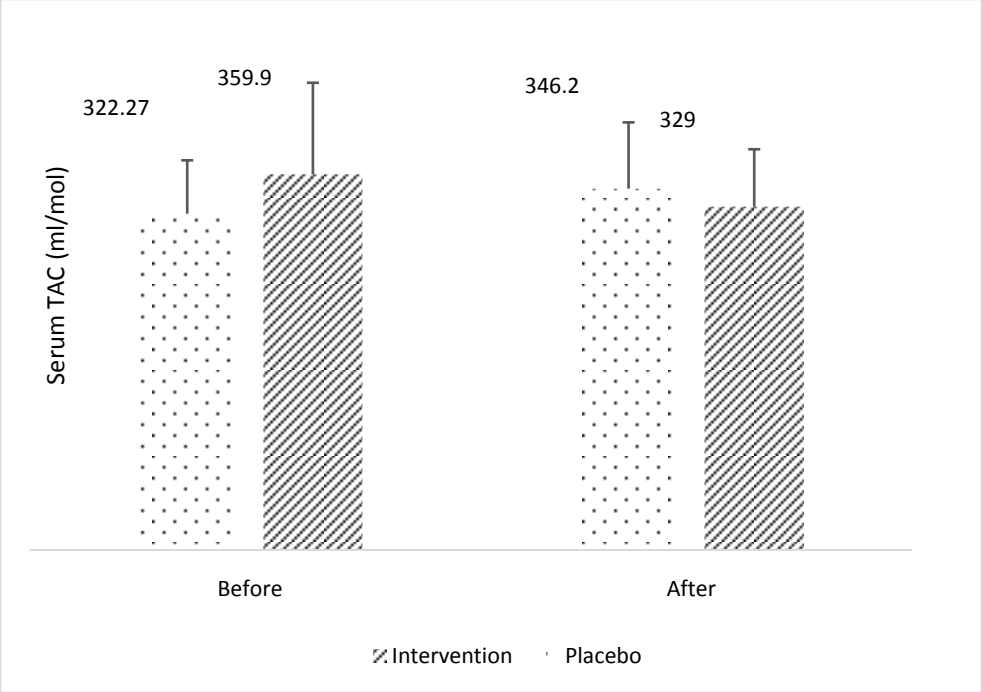

Figure 1. Serum TAC levels before and after study.

REFERENCES

[1]. Montazerifar F, Karajibani M, Sanadgol H, Hashemi M. Effect of peritoneal dialysis on antioxidant defense system and oxidative stress. Hong Kong Journal of Nephrology. 2012 10//;14(2):33-7.
[2]. Sundl I, Roob JM, Meinitzer A, Tiran B, Khoschsorur G, Haditsch B, et al. Antioxidant status of patients on peritoneal dialysis: associations with inflammation and glycoxidative stress. Peritoneal Dialysis International. 2009;29(1):89-101.
[3]. Gokal R, Mallick N. Peritoneal dialysis. The Lancet. 1999;353(9155):823-8.
[4]. Kamgar M, Zaldivar F, Vaziri ND, Pahl MV. Antioxidant therapy does not ameliorate oxidative stress and inflammation in patients with end-stage renal disease. Journal of the National Medical Association. 2009;101(4):336-44.
[5]. Dursun B, Dursun E, Capraz I, Ozben T, Apaydin A, Suleymanlar G. Are uremia, diabetes, and atherosclerosis linked with impaired antioxidant mechanisms? Journal of Investigative Medicine. 2008;56(2):545-52.
[6]. Kocak H, Gumuslu S, Ermis C, Mahsereci E, Sahin E, Gocmen AY, et al. Oxidative stress and asymmetric dimethylarginine is independently associated with carotid intima media thickness in peritoneal dialysis patients. American journal of nephrology. 2008;28(1):91-6. PubMed PMID: 17914250. Epub 2007/10/05. eng.
[7]. Samouilidou E, Grapsa E. Effect of dialysis on plasma total antioxidant capacity and lipid peroxidation products in patients with end-stage renal failure. Blood Purif. 2003;21(3):209-12. PubMed PMID: 12784045. Epub 2003/06/05. eng.
[8]. Daschner M, Lenhartz H, Bötticher D, Schaefer F, Wollschläger M, Mehls O, et al. Influence of dialysis on plasma lipid peroxidation products and antioxidant levels. Kidney international. 1996;50(4):1268-72.
[9]. Montazerifar F, Hashemi M, Karajibani M, Dikshit M. Effect of combined vitamins C and E supplementation on oxidant/antioxidant status in hemodialysis patients. Mediterranean Journal of Nutrition and Metabolism. 2010;3(2):159-63.

[10]. Durak İ, Kaçmaz M, Elgün S, Öztürk HS. Oxidative stress in patients with chronic renal failure: effects of hemodialysis. Medical Principles and Practice. 2004;13(2):84-7.

[11]. Ahmadpoor P, Eftekhar E, Nourooz-Zadeh J, Servat H, Makhdoomi K, Ghafari A. Glutathione, glutathione-related enzymes, and total antioxidant capacity in patients on maintenance dialysis. Iran J Kidney Dis. 2009;3(1):22-7.

[12]. Boudouris G, Verginadis, II, Simos YV, Zouridakis A, Ragos V, Karkabounas S, et al. Oxidative stress in patients treated with continuous ambulatory peritoneal dialysis (CAPD) and the significant role of vitamin C and E supplementation. International urology and nephrology. 2013 Aug;45(4):1137-44. PubMed PMID: 23212145.

[13]. Himmelfarb J, Stenvinkel P, Ikizler TA, Hakim RM. The elephant in uremia: oxidant stress as a unifying concept of cardiovascular disease in uremia. Kidney international. 2002;62(5):1524-38.

[14]. Boudouris G, Verginadis II, Simos YV, Zouridakis A, Ragos V, Karkabounas SC, et al. Oxidative stress in patients treated with continuous ambulatory peritoneal dialysis (CAPD) and the significant role of vitamin C and E supplementation. International urology and nephrology. 2013;45(4):1137-44.

[15]. Coombes JS, Fassett RG. Antioxidant therapy in hemodialysis patients: a systematic review. Kidney Int. 2012 Feb;81(3):233-46. PubMed PMID: 21975860. Epub 2011/10/07. eng.

[16]. Eiselt J, Racek J, Opatrný Jr K, Trefil L, Stehlík P. The effect of intravenous iron on oxidative stress in hemodialysis patients at various levels of vitamin C. Blood purification. 2006;24(5-6):531-7.

[17]. Chan D, Irish A, Croft KD, Dogra G. Effect of ascorbic acid supplementation on plasma isoprostanes in haemodialysis patients. Nephrology Dialysis Transplantation. 2006;21(1):234-5.

[18]. Fumeron C, Nguyen-Khoa T, Saltiel C, Kebede M, Buisson C, Drüeke TB, et al. Effects of oral vitamin C supplementation on oxidative stress and inflammation status in haemodialysis patients. Nephrology Dialysis Transplantation. 2005;20(9):1874-9.