International Journal of Health Sciences and Research
www.ijhsr.org ISSN: 2249-9571

Original Research Article

Oxidative Stress in Type 2 Diabetes Mellitus Patients with Microalbuminuria

Shailja Chambial[1*], Rajesh Kumar Sharma[2], Bharat Kumar Gupta[3], Akash Gupta[4], Debapriya Bandhopadhyay[5]

[1]Senior Resident, Department of Biochemistry, AIIMS, Jodhpur, Rajasthan, India
[2]Additional professor, Department of Physiology, AIIMS, Jodhpur, Rajasthan, India
[3]Professor, Department of Biochemistry, Subharti Medical College, Swami Vivekananda Subharti University, Meerut, Uttar Pradesh, India
[4]Assistant Professor, Dept. of Biochemistry, Saraswathi Institute of Medical Sciences, Hapur, Uttar Pradesh, India
[5]Assistant Professor, Department of Biochemistry, AIIMS, Bhubaneshwar, Orissa, India

[*]Correspondence Email: shailjachambial@yahoo.com

Received: 27/06//2013 *Revised: 16/09/2013* *Accepted: 26/09/2013*

ABSTRACT

Background & Aims: To find out whether any correlation exists between the levels of Plasma Malondialdehyde (MDA) and Whole blood Glutathione peroxidase (GPx) in patients of type 2 DM with microalbuminuria.
Methods: The study was carried out in total of 100 type 2 diabetes mellitus patients with & without microalbuminuria and 50 healthy controls. Fasting serum sugar (FSG), lipid profile, HbA1c%, urea, creatinine, MDA & GPx were measured using standard methods.
Results: MDA was significantly higher in type 2 DM with microalbuminuria in comparison to type 2 DM without microalbuminuria.
In type 2 DM with microalbuminuria the albumin creatinine ratio (ACR) was significantly correlated with FSG($r=0.749$, $p<0.001$), HbA1c% ($r=0.799$, $p<0.001$), MDA($r=0.695$, $p<0.001$), cholesterol($r=0.716$, $p<0.001$), LDL-Cholesterol ($r=0.621$,$p<0.001$)though ACR & GPx were not found to be significantly correlated ($r= -0.169$, $p=0.241$). In the same group HbA1c% was significantly correlated with MDA ($r=0.791$, $p<0.001$). We did not find any significant correlation between MDA & GPx levels in type 2 DM with microalbuminuria.
Conclusion: Oxidative stress, hyperglycemia & microalbuminuria go hand in hand in type 2 diabetes mellitus.

Key words: MDA, GPx, Type 2 DM, Microalbuminuria, Albumin Creatinine Ratio.

INTRODUCTION

According to the WHO, there were 31.7 million people suffering from diabetes in 2000 in India. This number is estimated to increase to an alarming level of 79.4 million by 2030.[1]

According to a population based survey the prevalence of overt nephropathy was found to be 2.2% and that of microalbuminuria was 26.9% in study conducted in south India.[2] Diabetic nephropathy (DN) has been classically defined as increased protein excretion in urine. Early stage is

characterized by a small increase in urinary albumin excretion (UAE), also called microalbuminuria or incipient DN. Screening for diabetic nephropathy must be initiated at the time of diagnosis in patient with Type 2 diabetes mellitus[3] since approximately 7% of them already have microalbuminuria at that time.[4]

Oxidative stress is considered to be a unifying link between DM and its complications including nephropathy. Lipid peroxidation is an oxidative damage involving the cell membranes, lipoproteins under condition of oxidative stress. One of the major decomposition products of lipid peroxidation is malondialdehyde (MDA)[5] which originates from breakdown of fatty acids and hence measuring the level of MDA in biological samples is a measure of oxidative stress. Antioxidant system in the body primarily works against the deleterious effects of oxidative stress. One of them Glutathione peroxidase (GPx), is selenium containing enzyme and has four known isozymes. The classical GPx (GPx-1) acts as cellular antioxidant and can metabolize hydrogen peroxide and a range of organic peroxides, including cholesterol and long-chain fatty acid peroxides.[6] Another isoform, GPx-3 originates mainly from kidney[7] and contributes to plasma GPx levels and acts mainly as extracellular antioxidant. It is generally believed that the protective effect of GSH against the oxidative breakdown of lipids is mediated through GPx by the reduction of endogenously formed hydroperoxides of unsaturated fatty acids to hydroxyl derivatives.[8] Thus, it may protect the proximal tubules of the kidney from localized peroxide-induced damage and hence it's relevance in nephropathy.

There are studies available relating oxidative stress, antioxidant status and microalbuminuria in Type 2 Diabetes Mellitus patients in literature but very few exploring fundamental causes of diabetic incipient nephropathy in North Indian population, especially in people of western U.P, hence present study was planned.

MATERIALS & METHODS

The present study was started after obtaining ethical clearance from the institutional committee. The informed consent from the patients was also taken. Patients attending in and out-patient department of Medicine of Subharti CSS hospital were included. They were diagnosed as type 2 Diabetes Mellitus clinically & on the basis of the criteria laid down by American Diabetic Association (ADA) 2007.

Patients with blood pressure >130/80mm Hg, other causes of proteinuria like fever, urinary tract infection, prostatitis, congestive heart failure and hematuria etc., those on Angiotensin Receptor Blocker & ACE inhibitors. & patients on Vitamin A, C and E supplements, drug induced Diabetes & gestational Diabetes Mellitus were excluded from the study.

Albumin in urine was measured using Nyocard albumin kit (Axis Shield, Scotland). Urine creatinine was estimated after dilution of the same spot urine sample using Vitros 250 auto analyzer. The urine albumin creatinine ratio was calculated and value less than 30mg/g creatinine was considered normal and 30-300 mg/g creatinine was considered as micro-albuminuria. Type 2 DM patients with persistent microalbuminuria were selected and their fasting venous blood sample was collected for routine investigations like fasting serum glucose, lipid profile, urea, creatinine which were done on Vitros 250 autoanalyser. Glycosylated hemoglobin (HbA1c%) was estimated using Nyocard HbA1c kit. Sample was stored at -80^{0}C in a deep freezer for estimation of MDA & GPx.

On the basis of microalbuminuria subjects were categorized into:-
Group 1: age and sex matched 50 healthy volunteers (n=50)
Group 2: patients of type 2 Diabetes mellitus without microalbuminuria (n=50)
Group 3: patients of type 2 Diabetes mellitus with microalbuminuria (n=50)
MDA was estimated in plasma using OxiSelect™ TBARS Assay Kit & Randox enzyme assay kit was used for measurement of Glutathione peroxidase in whole blood. [9]
Data was statistically analyzed using the software Graphpad Instat (U.S.A.).The data was analyzed using unpaired Student's T test. $p<0.05$ was considered significant

RESULT

The result is depicted in the given tables.

Table I: Age, & gender distribution among controls (Group 1), diabetic without microalbuminuria (Group-2) and diabetic with microalbuminuria (Group-3).

	Group 1 Mean ± SD (n=50)	Group 2 Mean ± SD (n=50)	Group 3 Mean ± SD (n=50)
Age (years)	48± 8.3	46± 10.2	54 ± 10.4
Sexdistribution:			
Male	30	28	33
Female	20	22	17

DISCUSSION

In our study HbA1c% levels & fasting serum glucose values though higher in group 3 but were not statistically significant when compared with group 2 (p value >0.05).
The urea & creatinine level was not statistically different in the three groups and the analytes in all the three groups were within the reference range.

Total cholesterol values in group 2 and 3 were significantly raised when compared with the values in group 1 but no significant difference between group 2 and 3 was observed. Similar observations were made by several other workers also.[10-14] It is likely that excretion of protein in urine as in nephrotic syndrome with the resultant hypoalbuminemia leads to an upregulation of 3-hydroxy- 3-methylglutaryl CoA reductase with a consequent hyper-cholesterolemia.[15]

Triglycerides levels were significantly higher in group 3 when compared with group 1 and 2 and higher in group 2 on comparison with group 1 ($p<0.01$) Similar findings were also observed by Pasupathy et al.[16]

HDL- cholesterol values of group 2 and 3 are significantly lower than in group1 ($p<0.01$).The values in group 2 are higher but statistically not significant on comparison with group 3.Similar decreasing trend in HDL values was seen by other workers.[10,12,14,17]

LDL cholesterol values in group 2 and 3 were significantly higher when compared with the values in group 1($p<0.01$) though the values in group 3 were higher but statistically not significant when compared with values in group 2. Our findings are supported by various studies.[10-14]

The MDA levels were significantly higher in group 2 when compared with group 1 (p value<0.01). The MDA levels were significantly higher in group 3 when compared to group 1 and group 2 (p value<0.01, both groups).Many studies support this finding.[17-21] Malondialdehyde (MDA) is frequently measured as an indicator of oxidative stress in vivo. In type 2 DM patients there is increased level of MDA as hyperglycemia results in the generation of ROS, increasing oxidative stress in affected tissues, which are damaged by the consequent activation of nuclear factor kappa B, along with AGE formation and activation of the protein kinase C, sorbitol and hexosamine pathways.[21-24]

The GPx levels were significantly lower in group 2 and group 3 when compared with group 1 (p value<0.01). No significant difference was observed amongst values of group 3 and group 2 (p value

>0.05). Similar results were observed by others.[18] The decreased GPx activity in type 2 DM patients with microvascular complications was observed by Kusuvulu et al. They stressed that superoxide dismutase is required for scavenging superoxide radicals [25] and decreased activity of SOD leads to increased levels of superoxide radicals which will cause inhibition of GPx.[26] Significantly decreased GPx mRNA expression along with other antioxidant enzymes has been observed in patients of diabetic nephropathy under hyper-glycemic conditions.[27] Patients of diabetes with and without microalbuminuria in the present study had similar level of fasting serum glucose and HbA1c% .This indicates that glycoxidation induced oxidative stress is not different in these two groups. This could be a possible explanation for our finding.

Table II: Mean and SD of biochemical parameters in controls (Group 1), diabetic without microalbuminuria (group-2) and diabetic with microalbuminuria (Group-3).

	Group 1 Mean ± S D(n=50)	Group 2 Mean ± S D(n=50)	Group 3 Mean ± S D(n=50)
HbA1c(%)	4.25±0.80	7.94±1.22 [a]	8.44±1.52 [b]
FSG(mg/dl)	85.4±12.8	197± 47.53 [a]	218±53.1 [b]
Urea(mg/dl)	19.9±4.82	19.52± 6.32	21.5±7.27
Creatinine (mg/dl)	0.61± 0.25	0.64± 0.28	0.68± 0.28
T.Cholesterol(mg/dl)	182.076±15.81	227.92± 26.69 [a]	229.92±22.14 [b]
HDL -c(mg/dl)	45.46± 5.84	38.08± 5.45 [a]	37.22±4.72 [b]
LDL-c (mg/dl)	107.1676±17.35	153.563± 23.98 [a]	154.57±23.84 [b]
Triglycerides (mg/dl)	136.87± 10.94	177.99±24.44 [a]	215.648±34.38 [b*]
Albumin Creatinine Ratio(mg/g)	10.98±3.74	19.74±5.01	91.916±33.51 [b*]
Malondialdehyde (nmol/mL)	1.34± 0.58	4.638± 1.45 [a]	6.47±1.63 [b*]
Glutathione Peroxidase (U/L)	7699.768±731.63	5391.76.±886.23 [a]	5266.26±970.197 [b]

[a] = Mean ± SD comparison between group 2 & 1 is significant (p<0.05); [b] = Mean ± SD comparison between group 3 & 1 is significant (p<0.05); [*] = Mean ± SD comparison between group 3 & 2 is significant (p<0.05);

Table 3 below shows the correlation between different variables in Type 2 DM subjects with microalbuminuria. In this group albumin creatinine ratio was significantly correlated with HbA1c%. Many workers have observed similar findings indicating that hyperglycemia in type 2 DM is associated with microalbuminuria and HbA1c is an independent risk factor for development of incipient nephropathy in Type 2 DM patients. [11,13,27-34]

Table III: Correlation between different variables in Group 3 (diabetics with microalbuminuria).

S.No	Parameter	r value	p value
1.	ACR vs HbA1C%	0.799	<0.001
2	ACR vs FSG	0.749	<0.001
3.	ACR vs MDA	0.695	<0.001
4.	ACR vs GPx	-0.169	0.241
5.	ACR vs Cholesterol	0.716	<0.001
6.	ACR vs HDL	-0.256	0.721
7.	ACR vs Triglyceride	0.136	0.347
8	ACR vs LDL	0.621	<0.001
9	HbA1C% vs MDA	0.791	<0.001
10	HbA1C% vs GPx	-0.241	0.092
11	MDA vs GPX	-0.215	0.133

ACR and malondialdehyde values were also significantly correlated in this group. This is an unique correlation which has not been found in any Indian study to our knowledge. Oxidative stress sustained in diabetes by hyperglycemia and glyco-oxidation products such as HbA1c and AGE and the absence of an appropriate compensatory response from the endogenous antioxidant network have been implicated in systemic endothelial dysfunction [26,35] and microalbuminuria is considered a marker of endothelial dysfunction.[36,37] Ha et al also showed that oxidative stress is one of the important mediators of vascular complications in diabetes including nephropathy.[38]

A negative trend was seen between ACR and GPx. Others have observed negative correlation between microalbuminuria and GPx in type 2 DM patients with microalbuminuria.[11,39]

Patients with uncontrolled Type 2 diabetes have severely deficient synthesis of glutathione (GSH).[24,35,40] It is generally believed that the protective effect of GSH against the oxidative breakdown of lipids is mediated through GPx by the reduction of endogenously formed hydroperoxides of unsaturated fatty acids to hydroxyl derivatives.[8]

In the present study there is no uniform trend in the values of ACR on comparison with triglyceride levels in Type 2 DM with microalbuminuria (r = 0.136, p value = 0.3471). Some studies have supported these findings.[13] Total cholesterol and LDL cholesterol were significantly correlated with ACR independently. Others have found significant correlation between total cholesterol, LDL cholesterol and progression of nephropathy in type 2 DM patients.[41] Agarwaal et al found significant correlation between nephropathy and HDL-c.[41] In the present study decreasing trend in HDL-c values was seen with increase in ACR in patients of Type 2 DM. Our result may not be statistically compatible with these authors because of the fact that the number of patients enrolled in their study was large (n=323) as compared to our study (n=50). Atherogenic lipoproteins can infiltrate into the glomerular endothelium and mesangial cells, initiating a cascade of events similar to atherosclerosis.[42] ApoB was found to be associated with a declining glomerular filtration rate in patients with chronic renal disease and renal dyslipidemia was predominantly associated with the accumulation of ApoB-containing lipoproteinsin in both sclerotic and nonsclerotic glomeruli. [43,44] The observation of the study in Taiwanese type 2 diabetic patients suggested that ApoB-containing lipoproteins could also initiate early glomerular injury leading to incipient diabetic nephropathy with micro-albuminuria.[45]

HbA1c% was correlated independently with MDA, significant positive correlation was observed .Our findings are supported by observations of Vincintini et al. [28]

GPx showed a negative trend which was not significant on correlation with HbA1C% Ramakrishna et al had observed inverse correlation between these two parameters but their findings were in patients of type 1 diabetes.[46]

Between MDA & GPx a negative trend was observed which suggests permanent structural membrane alterations in diabetes as well as increased production of ROS and decreased antioxidants in the circulation.[47]

CONCLUSION

HbA1c%, FSG, parameters of lipid profile were significantly correlated with urine albumin creatinine ratio; hence it may be concluded that higher levels of these biochemical parameters can be potential risk factors for development of micro-albuminuria in Type 2 DM patients. Malondialdehyde levels may be suggested as additional parameter for biochemical evaluation to evaluate kidney damage in Type 2 DM patients as in our study the Plasma Malondialdehyde levels are not only high but the levels are significantly correlated with microalbuminuria. Glutathione peroxidase did not show any significant correlation with ACR as well as MDA independently which may be an indication that its role in diabetic incipient nephropathy is not as pronounced as expected.

Conflict of interest: Authors declare that there is no conflict of interest related to this work.

REFERENCES

1. Wild S, Roglic G, Green A et al. Global prevalence of Diabetes: Estimates for the year 2000 and projections for 2030. Diabetes Care 2004;27:1047-1053
2. Unnikrishnan RI, Rema M, Pradeepa R et al. Prevalence and risk factors of diabetic nephropathy in an urban south Indian population: The Chennai Urban Rural Epidemiology Study (CURES 45). Diabetes Care. 2007;30(8):2019-24
3. American Diabetes Association (ADA): Nephropathy in Diabetes (Position Statement): Diabetes Care. 2004; 27:578-583.
4. Adler AL, Steven RJ, Manley SE et al. Development and progression of nephropathy in Type 2 Diabetes. The United Kingdom Prospective Study (UKPDS); Kidney International. 2003;(63):225-232
5. Halliwell B & Gutteridge JMC. Free Radicals in Biology and Medicine. Oxford University Press. 1997
6. Sunde RA. Selenium. In: Handbook of Nutritionally Essential Mineral Elements, Vol. 18, p. 493–556, O'Dell B.L. And Sunde R. A. (Eds), Marcel Dekker, New York
7. Avissar N, Ornt DB, Yagil Y et al. Human kidney proximal tubules are the main source of plasma glutathione peroxidase. Am. J.Phys. 1994;266:C367–C375
8. Fujiwara Y, Kondo T, Murakami K et al. Decrease of the inhibition of lipid peroxidation by glutathione-dependent system in erythrocytes of non-insulin dependent diabetics. Klin Wochenschr 1989;15(67):336–41
9. Pagelia DE, Valentine WN. Studies on quantitative and qualitative characterization of erythrocytic glutathione peroxidase. J.Lab Clin Med 1967;70(1):158-169
10. Piarulli F, Sartore G, Ceriello A et al.Relationship between Glyco-oxidation, antioxidant status and microalbuminuria in Type 2 Diabetic patients. Diabetologia. 2009;52(7):1419-25
11. Farvid MS, Djalali M, Siassi F et al. Association of glomerular and tubular dysfunction with glycaemic control, lipid, lipoprotein, apolipoprotein and antioxidant status in Type 2 Diabetes Mellitus. Singapore Med J. 2007;48(9):840-46
12. Kavitha G, Ramani G, Dhass Priya K et al. Oxidative Stress, Interleukin (Il-6) and atherogenic index of plasma in diabetic nephropathy. International Journal of Applied Biology and Pharmaceutical Technology. 2011;2(2): 211-17
13. Varghese A, Deepa R, Rema M et al. Prevalence of microalbuminuria in Type 2 Diabetes Mellitus at a diabetes centre in Southern India. Postgrad Med J. 2001;77(908):399-402
14. Aryal M, Jha B. Assessment of proteinuria as a marker of nephropathy in Type 2 Diabetes Mellitus. Nepal Med Coll J. 2006;8(4):250-53
15. Vaziri ND, Sato T, Liang K. Molecular mechanism of altered cholesterol metabolism in focal glomerulosclerosis. Kidney International 2003;63:1756–63
16. Pasupathi P, Farook J, Chinnaswamy P. Oxidant-Antioxidant status, high sensitive C-reactive protein and Homo Cysteine levels in Type 2 Diabetic Patients with and without microalbuminuria. Int J Biol Med Res. 2010;1(3); 04-08
17. Ozdemir G, Ozden M, Maral H et al. Malondialdehyde, Glutathione, Glutathione Peroxidase And Homocysteine Levels In Type 2 Diabetic Patients With And Without Microalbuminuria. Ann Clin Biochem. 2005;42(2):99-104
18. Neri S, Bruno CM, Raciti C et al. Alteration of oxide reductive and haemostatic factors in Type 2 Diabetics. J. Intern. Med. 1994;336: 495-500
19. Aslan M, Sabuncu T, Kocyigit A et al. Relationship between total oxidant status and severity of diabetic

nephropathy in Type 2 Diabetic patients. Nutr Metab Cardiovasc Dis. 2007; 17(10):734-40.
20. Ozben T, Nacitarhan S, Tuncer N. Plasma and urine malondialdehyde levels in non-insulin dependent diabetic patients with and without microalbuminuria.. Int J Clin Lab Res. 1995;25(3):162-64
21. Brownlee M. The pathophysiology of diabetic complications: A unifying mechanism. Diabetes. 2005;54:1615–1625
22. Nishikawa T, Edelstein D, Du XL et al. Normalizing mitochondrial superoxide production blocks three pathways of hyperglycaemic damage. Nature. 2000;404:787–790
23. Lee TS, McGregor LC, Fluharty SJ et al. Differential regulation of protein kinase C and (Na, K)-adenosine triphosphatase activities by elevated glucose levels in retinal capillary endothelial cells. J Clin Invest. 1989;83:90–94.
24. Evans JL, Goldfine ID, Maddux BA et al. Oxidative stress and stress activated signaling pathways: A unifying hypothesis of Type 2 diabetes. Endocr Rev 2002;23:599–622
25. Kusuvulu MM, Giri R, Rao KB et al. Lipid peroxidation and antioxidant enzyme levels in type 2 diabetics with microvascular complications. Diabetes & Metabolism. 2000;26:387-392
26. Blum J, Fridovich I. Inactivation of Glutathione peroxidase by superoxide radical. Arch. Biochem Biophys. 1985;240:500-508
27. Hodgkinson AD, Bartlett T, Oates PJ et al. The response of antioxidant genes to hyperglycemia is abnormal in patients with Type 1 Diabetes and diabetic nephropathy. Diabetes. 2003;52:846-51
28. Vincintini JMS, Valentini JMS, Grotto DMS et al. Association among microalbuminuria and oxidative stress biomarkers in patients with Type 2 diabetes. Journal of Investigative Medicine: 2011;59:649-654
29. Gall MA, Hougaard P, K Borch-Johnsen et al. Risk factors for development of incipient and overt diabetic nephropathy in patients with non-insulin dependent Diabetes Mellitus: Prospective, Observational Study. BMJ. 1997;314(7083):783-8
30. Park JY, Kim HK, Chung YE et al. Incidence and determinants of microalbuminuria in Koreans with Type 2 Diabetes. Diabetes Care. 1998;21(4):530-4
31. Mordchai R, David B, Dorit RS et al. Main risk factors for nephropathy in Type 2 Diabetes Mellitus are plasma cholesterol levels, mean blood pressure, and hyperglycemia. Arch Intern Med. 1998;158:998-1004
32. Amini M, Safaei H, Aminorroaya A. The incidence of microalbuminuria and its associated risk factors in Type 2 Diabetic patients in Isfahan, Iran. Rev Diabet Stud. 2007;4(4):242-248
33. Kassab A, Ajmi T, Issaoui M et al. Homocysteine Enhances LDL Fatty Acid Peroxidation, Promoting Microalbuminuria In Type 2 Diabetes. Ann Clin Biochem. 2008;45:476–80
34. Sheikh SA, Baig JA, Iqbal T et al. Prevalence of microalbuminuria with relation to glycemic control in Type-2 Diabetic patients in Karachi. J Ayub Med Coll Abbottabad. 2009;21(3):83-86
35. Maxwell SR, Thomason H, Sandler D et al. Antioxidant status in patients with uncomplicated insulin-dependent and non-insulin dependent diabetes mellitus. Eur J Clin Invest. 1997;271:484–490
36. Stehouwer CDA. Von Willebrand factor, dysfunction of the vascular endothelium, and the development of renal and vascular complications in diabetes. In: Mogensen CE 3rd ed. The kidney and hypertension in diabetes mellitus. Kluwer, Boston, 1997 pp 155–163
37. Dinneen SF, Gerstein HC. The association of microalbuminuria and mortality in non-insulin-dependent

diabetes mellitus. Arch Int Med. 1997;157:1413–1418
38. Hunjoo Ha et al. Future Of Interventions In Diabetic Nephropathy: Antioxidants Peritoneal Dialysis International. 1999;19(S):228-233
39. Kronhauser C, Garcia RJR, Wrobel K et al. Serum Se and Gpx concentration in Type 2 Diabetes Mellitus patients. Prim Care Diabetes. 2008;2(2):81-85
40. Sekhar RV, McKay SV, Patel SG et al. Glutathione synthesis is diminished in patients with uncontrolled diabetes and restored by dietary supplementation with cysteine and glycine. Diabetes Care. 2011;34(1):162-7
41. Agarwal N, Sengar NS, Jain PK, Khare R. Nephropathy in newly diagnosed Type 2 Diabetics with special stress on the role of hypertension. Journal of Association of physicians of India. 2011; 59:145-47
42. Kamanna VS, Roh DD, Kirschenbaum MA. Hyperlipidemia and kidney disease:concepts derived from histopathologyand cell biology of the glomerulus. Histol Histopathol. 1998; 13:169–179.
43. Samuelsson O, Aurell M, Knight-Gibson C et al. Apolipoprotein-B containing lipoprotein and progressionof renal insufficiency. Nephron.1993; 63:279–285
44. Samuelsson O, Mulec H, Knight-Gibson C et al. Lipoprotein abnormalities are associated with increased rate of progression of human chronic renal insufficiency. Nephrol Dial Transplant 12:1908–1915,Observations
45. Tseng,Chin-Hsiao. Apolipoprotein B Is an Independent Risk Factor for Microalbuminuria in Taiwanese Patients With Type 2 Diabetes. Letters Diabetes Care;2003: 26(10): 2965
46. Ramakrishna V, Jailkhani R. Evaluation of oxidative stress in Insulin Dependent Diabetes Mellitus (IDDM) patients. Diagnostic Pathology. 2007;2(2):22-26
47. Sundaram RK, Bhaskar A, Vijayalingam S et al. Antioxidant status and lipid peroxidation in Type 2 diabetes mellitus with and without complications. Clin Sci (Lond) 1996; 90: 255–60.

How to cite this article: Chambial S, Sharma RK, Gupta BK et. al. Oxidative stress in type 2 diabetes mellitus patients with microalbuminuria. Int J Health Sci Res. 2013;3(10):29-36.

****