Original Research Paper Biochemistry

EVALUATION OF ASSOCIATION OF SERUM MAGNESIUM AND HS-CRP IN TYPE 2 DIABETES MELLITUS

Dr. Premjeet Kaur MD Biochemistry, Demonstrator, Department of Biochemistry, G. M. C. Chandigarh. -Corresponding author

ABSTRACT

Type 2 diabetes mellitus is now recognized as an inflammatory condition associated with insulin resistance and abnormal endothelial vascular reactivity. High sensitive C-reactive protein (hs-CRP) is an established marker of low grade systemic inflammation.Low magnesium level has been linked to reduced insulin sensitivity and increased risk of type 2 diabetes mellitus. Low magnesium has a direct role in promoting endothelial dysfunction by generating a pro-inflammatory, prothrombotic and proatherogenic environment. Hypomagnesaemia has been implicated in adversely affecting diabetic complications. Because serum Magnesium and hs-CRP reflect closely related component of the same disease process, a strong relationship between these variables may be anticipated. We selected 50 patients of Type-2 diabetes mellitus and 50 normal healthy individuals to evaluate the association between serum magnesium and hs-CRP. Statistical analysis depicted a non-significant (p> 0.05) negative correlation (r=-0.20) between serum magnesium and hs-CRP in type 2 diabetic patients. This data supports the hypothesis that hypomagnesemia and hs-CRP may be involved independently in the pathogenesis of diabetes and its complications.

KEYWORDS

INTRODUCTION: Type 2 diabetes mellitus is now recognized as an inflammatory condition associated with insulin resistance and abnormal endothelial vascular reactivity. [1]. Independent of the triggering agent and of the initial events, any process linked to chronic inflammation which decrease insulin action and insulin resistance will lead to worsening of inflammation in a vicious cycle. One of the most sensitive acute-phase reactants in humans is hs-CRP.[2][3]

There is accumulating evidence that the altered metabolism of some micronutrients in diabetes mellitus might have a specific role in the pathogenesis and complications of the disease. Magnesium deficiency is the most evident disturbance of metal metabolism in insulin-dependent diabetes mellitus. Mg depletion negatively affects the glucose homeostasis and insulin sensitivity in diabetic patients [4] as well as the complications such as retinopathy, thrombosis and hypertension. [5] Clinically, hypomagnesaemia is defined as a serum Mg concentration less than or equal 1.6 mg/dl or >2 SD below the mean of the general population. [6][7] Preventing hypomagnesaemia may therefore be beneficial in the management of the diabetics. [8] The Recommended Daily Allowance (RDA) for magnesium is 6 mg/kg/day Jerry L.et al 2000). Low Magnesium levels may promote endothelial cell dysfunction and thrombogenesis via increased platelet aggregation and vascular calcification. [9]

Because hs-CRP and Mg seems to be closely related component of the same disease process, a strong relationship between these variables may be anticipated.

MATERIALS AND METHODS: The present study was a case control prospective study undertaken in the Department of Biochemistry in collaboration with Department of Medicine, Sri Guru Ram Das Institute of Medical Sciences and Research, Amritsar. A total of 100 subjects willing to participate in the study with informed consent were included in the study. 50 patients of poorly controlled Type 2 Non Insulin Dependent Diabetes Mellitus (NIDDM) between 40-65 yrs of age, of either sex whose HbA1c was >7% and 50 healthy, age and sex matched controls from the same population but without any disease and without family history of DM.

Exclusion criteria: Patients suffering from type-1 DM, patients with acute complications of DM like Diabetic ketoacidosis, history of acute infections, other ailments like gross congestive heart failure, tuberculosis, gout, rheumatoid arthritis and skeletal muscle injury, serum creatinine > 1.5mg/dl, renal failure and those giving positive dip stick test for proteinuria were not included in the study.

The patients and controls were screened for fasting blood sugar (FBS), lipid profile, serum hs-CRP and magnesium and the values were compared with that of normal healthy subjects. Hs-CRP was estimated by Quantia –CRP (M A Mendall et al 1996) [10], a turbidimetric immunoassay. Magnesium was estimated by Calmagite method. [11] Magnesium combines with Calmagite in an alkaline medium to form a red colored complex the intensity of which is directly proportional to the amount of magnesium present in the sample. FBS was estimated by GOD-POD Method (Trinder 1969)[12]. HbA1C by Nycocard Reader (Jeppson 2002) [13] Total Serum Cholesterol was be estimated by CHOD-PAP Method (Allain C.C. et al 1974) [14] Serum Triglyceride was be estimated by GPO-Trinder Method. (McGowan MW et al 1983) [15] Serum High Density Cholesterol (HDL-C) was estimated by Phosphotungstic Acid Method (Gordon T. Et al 1977) [16]

Result: There was no significant effect of age (p>0.05) and sex distribution (p > 0.05) in the study. Table 1 shows that FBS, HbA1c, total cholesterol, triglyceride and hs-CRP levels were significantly high in cases when compared to controls while Mg and HDL-C were significantly low in cases as compared to the controls.

Table 1 : Comparison of various parameters estimated in patients and controls

	Cases (Mean±SD)	Controls (Mean±SD)	p value
Fasting blood sugar (mg/dl)	194.38 ± 53.60	100.30 ± 12.46	< 0.001*
HbA1c (%)	8.758 ± 1.83	5.148 ± 0.51	< 0.001*
Total cholesterol (mg/dl)	223.20 ± 45.41	174.46 ± 33.90	< 0.001*
Triglycerides(mg/dl)	224.70 ± 76.77	161.14 ± 32.42	< 0.001*
HDL-C(mg/dl)	40.48 ± 8.18	55.00 ± 12.04	< 0.001*
hs-CRP (mg/dl)	1.29 ± 1.79	0.57 ± 0.09	< 0.005**
Magnesium (mEq/L)	1.09 ± 0.29	2.09 ± 0.29	<0.001*

*P<.001 =highly significant
**p<0.05-significant
***p>0.05- non significant

Table 2 shows that FBS and HbA1c were significantly correlated with hs-CRP and Magnesium in type 2 diabetics. Total cholesterol, triglycerides and HDL-C had no significant correlation with hs-CRP

but a significant correlation with MG in type 2 diabetic patients.

Table 2: Correlation of hs-CRP and Magnesium in type 2 Diabetes Mellitus with FBS, HbA1c and lipid profile parameters

	hs –CRP (mg/dl)		Magnesium (mEq/L)	
	R	P	R	P
Fasting blood sugar(mg/dl)	0.404	0.004**	-0.541	<0.001*
HbA1c (%)	0.432	0.002**	-0.320	<0.001*
Total cholesterol (mg/dl)	0.017	0.906***	-1.89	0.01**
Triglycerides(mg/dl)	0.151	0.294***	- 0.011	<0.001*
HDL-C(mg/dl)	-0.051	0.724***	0.176	0.033**

*P<.001 =highly significant
**p<0.05-significant
***p>0.05- non significant

Table 3 shows a highly significant positive (r=0.506; p <0.001) correlation between hs-CRP and Mg

Table 3: Correlation between microalbumin in urine and serum hs-CRP in patients of type 2 diabetes mellitus (cases)

Parameter	Mean ± SD	R	P
Magnesium (mEq/L)	1.09 ± 0.29	-0.20	0.163 ***
hs-CRP (mg/dl)	1.29 ± 1.79		

***p>0.05- non significant

Discussion: In present study significantly high levels of hs-CRP (p=0.004) were observed in T2DM than controls (Table 1). A similar observation was noted by other studies. [17] [18] [19] The biological mechanisms through which hs-CRP increases risk of type 2 diabetes is not well understood. Hs-CRP may have indirect influence on insulin resistance and insulin secretion through altered innate immune response. [20] A highly significant positive correlation was found between hs-CRP and FBS as well as HbA1c (Table 2) supported by the other studies. [17] Observational studies have also suggested an independent role of hs-CRP in development of insulin resistance and diabetes, but it is unclear whether the association is a causal one or the consequence of inefficacy of the adipose tissue and other confounding factors. [22] In our study, there existed non significant correlation between hs-CRP and total cholesterol, triglyceride and HDL (Table 2) which was supported by Chen Chung Fu et al [21] and D A Muttur et al [22] No significant correlation between hs-CRP and lipids profile parameter may suggest that hs-CRP act as an independent marker for cardiovascular risk factor.

In our study, a highly significant difference in levels of magnesium were found between patients and controls (p<0.001). Similar observations were found in other studies. [23] [24] [25]. Various clinical studies demonstrated that diabetics with hypomagnesemia showed reduced pancreatic β-cell activity. Insulin receptor autophosphorylation is dependent on intracellular Mg^{2+} concentrations, making Mg^{2+} a direct factor in the development of insulin resistance. Conversely, insulin is an important regulator of Mg^{2+} homeostasis. In the kidney, insulin activates the renal Mg^{2+} channel melastatin type 6 that determines the final urinary Mg^{2+} excretion. Consequently, patients with T2DM and hypomagnesemia enter a vicious circle in which hypomagnesemia causes insulin resistance and insulin resistance reduces serum Mg^{2+} concentrations. [26] [27] [28] Patients with hypomagnesemia show a more rapid disease progression and have an increased risk for diabetes complications.[29] Preventing hypomagnesemia may therefore be considered in the management of the disease. [30] [31] In our study we found a highly significant positive correlation between FBS, HbA1c and Mg. These findings were supported by Sharma A et al (2007) [32], R D Ankush et al (2009) [5] showed a strong positive correlation (p <0.001). Mg depletion has a negative impact on glucose homeostasis and insulin sensitivity in patients with type 2 diabetes as well as on the evolution of complications such as retinopathy, thrombosis and hypertension. [3] Insulin secretion requires magnesium: magnesium deficiency results in impaired insulin secretion. It has been suggested that hypomagnesemia may induce altered cellular glucose transport, reduced pancreatic insulin secretion, defective post receptor insulin signaling, and/or altered insulin–insulin receptor interactions. [33] In our study, there existed non significant correlation between magnesium and total cholesterol, triglyceride and HDL (Table II) which was supported by Hamid Nasri et al (2008) [34] No significant correlation between magnesium and lipids profile parameter may suggest that magnesium act as an independent marker for cardiovascular risk factor.

Very few studies have been conducted on the analysis of the correlation between serum Magnesium and hs-CRP in the type 2 diabetic patients. In our study, on statistical analysis there was non-significant (p> 0.05) negative correlation (r=-0.20) between serum magnesium and hs-CRP. This data supports the hypothesis that hypomagnesemia and hs-CRP may be involved independently in the pathogenesis of diabetes and its complications which might require further evaluation. One drawback in our study can be small sample size. A study by G Romero et al (2011) [35] showed that severe hypomagnesemia (Odds Ratio (OR): 8.1 and Confidence Interval (CI) 95%: 3.6-19.4 and OR: 3.7; CI 95%: 1.1-12.1) was strongly associated with elevated hs-CRP in patients of metabolic syndrome.

References

1. 1Crook M. Type 2 diabetes mellitus: a disease of the innate immune system? An update. Diabet Med 2004. 21: 203–207.
2. 2José Manuel Fernández-Real and Wifredo Ricart. Insulin Resistance and Chronic Cardiovascular Inflammatory Syndrome Endocrine Reviews 2003. 24(3):278-301.
3. 3A Rizvi. The role of inflammation in diabetes and its complications. Southern Medical Journal 2006. 99(1):8-9.
4. 4Chetan P. Hans, R. Sialy and Devi D. Bansal. Magnesium deficiency and diabetes mellitus 2002. Current science. 83:1456-1463.
5. 5R.D. Ankush, A N Suryakar and NR Ankush. Hypomagnesemia in type 2 diabetes mellitus patients: A study on the status of oxidative and nitrosative stress. IJCB 2009. 24(2):184-189.
6. 6Pham PC, Pham PM, Pham PA, Pham SV, Pham HV, Miller JM. Lower serum magnesium levels are associated with more rapid decline of renal function in patients with diabetes mellitus type 2. Clin. Nephrol 2005. 63: 429–436.
7. 7Voets T, Nilius B, Hoefs S, Kemp AW, Droogmans G, Bindels RJ, Hoenderop JG. TRPM6 forms the Mg2+ influx channel involved in intestinal and renal Mg2+ absorption. J Biol Chem2004. 279:19–25.
8. 8Monika K. Waltia, Michael B. Zimmermanna, Giatgen A. Spinasb, Richard F. Hurrella. Low plasma magnesium in type 2 diabetes. Swiss med wkly 2003. 133:289–292
9. 9Maier JAM, Malpuech Brugere C, Zunowska W, Rays-siguier Y, Mazur A. Low Mg promotes endothelial cell dysfunction, Implications for Atherosclerosis inflammatory and thrombosis. Biochem Biophys Acta 2004. 1689; 13-21.
10. 10Ma Mendall, Praful Patel et al. BMJ 1996. 312:1061-1065.
11. 11Grindler E. et al. Calmagite method. Clin Chem 1971. 17:662.
12. 12Trinder, P.Ann. GOD-POD method. Clin BioChem 1969. 6:24.
13. 13Jeppson JO et.al., Approved IFCC Reference Method for the Measurement of HbA1C in human Blood. Clin Chem Lab Method 2002. 40(1):78-89.
14. 14Allian C. C. , Poon L, S. , Chan C. S. G. , Richmond W. and Fu P. , Clin. Chem 1974. 20:47.
15. 15McGowan MW et al. Clin Chem 1983. 6:24-27.
16. 16Gordon T. et al. Am. J. Med.1977. 62:707.
17. 17Safiullah Amanullah, Abdulla Jarari at al. Association of hs-CRP with Diabetic and Non-diabetic individuals. Jordan Journal of Biological Sciences 2010. 3:7-12.
18. 18Fen-qin Chen, Jiao Wang, Xiao-bo Liu et al. Levels of Inflammatory Cytokines in Type 2 Diabetes Patients with Different Urinary Albumin Excretion Rates and Their Correlation with Clinical Variables. Journal of Diabetes Research. Volume 2013, Article ID 138969, 6 pages
19. 19Zhen Wang at al. Analysis of Inflammatory Mediators in Prediabetes and Newly Diagnosed Type 2 Diabetes Patients. Journal of Diabetes Research.Volume 2016, Article ID 7965317, 10 pages
20. 20Pradhan A, Manson J, Rifai N et al. C-reactive protein, interleukin 6 and risk of developing type 2 diabetes mellitus. JAMA 2001; 286: 327- 334.
21. 21Chen-Chung Fu, Yin-Ming Li. Association of C-reactive protein and traditional risk factors with nephropathy in the elderly patients with diabetes. Taiwan Geriatrics & Gerontology 2008. 3(3):202-212.
22. 22D A Muttur at al . Analyzing Anthropometry And Metabolic Variables Associated With Microalbumin And C-Reactive Protein As Markers Of Early Glomerular Dysfunction Among Mauritian Patients Suffering From Type II Diabetes. The Internet Journal of Laboratory Medicine 2010. 4:219-226.
23. 23Barbagallo M et al. Serum ionized magnesium in diabetic older persons. Metabolism 2014; 63(4):502-9.
24. 24Chutia H, Lynrah KG.Association of Serum Magnesium Deficiency with Insulin Resistance in Type 2 Diabetes Mellitus. J Lab Physicians 2015. Jul-Dec; 7(2):75-8.
25. 25Ramadass S, Basu S, Srinivasan AR. SERUM magnesium levels as an indicator of status of Diabetes Mellitus type 2. Diabetes Metab Syndr 2015.9(1):42-5.
26. 26Lima MdeL, Cruz T, Rodrigues LE, et al. Serum and intracellular magnesium deficiency in patients with metabolic syndrome--evidences for its relation to insulin resistance. Diabetes Res Clin Pract 2009.83:257–262.
27. 27Gunther T.. The biochemical function of Mg^{2+} in insulin secretion, insulin signal transduction and insulin resistance. Magnes Res 2010. 23(1):5-18.
28. 28Lisanne M.M. Gommers, Joost G.J. Hoenderop, René J.M. Bindels and Jeroen H.F. de Baaij. Hypomagnesemia in Type 2 Diabetes: A Vicious Circle? Diabetes Jan

2016; 65(1): 3-13.
29. Dasgupta A, Sharma D, Saikia UK. Hypomagnesemia in type 2 diabetes mellitus. Indian J Endocrinal Metab 2012. Nov;16(6) :1000-3
30. Jerry L. Nadler MD. Diabetes and Mg: The emerging role of oral Mg supplementation. Clinical Research and Laboratory News of Cardiologists. Third Quarter 2000.
31. Mooren FC, Kruger K, Volker K, Golf SW, Wadepuhl M, Kraus A. Oral magnesium supplementation reduces insulin resistance in non-diabetic subjects - a double-blind, placebo-controlled, randomized trial. Diabetes Obes Metab 2011. Mar; 13(3):281-4.
32. Sharma A, Dabla S, Agrawal RP, Barjatya H, Kochar DK, Kothari RP. Serum magnesium: an early predictor of course and complications of diabetes mellitus. J Indian Med Assoc 2007. 105(1):16-20.
33. Pham PCT, Pham PMT, Pham SV, Miller JM, Pham PTT. Hypomagnesemia in patients with type-2 diabetes. Clin J Am Soc Nephrol 2007. 2:366-373.
34. Hamid Nasri, Hamid Reza. Lipids in association with serum magnesium in diabetes mellitus patients. Bratisl Lek Listy 2008. 109:302-306.
35. Guerrero-Romero F, Bermudez-Pena C, Rodriguez-Moran M.Severe hypomagnesemia and low-grade inflammation in metabolic syndrome. Magnes Res 2011. 24(2):45-53.