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Research Article

# Study of the Serum Levels of Iron, Ferritin and Magnesium in Diabetic Complications

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**ABSTRACT**

Alteration in mineral status has been found to be associated with impaired insulin release, resistance and dysglycemia. To study the mineral status in diabetic complications - thirty patients each with diabetic neuropathy, nephropathy and retinopathy along with thirty uncomplicated diabetes mellitus patients and age matched thirty healthy controls were recruited for this study. Estimation of glycemic status, magnesium, iron and ferritin levels was done. Magnesium levels were found to be significantly decreased in the microvascular complications which correlated negatively with glycated hemoglobin. Iron and ferritin levels were found to be significantly increased in diabetic neuropathy, nephropathy and retinopathy. Hypomagnesemia, increased iron levels and hyperferritinemia seem to be associated with microvascular complication of diabetes mellitus as either causative factors or as a consequence of the disease. Screening of patients for these factors might help in the management of progression of the disease.

**Key words**:

## INTRODUCTION

Long standing metabolic derangement in Diabetes mellitus is associated with permanent and irreversible functional and structural changes in the vascular system resulting in development of complications affecting the kidney, eye and nervous system[1]. Impaired insulin release, insulin resistance and glucose intolerance have been found to be associated with alteration in mineral status in diabetic patients[2]. Various mechanisms have been proposed for the onset of diabetes and the development and progression of diabetic complications one of which is alteration in the mineral status[3-5]. Magnesium and iron are very important cations needed for the optimum cellular process. Magnesium is an abundant intracellular ion playing a vital role in insulin secretion and activity. Insulin is involved in the transport of magnesium through the cellular membrane and in the intracellular supply[6]. Hypomagnesemia may enhance endothelial cell dysfunction and thrombogenesis by increased platelet aggregation and vascular calcification. Low magnesium levels also seem to result in proinflammatory and profibrinogenic response reducing the protective enzymes against oxidative stress[7-11]. Iron is a transition metal that acts as an oxidant. Increased accumulation of iron affects insulin synthesis and secretion in the pancreas[12]. Iron deposition in the liver may cause insulin resistance by interfering with the ability of insulin to suppress hepatic glucose production. Catalytic iron converts poorly reactive free radicals like $H_2O_2$ into highly reactive ones like hydroxyl radical and superoxide anion that can initiate and propagate the cascades leading to oxidative damage. Several studies have shown the role of oxidative stress in diabetic patients with iron overload[13-16]. Ferritin is an index of body iron stores and is an inflammatory marker. Body iron stores are positively associated with the development of glucose intolerance, type 2 diabetes mellitus. Studies involving the levels of magnesium, iron and ferritin in microvascular complications of diabetes mellitus have yielded controversial results. In this study, we evaluated the magnesium, iron and ferritin status in patients with diabetic neuropathy, retinopathy and nephropathy.

## MATERIAL AND METHODS

The study was conducted at SRM Medical College Hospital & Research Centre after obtaining the approval from the Institutional Ethical Committee.

Thirty patients each diagnosed with diabetic retinopathy, neuropathy and nephropathy were recruited along with 30 diabetic patients without complications. Control group comprised of thirty healthy volunteers. All the subjects were in the age group of 35 - 70 years. Subjects with ESRD (end stage renal disease), anemia, those on mineral and iron supplements, drugs like aminoglycosides, amphotericin B, Cetuximab, cyclosporine, digoxin, diuretics were excluded from the study.

After obtaining consent from the patient and eliciting the relevant history, fasting blood samples were collected for the estimation of glucose, $HbA_{1c}$, lipid profile, iron, ferritin and magnesium. Urine samples were collected for microalbumin analysis. Plasma glucose, lipid profile, iron, magnesium and urine microalbumin were estimated using standard kits in Beckman Coulter AU 400 chemistry

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Table 1: Demographic and biochemical parameters of diabetic patients with and without complications and healthy controls

| Parameters | Control | Diabetes | Neuropathy | Nephropathy | Retinopathy |
|---|---|---|---|---|---|
| FBS (mg/dL) | 83.8 ± 8.2 | 123.1 ± 22.4 * | 148.1 ± 19.9 * | 158.7 ± 27.3 * | 187.5 ± 93.8 * |
| Hb (g/dL) | 14.6 ± 1.57 | 14.8 ± 1.56 | 14.7 ± 1.49 | 14.1 ± 1.19 | 14.0 ± 1.06 |
| T.Chol (mg/dL) | 165.7 ± 10.37 | 242.3 ± 24.45 * | 242.3 ± 28.46 * | 241.9 ± 27.1 * | 244.8 ± 46.2 * |
| TGL (mg/dL) | 94.3 ± 16.7 | 164.3 ± 32.77 * | 166.0 ± 20.47 * | 182.1 ± 41.33 * | 180.1 ± 38 * |
| HDL (mg/dL) | 41.0 ± 3.41 | 54.5 ± 3.52 | 60.8 ± 7.05 | 56.9 ± 11.41 | 57.0 ± 11.1 |
| LDL (mg/dL) | 99.7 ± 15.9 | 130.3 ± 22.3 * | 145.2 ± 23.4 * | 140.4 ± 40.85 * | 140.7 ± 32.8 * |
| Microalbumin (mg/L) | 9.2 ± 4.06 | 101.3 ± 77.01 * | 116.3 ± 67.43 * | 423.1 ± 63.49 * | 129.9 ± 58.2 * |

* $p < 0.05$ is considered statistically significant

Table 2: HbA1c, Iron, Ferritin, Magnesium levels in diabetic neuropathy in comparison with diabetic and control subjects

| Parameters | Controls (n= 30) | Diabetes (n= 30) | Diabetic neuropathy (n= 30) | p value |
|---|---|---|---|---|
| HbA1c (%) | 5.0 ± 0.45 | 6.7 ± 0.11 | 7.5 ± 1.7 | < 0.001 * |
| Iron (µg/dL) | 145.3 ± 21.25 | 200.9 ± 19.9 | 225.5 ± 34.2 | < 0.001 * |
| Ferritin (ng/mL) | 192.2 ± 60.5 | 304.0 ± 41.22 | 351.9 ± 24.2 | < 0.001 * |
| Magnesium (mg/dL) | 1.9 ± 0.11 | 1.5 ± 0.11 | 1.4 ± 0.12 | < 0.01 * |

* $p < 0.05$ is considered statistically significant

Table 3: HbA1c, Iron, Ferritin, Magnesium levels in diabetic nephropathy in comparison with diabetic and control subjects

| Parameters | Controls (n= 30) | Diabetes (n= 30) | Diabetic nephropathy (n= 30) | p value |
|---|---|---|---|---|
| HbA1c (%) | 5.0 ± 0.45 | 6.7 ± 0.11 | 7.4 ± 0.74 | < 0.001 * |
| Iron (µg/dL) | 145.3 ± 21.25 | 200.9 ± 19.9 | 261.9 ± 27.5 | < 0.001 * |
| Ferritin (ng/mL) | 192.2 ± 60.5 | 304.0 ± 41.22 | 357.5 ± 28.5 | < 0.001 * |
| Magnesium (mg/dL) | 1.9 ± 0.11 | 1.5 ± 0.11 | 1.4 ± 0.1 | < 0.001 * |

* $p < 0.05$ is considered statistically significant

analyzer. Ferritin levels were estimated by immunoenzymatic assay using TOSOH AIA immunoanalyzer. All data are expressed as mean ± SD. Statistical analysis was done using SPSS software. Statistical significance for the diabetic complications and controls were analyzed by using ANOVA and Pearson's correlation and $p < 0.05$ was considered as statistically significant.

## RESULTS AND DISCUSSION

Our study shows that serum magnesium levels are decreased in micro vascular complications of Diabetes mellitus - Retinopathy, Nephropathy and Neuropathy. Similar results were observed with inverse correlation with glycemic control and a strong association with diabetic retinopathy[17-20]. The various causes of low magnesium in diabetes include diets low in magnesium, osmotic diuresis causing high renal excretion of magnesium, insensitivity to insulin affecting the intracellular magnesium transport causing loss, rampant use of diuretics promoting magnesium wasting and reduced tubular reabsorption due to insulin resistance[21-25]. There are reports of higher incidence of retinopathy in diabetics with hypomagnesemia[26,27]. In our study, diabetic retinopathy patients had lower magnesium values compared to diabetic patients without any complications. Hypomagnesemia is considered as a possible risk factor for the development and progression of diabetic retinopathy[28]. Diabetic nephropathy patients had significant hypomagnesemia in our study. Sakaguchi et al observed that hypomagnesemia was significantly associated with progression of ESRD in patients with diabetic nephropathy but not in those with non-diabetic CKD[29]. Serum magnesium levels were significantly lower in diabetic neuropathy patients when compared to diabetic patients without complications. Low intracellular levels were reported in diabetic peripheral neuropathy subjects with improvement in nerve conduction following supplementation[30] suggesting that magnesium depletion might be linked to the pathogenesis of this complication. A negative correlation was observed between serum magnesium levels and glycated hemoglobin in all the three studied microvascular complications emphasizing its role in the progression of the disease. Our study demonstrated an increase in iron and ferritin levels in diabetic micro vascular complications. Ferritin is the storage form of iron and it releases iron in a controlled fashion whihc plays a vital role in the maintenance of intracellular iron balance. Iron is a potent pro-oxidant and reactive oxygen species have been shown to interfere with insulin signaling at the cellular level[31]. Shi zhao et al used PC-12 cells as invitro cellular models of peripheral neuropathy and demonstrated that iron overload aggravates oxidative stress injury in neural cells under high glucose concentration and that the Nrf2/ARE signaling pathway might play an important role in pathogenesis of neuropathy. Extensive glycation of basement membrane/internal elastic lamina proteins generates adduct capable of binding adventitious iron. The resultant

Table 4: HbA1c, Iron, Ferritin, Magnesium levels in diabetic retinopathy in comparison with diabetic and control subjects

| Parameters | Controls (n= 30) | Diabetes (n= 30) | Diabetic retinopathy (n= 30) | p value |
|---|---|---|---|---|
| HbA1c (%) | 5.0 ± 0.45 | 6.7 ± 0.11 | 8.1 ± 1.0 | < 0.001 * |
| Iron (µg/dL) | 145.3 ± 21.25 | 200.9 ± 19.9 | 260.2 ± 23.3 | < 0.001 * |
| Ferritin (ng/mL) | 192.2 ± 60.5 | 304.0 ± 41.22 | 356.7 ± 28.0 | < 0.001 * |
| Magnesium (mg/dL) | 1.9 ± 0.11 | 1.5 ± 0.11 | 1.4 ± 0.1 | < 0.001 * |

* $p < 0.05$ is considered statistically significant

Table 5: Correlation between Magnesium levels and HbA1c in diabetic retinopathy, diabetic neuropathy and nephropathy

| | Parameters | Correlation coefficient | p value |
|---|---|---|---|
| Diabetic neuropathy | Mg vs. HbA1c | - 0.421 | < 0.01 |
| Diabetic nephropathy | Mg vs. HbA1c | - 0.404 | < 0.01 |
| Diabetic retinopathy | Mg vs. HbA1c | - 0.480 | < 0.01 |

subendothelial glycochelates catalytically destroy endothelium derived nitric oxide leading to imbalance of vasodilator vs vasoconstrictor factors resulting in a relative state of chronic vasoconstriction. The diminished blood flow to peripheral nerves leads to neuronal hypoxia and eventual nerve death[32]. Oxidative stress factors such as hyperglycemia, advanced glycation end products and hyperlipidemia contribute to the availability of intracellular iron that can generate and viciously worsen oxidative stress and renal damage. Iron content in the kidney has been shown to be increased in animal models of diabetes. There is considerable evidence that one renal insufficiency develops, regardless of etiology, it tends to progress over time. The pathogenic role of iron in progression of kidney disease is indicated by the observation that progression can be prevented either by an iron-deficient diet or chelators (33,34,35). Hsu et al found that hyperferritinemia may be an independent risk factor of nephropathy in patients with type 2 diabetes[36]. In the diabetic eye, there is an impairment of iron homeostasis, leading to iron overload. The mechanisms involved in this include: (1) Destruction of heme molecules induced by hyperglycemia (2) intra retinal and vitreal hemorrhages (3) Overexpression of the renin-angiotensin system. The main consequences of iron overload are the following: (1) Retinal neurodegeneration due to the increase of oxidative stress (2) increase of AGE-RAGE binding (3) Defective phagocytosis of retinal pigment epithelium, which generates the accumulation of autoantigens and the synthesis of pro inflammatory cytokines. Iron catalyses the binding of the AGEs to the specific receptor, a crucial step in the pathogenesis of Diabetic retinopathy[37]. The observational nature of our study design precludes proving a caustive relationship between the altered mineral status and the diabetic microvascular complications. The cause of deficiency of magnesium is not established as quantification was not done for magnesium in dietary sources nor was urinary magnesium measured in this study. Serum albumin level was not measured to reflect accurate level for serum magnesium. Therefore, further studies are needed to address these issues. Hypomagnesemia, increased iron levels and hyperferritinemia seem to be associated with microvascular complications of Diabetes mellitus. Routine screening of these elements might help in mitigating the progression of diabetes mellitus towards complications by either magnesium supplementation or iron chelation therapy.

## REFERENCES


1. 1999 United states Renal Data System Annual Report: National Technical Information Service. US Department of Health and Human Services, Springfield, VA.
2. Paolisso G, Sgambato S, Pizza G, Passariello N, Varricchio M, D'Onofrio F. Improved insulin response and action by chronic magnesium administration in aged NIDDM subjects. Diabetes Care 1989, 12:265-69.
3. Wilson JG, Lindquist JH, Grambow SC, Crook Ed, Maher JF. Potential role of increased iron stores in diabetes. Am J Med Sci. 2003;325: 332-339.
4. Kahn SE, Hull RL, Utzschneider KM. Mechanism linking obesity to insulin resistance and type 2 diabetes. Nature 2006; 444: 840-6.
5. Dongiovanni P, Valenti L, Ludovica Francanzani A, Gatti S, Cairo G, Fargion S. Iron depletion by desferroxamine upregulates glucose uptake and insulin signaling in hepatoma cells and in rat liver. Am J Pathol 2008; 172:738-47.
6. Valk HW. Magnesium in diabetes mellitus. Nethelands J Med 1999; 54: 139-46.
7. Rayssignier Y. Role of magnesium and potassium in the pathogenesis of arteriosclerosis. Magnesium 1984; 3: 226-238.
8. Shivakumar K. Pro-fibrinogenic effects of magnesium deficiency in the cardiovascular system. Magnes Res. 2002; 15: 307-315.
9. Maier J, Malpuech-Brugere C, Zimowska W, Rayssignier Y, Mazur A. Low magnesium promotes endothelial cell dysfunction: implications for atherosclerosis, inflammation and thrombosis. Biochim Biophys Acta 2004; 1689: 13-21.
10. Zhou Q, Olinescu RM, Kummerow FA. Influence of low magnesium concentrations in the medium on the antioxidant system in cultured human arterial endothelial cells. Magnes Res 1999; 12: 19-29.

11. Hartwig A. Role of magnesium in genomic stability. Magnes Res 2001; 475: 113-121.
12. Rahier JR, Loozen S, Goebbels RM. The hemochromatotic human pancreas: a quantitative immuno-histochemical and ultrastructural study. Diabetologia 1987; 30: 5-12.
13. Lassman MN, Genel M, Wise JK, Hendler R, Felig P. Carbohydrate homeostasis and pancreatic islet cell function in thalassemia. Ann Intern Med.1974; 80: 65-67.
14. Fernandez-Real JM, Lopez-Bermejo A, Ricart W. Cross-talk between iron metabolism and diabetes. Diabetes 2002; 51: 2348-2354.
15. Niederau C, Berger M, Stremmel W, Starke A, Strohmeyer G, Ebert R, Siegel E, Cruetzfeldt W. Hyperinsulinemia in non-cirrhotic hemochromatosis: impaired hepatic insulin degradation? Diabetologia 1984; 6: 441-444.
16. Walter RM, Uriu-Hare JY, Olin KL, Oster MH, Anawalt BD, Critchfield JW. Copper, zinc, manganese and magnesium status and complications of diabetes mellitus. Diabetes Care 1991; 14: 1050-56.
17. Diwan AG, Pradhan AB, Lingojwar D, Krishna KK, Singh P, Almelkar SI. Serum zinc, chromium and magnesium levels in type 2 diabetes. Int J Diab Dev Countries 2006; 26: 122-23.
18. Tripathy S, Sumathi S, Raj GB. Minerals and nutritional status of type 2 diabetic subjects. Int J Diab Dev Countries 2004; 24: 27-28.
19. Sharma A, Dabla S, Agarwal RP, Barjatya H, Kothari RP, Kochar DK. Serum magnesium: an early predictor of course complications of diabetes mellitus. J Indian Med Assoc 2007; 105:16-20.
20. Kin DJ, Xun P, Liu K, Loria C, Yokota K, Jacobs DR. Magnesium intake in relation to systemic inflammation, insulin resistance and the incidence of Diabetes. Diabetes Care 2010; 33: 2604-10.
21. Schultze MB, Schultz M, Heidemann C, Schienkiewitz A, Hoffman K, Boeing H. Fiber and magnesium intake and incidence of type 2 diabetes: A prospective study and meta-analysis. Arch Intern Med 2007; 167: 956-965.
22. Paolisso G, Sgambato S, Passariello N, Guigliano D, Scheen A, D'Onofrio F. Insulin induces opposite changes in plasma and erythrocyte magnesium concentrations in normal man. Diabetologia 1986; 29: 644-47.
23. Pham C, Pham PM, Pham SV, Miller JM. Pham PT. Hypomagnesemia in patients with type 2 diabetes. Clin J Am Soc Nephrol. 2007; 2: 366-73.
24. Limaye CS, Londhey VA, Nadkar MY, Borges NE. Hypomagnesemia in critically ill medical patients. J Assoc Physicians India 2011; 59: 19-22.
25. Dasgupta A, Saikia UK, Sharma D, Dutta Chowdhury S. Quadriparesis in diabetes due to dyselectrolytemia. Indian J Endorinol Metab 2010; 14:27-29.
26. Fujii S, Takemura T, Wada M, Akai T, Okuda K. Magnesium levels in plasma erythrocytes and urine in patients with diabetes mellitus. Horm Metab Res 1982; 14: 61-62.
27. Nazar Haddad S, Salah Zuhair. Serum magnesium and severity of diabetic retinopathy. Mjbu 2010; Vol 28: 1-4.
28. Yusuke Sakaguchi, Tatsuya Shoji, Terumasa Hayashi. Hypomagnesemia in type 2 diabetic nephropathy: a novel predictor of end-stage renal disease. Diabetes Care 2012; 35(7):1591-7.
29. De leeuw, Engelen W, De Block, Van Gaal L. Long term magnesium supplementation influences favourably the natural evolution of neuropathy in Mg-depleted type 1 diabetic patients. Magnes Res 2004; 17(2);109-14.
30. Hussain MAM, Varghaz Z, Polities D. Insulin resistance and iron overload. Ann Clin Biochem.1983; 20(2):77-79.
31. Mingwei Qian, Ulf T Brunk, Galen M Pieper, John W Eaton. Diabetic peripheral neuropathy: possible involvement of iron bound to glycated basement membrane proteins. Pediatric Research 1998;43: 83-84.
32. Johnson WT, Evans GW. Effects of the interrelationship between dietary protein and minerals on tissue content of trace metals in streptozotocin-diabetic rats. J Nutrit 1984; 114:180-90.
33. Nankivell BJ, Chen J, Boadle Ra, Harris DCH. The role of tubular iron accumulation in the remnant kidney. J Am Soc Nephrol 1994; 4:1598-1607.
34. Nankivell BJ, Boadle Ra, Harris DCH. Iron accumulation in human chronic renal disease. Am J Kid Dis 1992; 20:580-584.
35. Y H Ysu, Huang MC, Chang HY, Shin SJ, Wahlqvist ML, Chang YL et al. Association between serum ferritin and microalbuminuria in type 2 diabetes in Taiwan. Diabetic Medicine 2013; 30(11):1367-73.
36. Andreea Ciudin, Hernandez C, Simo R. Iron overload in Diabetic Retinopathy: A Cause or a Consequence of Impaired Mechanism? Experimental Diabetes Research 2010, Article ID 714108, 8 pages, 2010. doi10.1155/2010/714108.
37. Yamagishi SI, Ueda S, Mastui T, Nakamura, Sokuda. Role of advanced glycation end products (AGEs) and oxidative stress in Diabetic Retinopathy. Current Pharmaceutical Design 2008;14(10):962-968.