

**OXIDATIVE STRESS IN CHRONIC RENAL FAILURE****G.ARUNA^{1*} AND K.AMBIKA DEVI²**^{1*} *Department of Biochemistry, ASRAM medical college, Eluru- 534002, Andhra Pradesh, India.*² *Department of Biochemistry, ASRAM medical college, Eluru, Andhra Pradesh, India.***ABSTRACT**

Chronic renal failure applies to the process of continuing significant irreversible reduction in the nephron number and corresponds to Chronic Kidney Disease (CKD) with the classification of 3-5. Lipid peroxidation is a continuous chain reaction providing a continuous supply of free radicals. Vitamin E an antioxidant forms the 1st line of defense against peroxidation of membranes of the cell and subcellular organelles. To compare Malondialdehyde, Vitamin E and other biochemical parameters in chronic renal failure with those of healthy controls. Sixty cases of both sexes with chronic renal failure from 35-60 years age group were selected. Control group includes 20 healthy individuals. Urea, Creatinine, Malondialdehyde, Vitamin E were compared in chronic renal failure with those of healthy controls. Creatinine Clearance was estimated by using Cock Croft and Gault formula to assess the renal function. The results were statistically analyzed by t- test and Pearson correlation. MDA levels were found statistically highly significant in CRF patients as compared to controls. Vitamin E levels showed significant decrease in CRF patients as compared to control group. The groups mean predicted Creatinine Clearance from the Cock Croft and Gault formula was 7.24 ml/min. Serum MDA and Serum Creatinine were positively correlated with severity and duration of CRF. Vitamin E was decreased in all the cases of CRF.

KEY WORDS :- Malondialdehyde, CRF, Vitamin E.**G.ARUNA**Department of Biochemistry, ASRAM medical college,
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INTRODUCTION

Chronic renal failure applies to the process of continuing significant irreversible reduction in the nephron¹⁹ number and corresponds to Chronic Kidney Disease (CKD) with the classification of 3-5¹⁹. Chronic renal failure implies longstanding and usually progressive, impairment in renal function²³. Lipid peroxidation is a continuous chain reaction providing a continuous supply of free radicals. Vitamin E an antioxidant forms the 1st line of defense against peroxidation of membranes of the cell and subcellular organelles. Creatinine level in serum appears to be a better index of the severity of the degree of failure in patients suffering from chronic renal failure, while serum urea concentration shows a better correlation with degree of failure in acute renal failure¹⁶. Serum malondialdehyde (MDA) level in CRF patients increases resulting in alteration of LDL. Malondialdehyde is the end product of lipid peroxidation, caused by the oxygen free radicals²³.

MATERIALS AND METHODS

Patients admitted in the Medical, Nephrology and Urology wards and outpatients of the said departments with a diagnosis of chronic renal failure in ASRAM HOSPITAL formed the subjects of this study. Sixty cases of both sexes with chronic renal failure from 35-60 years age group were selected. Control group includes 20 healthy individuals. Urea, Creatinine, Malondialdehyde, Vitamin E were compared in chronic renal failure with those of healthy controls. Creatinine Clearance was estimated by using Cock Croft and Gault formula to assess the renal function. Inclusion criteria:- Hypertensive Nephropathy, Diabetic Nephropathy, Analgesic induced Nephrology, Glomerulonephritis. Exclusion criteria: Osteoporotic disorders, Pregnant women, Acute Renal Failure. Blood Urea was estimated

quantitatively by Urease Glutamate Dehydrogenase method using commercial kit¹⁰, Serum Creatinine by Jaffe's method²⁴ using commercial kit, Serum Malondialdehyde by Thiobarbituric acid method by chemical analysis, Serum Vitamin E by Emmerie - Engel method by chemical analysis.

STATISTICAL ANALYSIS

Statistical analysis was done by t- test. Correlation coefficient was calculated using Pearson correlation. The results were expressed as mean \pm SD. The level of significance is assessed based on p value. p-value of < 0.05 was accepted as statistically significant.

RESULTS

The Serum levels of Urea, Creatinine, and MDA was significantly higher in cases when compared to controls. The levels of Serum vitamin E was significantly lower in cases when compared to controls (Table 1 & 2). The mean of MDA in normal control was 192 nmol% as compared with chronic renal failure cases 450.7 nmol% (Table 3). There was a great difference in the two means of MDA providing good accuracy in diagnosing chronic renal failure patients. Statistically significant increase in MDA levels was found in CRF Patients as compared to controls ($p < 0.001$) and statistically highly significant decrease in mean 4.17 mg/L vitamin E levels in CRF patients as compared to mean 10.9 mg/L vitamin E controls ($p < 0.001$) (Table 3). There was a correlation between serum MDA and serum Vitamin E levels in Chronic Renal Failure. Significantly negative correlation was observed between MDA and Vitamin E (Table 4). Highly significant positive correlation was observed between MDA and serum creatinine (Table 4).

Table1
Control Group

Sl No.	Age (years)	Sex	Urea (Normal range 15-40 mg/dl)	Creatinine (Normal range 0.7-1.5 mg/dl)	MDA (Normal range 171.5-343.15 nmol%)	Vit E (Normal range 10-12 mg/L)
1	38	M	12	0.65	186	12
2	45	M	14	0.55	185	10
3	40	M	12	1.0	213	11.5
4	48	M	25	0.5	240	12
5	36	M	16	0.6	180	11
6	46	F	16	0.5	160	11.5
7	50	M	15	1.0	162	10
8	43	M	12	0.9	160	11
9	45	M	26	1.3	186	12
10	37	F	12	1.1	154	10
11	44	M	14	1.0	162	11
12	52	F	14	0.55	182	10.5
13	39	M	20	0.75	213	11.5
14	55	M	22	0.8	198	10
15	34	F	15	0.8	186	11.4
16	56	M	20	0.8	210	11
17	39	M	18	1.0	178	10.6
18	42	M	27	1.1	240	11.5
19	45	M	33	0.9	266	11.2
20	46	M	28	1.0	179	10
Mean			18.55	0.840	192	10.985
SD			6.304	0.228	30.248	0.711
SEM			1.410	1.410	6.764	0.159

Table 2
Cases (or) study group

S.No.	Age (Years)	Sex	Urea (Normal range 15-40 mg/dl)	Creatinine (Normal range 0.7-1.5 mg/dl)	MDA (Normal range 171.5-343.15 nmol%)	Vit E (Normal range 10-12 mg/L)	CG formula < 15 ml/ min
1	50	M	170	6.8	400	6	12.93
2	38	M	160	6.2	373	8.9	8.75
3	42	M	150	8	398	7.2	6.72
4	45	M	156	9.25	375	7	10.09
5	48	M	160	6	372	8	8.66
6	37	M	170	5.3	399	5.4	17.18
7	43	F	174	5.9	453	4.2	11.56
8	51	M	175	11.25	400	6.4	9.66
9	43	M	182	8.52	455	5.3	11.09
10	45	M	195	6.6	480	4.5	8.28
11	48	F	195	12.2	423	3.7	6.42
12	45	M	180	11	401	4.8	7.14
13	38	M	190	6.9	448	3	15.02
14	37	F	195	8.8	412	2	7.69
15	42	M	200	7.8	480	5	11.22
16	46	M	200	9.6	453	4.8	9.31
17	39	M	209	7.7	452	5	9.64
18	51	M	205	9.5	422	3.6	6.78
19	54	M	214	14	493	4.2	4.1
20	49	M	240	13.6	490	4	4.5
21	44	M	245	15	520	2.9	4.84
22	47	M	258	13.2	560	2.2	5
23	51	M	265	15	506	3.6	4.49
24	42	M	240	7	453	4.5	8.82
25	45	M	260	7.8	480	4.4	11
26	57	F	264	14	533	2	4
27	39	F	295	18	600	1.8	3
28	36	M	280	14	525	2.4	3.6
29	53	M	312	13	540	1.9	4.3
30	52	M	320	18	586	1.4	2.8

31	50	M	160	8	398	7	6.72
32	37	F	190	9	423	4	4.8
33	60	M	180	8.6	500	4	4.8
34	58	F	178	8	456	3.4	4.6
35	52	M	150	6	343	3.4	6
36	54	F	158	6.2	280	2.8	5.6
37	38	M	148	4	370	4.6	6.2
38	48	M	190	6.9	448	3	12
39	49	M	240	13	490	4	4.5
40	52	F	175	11	400	6	9
41	56	F	264	12	560	2	5
42	60	M	214	14	490	3	4.8
43	39	M	160	6.2	360	9	8
44	49	M	160	6	372	7	9
45	46	M	240	14	500	2.5	5
46	39	F	240	7	450	4.5	8
47	58	F	260	12	530	2.2	4.8
48	60	M	258	11.6	500	2	4.2
49	60	M	230	12	490	3	4.8
50	58	M	162	6	370	7	8
51	60	M	190	7	500	2	4.6
52	59	M	110	6	420	2.8	6
53	60	M	124	5.8	500	3	4.8
54	39	M	124	5.8	490	3	5
55	60	M	175	4	400	6.4	9.6
56	58	M	158	5	360	4	9.4
57	52	M	182	8	400	3.5	9.7
58	36	F	160	6	410	5	7
59	60	F	200	9.6	450	4.8	9.8
60	60	M	220	10	500	2	4.2
Mean			200.98	9.31	450.70	4.18	7.24
SD			47.1	3.5	65.1	1.9	3.0
SEM			6.08	0.45	8.4	0.25	0.39

TABLE 3

	Urea mg%	Creatinine mg%	MDA nmol%	Vit E mg / L
Controls Mean \pm S.D.	18.5 \pm 6.3	0.84 \pm 0.22	192 \pm 30.2	10.9 \pm 0.71
Cases Mean \pm S.D.	200.98 \pm 47.1	9.31 \pm 3.5	450.7 \pm 65.1	4.17 \pm 1.9
SE (diff between Means)	6.24	0.4546	10.7813	0.2922
t - Value	29.2435	18.6318	24.00	23.032
P - value	< 0.001 Highly significant	< 0.001 Highly significant	< 0.001 Highly significant	< 0.001 Highly significant

TABLE 4
Pearson Correlation of Serum Creatinine MDA Vs Vitamin E

Variable	Controls			Cases	
	Person Correlation	(r)	P value	Person Correlation (r)	P value
MDA Vs Serum Creatinine	r = -0.06		P < 0.001 Highly Significant	r = 0.71	P < 0.001 Highly Significant
	t = 4.26			t = 7.618	
MDA Vs Vitamin -E	r = 0.42		P < 0.1 Significant	r = -0.66	P < 0.001 Highly Significant
	t = 1.96			t = 6.6937	

DISCUSSION

The present study includes the analysis of Urea, Creatinine, MDA, Vitamin E in serum and calculation of creatinine clearance using Cockcroft Gault formula in Chronic Renal Failure patients. A total of 80 subjects have been included in the present study. They were classified into controls (20 subjects), Chronic Renal Failure (60 Subjects) of the same age group (30-60 years). The Serum levels of Urea, Creatinine, and MDA was significantly higher in cases when compared to controls. The levels of Serum vitamin E was significantly lower in cases when compared to controls (Table 1 & 2). The data suggests that there was a correlation between serum MDA and serum Vitamin E levels in Chronic Renal Failure. Other renal function tests performed to support the Clinical diagnosis include blood urea and serum creatinine. The present study indicates lipid peroxidation was a continuous chain reaction providing a continuous supply of free radicals that initiate further peroxidation. The mean of MDA in normal control was 192 nmol/L% as compared with chronic renal failure cases 450.7 nmol/L% (Table 3). There was a great difference in the two means of MDA providing good accuracy in diagnosing chronic renal failure patients. Student 't' test was done for comparison of data. In the present study, statistically significant increase in MDA levels was found in CRF Patients as compared to controls ($p < 0.001$) and statistically highly significant decrease in mean 4.17 mg/L vitamin E levels in CRF patients as compared to mean 10.9 mg/L vitamin E controls ($p < 0.001$) (Table 3). Kidney failure has a number of causes, lipid peroxidation of glomerular membrane due to free radicals, being one amongst them. In their study Dr. Suresh C.R. Wilma Delphine Sinia and Rajini Agarwal statistically found a significant increase in MDA levels in CRF patients as compared to controls ($p < 0.001$).³⁶ MDA showed significant positive correlation with serum creatinine ($r = 0.794$) in CRF patients. In this study also MDA showed significant positive correlation with serum creatinine ($r = 0.71$) in CRF patients and MDA showed significant

negative correlation with serum vitamin E ($r = -0.66$) in CRF patients (Table 4). Significantly negative correlation was observed between MDA and Vitamin E (Table 4). The same findings were also observed by Ratna Priya and Vasudha K.C. in their journal Reactive Oxygen species play a significant role in pathogenesis of chronic renal failure.³² Highly significant positive correlation was observed between MDA and serum creatinine (Table 4). Antioxidant (Vitamin E) forms the 1st line of defense against peroxidation of membranes of the cell and subcellular breaking antioxidant. Uremia related anorexia, vomiting or diarrhea plays a major role in vitamin E depletion due to deficient intestinal absorption. The reduction in plasma Vitamin E levels was also caused by the consumption of Vitamin E during free radical generation. Vitamin E was the main non enzymatic antioxidant of cellular membranes and vitamin E protects plasma lipids from peroxidation.^{11, 25} The groups mean predicted creatinine clearance from the Cockcroft and Gault formula was 7.24 ml/min (Table 2). In clinical practice, the Cockcroft and Gault formula has also been used to predict GFR. The Cockcroft and Gault formula, a simple, widely used and recommended means to assess renal function. Cockcroft and Gault formula can be calculated using a simple calculator or by mental arithmetic.³⁹

SUMMARY AND CONCLUSION

Chronic renal failure one of the commonest kidney disease was associated with Oxidative stress. In this study, it was found that serum MDA and serum creatinine were positively correlated with severity of CRF and vitamin E was decreased in all the cases of chronic renal failure. At present, K/DOQI guidelines suggest the Cockcroft and Gault formula should be used in clinical practice. The Cockcroft and Gault formula was devised to predict creatinine clearance.⁴³

Future Scope of Study

Malondialdehyde can be clubbed with the routine laboratory investigations like estimation of Blood urea, serum creatinine because of its direct involvement in glomerular pathology and

its simplicity of measurement for screening renal failure. The present study was simple, specific and sensitive. It can be performed by using a simple colorimeter and can be used for detection and screening of kidney damage.

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