e-ISSN: 0975-1556, p-ISSN:2820-2643

Available online on www.ijpcr.com

International Journal of Pharmaceutical and Clinical Research 2022; 14(7); 582-588

Original Research Article

Prospective Assessment of HS-CRP, MDA Levels in Type 2 Diabetic Patients and Also to Explore Their Association with Hba1c and Insulin Resistance

Amarendra Kumar Amar[1], Shabana[2], Deokant Deepak[3]

[1]Tutor, Department of Biochemistry, Jan Nayak Karpuri Thakur Medical College and Hospital, Madhepura, Bihar, India
[2]Tutor, Department of Biochemistry, Jan Nayak Karpuri Thakur Medical College and Hospital, Madhepura, Bihar, India
[3]Tutor, Department of Biochemistry, Jan Nayak Karpuri Thakur Medical College and Hospital, Madhepura, Bihar, India

Received: 15-05-2022 / Revised: 25-06-2022 / Accepted: 20-07-2022
Corresponding author: Dr. Amarendra Kumar Amar
Conflict of interest: Nil

Abstract

Aim: To evaluate hs-CRP, MDA levels in type 2 diabetic patients and also to explore their association with HbA1c and insulin resistance.

Methodology: A prospective evaluation study was carried out in the Department of Biochemistry, Jan Nayak Karpuri Thakur Medical College and Hospital, Madhepura, Bihar, India for 1 year period. The study population consisted of 100 subjects divided in to two groups viz., diabetic patients (type 2 diabetic subjects; n=50) and non- diabetic participants (n=50). 48 male and 52 female were including in this study. The age of the patients of both sex were 30-50 years. Biochemical analysis Fasting venous blood samples were collected from the study subjects and routine laboratory investigations were carried out by standardized protocols like serum insulin, HbA1c, hs-CRP, and malondialdehyde (MDA) and analyzed.

Results: Out of 100 patients, 48% were males and 52% were females. 37% patients belonged 30-40 years of age group, while most of the patients (63%) belonged to 40-50 years of age group. Mean age of control group was 37.9±4.5 years, while 40.2±7.1 years in type 2 diabetes mellitus patient group. Mean systolic and diastolic blood pressure in DM patients was higher as compared to control group. The mean serum hs-CRP and MDA levels were significantly high in type 2 diabetic patients compared with healthy patients. Hs-CRP and MDA levels are shown significant positive correlation with glycocylated hemoglobin (HbA1C), insulin resistance, triglycerides and negative correlation with HDL cholesterol.

Conclusion: The use of stringent blood glucose management, frequent monitoring of hs-CRP, and monitoring of MDA levels may be beneficial in the prevention of vascular problems in type 2 diabetics.

Keywords: Diabetes, hemoglobin, insulin, triglycerides

Introduction

Diabetes mellitus (DM) recognized as a common metabolic disorder worldwide caused by complex interactions of genetic, environmental and lifestyle factors. Type 2 diabetes mellitus is a serious public health concern globally and associated with lasting vascular problems, which leads to morbidity and death. Inflammation plays a critical role in the development of vascular problems in type 2 diabetes mellitus [1]. Impaired insulin production and sensitivity mechanism can be explained by oxidative stress, endoplasmic reticulum stress, amyloid deposition in the pancreas, lipotoxicity and glucotoxicity [2]. Chronic hyperglycemia and oxidative stress enhance the pro-inflammatory proteins with infiltrating macrophages secreting inflammatory cytokines which leads to systemic inflammation [3].

Blood glucose control in people with type 2 diabetes may be monitored over time using HbA1c, an essential biochemical measure [4]. When haemoglobin combines with glucose in the blood, the result is HbA1c [5]. In both healthy and diabetic individuals, HbA1c levels may be utilized as a risk factor for stroke and cardiovascular disease (CVD). The risk of cardiovascular disease (CVD) development may be reduced by 10% with a (0.2 percent) drop in HbA1c levels [6]. Several studies have shown that mothers of babies with high HbA1c levels are more likely to have cardiovascular disease (CVD) in the future [7].

When the liver produces Hs C-reactive protein in response to multiple cytokines, it is an acute phase reactant protein that serves as an indicator of low-grade systemic inflammation [8, 9]. Studies have shown that hs-CRP binds directly to oxidised LDLC and stimulates the production of plasminogen activator inhibitor-1, resulting in endothelial dysfunction and cardiovascular disease (CVD) [10-12]. Pro-inflammatory cytokines are released by infiltrating macrophages in response to hyperglycemia-induced oxidative stress, resulting in both local and systemic inflammation [13]. The synthesis of aldehydes such as malondialdehyde (MDA), propanol, hexanal, and 4-hydroxynonenal (4-HNE) has been linked to high levels of free radicals or reactive oxygen species (ROS), reactive nitrogen species (RNS). To find out if hs-CRP and MDA levels in type 2 diabetics correlate with HbA1c and insulin resistance, this research set out to do just that.

Materials and Methods:

A prospective evaluation study was carried out in the Department of Biochemistry, Jan Nayak Karpuri Thakur Medical College and Hospital, Madhepura, Bihar, India for 1 year period.

Inclusion and exclusion criteria

The study population consisted of 100 subjects divided in to two groups viz., diabetic patients (type 2 diabetic subjects; n=50) and non- diabetic participants (n=50). 48 male and 52 female were including in this study. The age of the patients of both sex were 30-50 years. We excluded the patients with positive history of smoking, alcohol, tobacco chewing, renal disease, inflammatory disorders, neoplastic disorders, thyroid disorders, liver dysfunction, and history of acute myocardial infarction, stroke and occlusive peripheral vascular disease.

Methodology

Biochemical analysis Fasting venous blood samples were collected from the study subjects and centrifuged at 3000 rpm for 15 min. Routine laboratory investigations were carried out by standardized protocols with vitros 350 fully automated analyzer. Serum insulin estimated by Enzyme Linked Immuno Sorbent Assay (ELISA), HbA1c estimated by (Ion Exchange Resin method) hs-CRP

was assessed by (latex turbidimetric immunoassay), malondialdehyde (MDA) estimated by Thiobarbituric Acid Reactive Substances (TBARS) method [15]. Post prandial venous blood samples collected for plasma glucose (PPG) analysis. Homeostasis model assessment for Insulin Resistance (HOMA-IR) HOMA- IR calculated by using fasting glucose and insulin values: HOMA – IR= fasting insulin X fasting glucose (m M/L)/22.51 [17].

Results:

Out of 100 patients, 48% were males and 52% were females. 37% patients belonged 30-40 years of age group, while most of the patients (63%) belonged to 40-50 years of age group. Mean age of control group was 37.9±4.5 years, while 40.2±7.1 years in type 2 diabetes mellitus patient group. Mean systolic and diastolic blood pressure in DM patients was higher as compared to control group.

Table 1: Gender and age distribution of patients

Variables	N=100
Gender	
Male	48
Female	52
Age	
30-40 years	37
40-50 years	63

Table 2: Comparison of baseline parameters in controls, type 2 diabetic patients

Parameters	Controls (N=50)	T2DM (N=50)	p-value
Age	37.9±4.5	40.2±7.1	0.46
Body mass index (BMI) kg/m^2	24.9±1.6	27.8±2.6	0.015
Waist/Hip ratio	1.05±0.03	0.97±0.14	0.022
Systolic BP(mmHg)	118.2±6.0	121.5±9.6	0.09
Diastolic BP (mm Hg)	76.8±6.2	81±8.0	0.15

The mean serum hs-CRP and MDA levels were significantly high in type 2 diabetic patients compared with healthy patients. hs-CRP and MDA levels are shown significant positive correlation with glycocylated hemoglobin (HbA1C), insulin resistance, triglycerides and negative correlation with HDL cholesterol.

Table 3: Comparison of FPG, PPG, HbA1C, HOMA-IR, Lipid profile, Liver profile, Renal profile hs-CRP and MDA levels in control and type 2 diabetic subjects

Parameters	Controls (N=50)	T2DM(N=50)
FPG(mg/dl)	81.4± 9.0	134.5±13.8
PPG(mg/dl)	105.8±9.2	190±24.0
HbA1C	5.5±0.8	8.8±1.2
Serum Triglycerides (mg/dl)	98.0±10.8	134.8±14.0
Serum cholesterol (mg/dl)	180.2±10.0	207.0±23.2
HOMA-IR	1.4±0.4	4.2±1.1
HDLcholesterol (mg/dl	43.7±2.2	40.0±3.4
LDLcholesterol (mg/dl)	110±11.0	137.0±14.8
Total Bilirubin(mg/dl	0.78±0.1	0.84±0.1
Direct Bilirubin(mg/dl)	0.2±0.09	0.18±0.07
Serum urea(mg/dl)	23.2±5.0	26.5±7.0
Serum creatinine(mg/dl)	0.78±0.4	0.80±0.8
hs-CRP(mg/L)	2.0±0.5	4.0±1.9
MDA(µmol/L	1.9±0.7	6.2±1.6

Discussion:

hs-CRP is a member of the class of acute phase reaction proteins and the hs-CRP level reflects the degree of systemic inflammatory response. A previous study demonstrated that the serum hs-CRP level was high in patients with DM, including those with macro vascular complications. It was also found that high hs-CRP was an independent predictor of cardiac risk in patients with T2DM [18]. Oxidative stress stimulates the inflammatory mediators which in turn enhances the production of reactive oxygen species (ROS) and reactive nitrogen species (RNS). Oxidative stress induces tumour necrosis factor alpha (TNF- α) secretion, it is linked to obesity related insulin resistance and vascular complications in type 2 diabetes mellitus [19, 20].

Meng *et al* [21] observed dynamic glucose characteristics in patients with T2DM with different concentrations of HbA1c through the CGMS for 72 h, and the results showed that the HbA1c levels were positively correlated with the 24 h MBG levels, but not with MAGE, which was consistent with the present results. In addition, the present study indicated that an increase in HbA1c levels also triggered an increase in hs-CRP levels. Sarinnapakorn *et al* [22] found that hs-CRP levels correlated with HbA1c levels in overweight female patients with T2DM. Kilpatrick *et al* [23] suggested that HbA1c was an independent predictor of the risk of retinopathy and nephropathy in patients with T1DM, and that high HbA1c levels were associated with the risk of cardiovascular mortality in the general Japanese population [24].

Chang *et al* [25] reported that there was a positive correlation between serum hs-CRP and MAGE in patients with T2DM, which was in agreement with the results of the present study. Su *et al* [26] reported that MAGE was significantly higher in patients with coronary artery disease than in patients without it. Other studies have reported that glycemic fluctuations are positively correlated with the carotid artery intima-media thickness in patients with T2DM [27, 28], suggesting that glycemic excursions are associated with macro vascular complications.

In the present study we observed significantly increased MDA levels in T2DM patients compared to healthy controls and also positive correlation with HbA1c and HOMA-IR. HbA1c is widely used as mean glycemic index in diabetes and also useful measurement for the vascular complications. Oxidative stress plays a crucial role in pathogenesis of diabetic vascular complication [29]. Chronic hyperglycemia in diabetic patients can increase production of free radicals through Amadori rearrangement [30]. In general, the ROS and RNS are continuously generated in physiological conditions and are eliminated by several antioxidant enzymes. Co-existence of inflammation, increased lipid peroxidation, dyslipidemia along with hyperglycemia conditions could pathologically increase the effect of oxidative stress [31]. However, the decreased efficiency of cellular antioxidant mechanisms with simultaneously enhanced lipid peroxidation along with increased insulin resistance and HbA1c may contribute factors of provoking inflammatory pathways and vascular complications in type 2 diabetes mellitus.

Misra *et al* [32] indicated that hs-CRP levels were higher in patients with T2DM with macrovascular complications than in those with T2DM without macro vascular complications. Another previous study reported the same findings, and also demonstrated that hs-CRP was positively correlated with macrovascular complications [33]. The associations between hs-CRP and HbA1c were analyzed using multivariate stepwise regression analysis, and the results indicated that HbA1c was a risk factor for hs-CRP and that glycemic excursions had a smaller effect on the systemic

inflammatory response than HbA1c did; therefore, when glycemic control treatment is planned, priority should be given to the reduction of the HbA1c levels followed by the reduction of glycemic excursions. This indicates that reducing the inflammatory response defers the occurrence and development of complications associated with DM. [34]

Conclusion:

Increased levels of hs-CRP and MDA in type 2 diabetes patients have the potential to be useful diagnostic indicators for the evaluation of endothelial dysfunction in these individuals. The use of stringent blood glucose management, frequent monitoring of hsCRP, and monitoring of MDA levels may be beneficial in the prevention of vascular problems in type 2 diabetics.

References:

1. Festa A, D'Agostino R, Howard G, Mykkanen L, Tracy RP, et al. ¨ Chronic Subclinical Inflammation as Part of the Insulin Resistance Syndrome. Cirction. 2000;102(1):42–47.
2. 2. Donath MY, Shoelson SE. Type 2 diabetes as an inflammatory disease. Nature Rev Immunol. 2011;11(2):98–107.
3. Wellen KE, Hotamisligil GS. Inflammation, stress, and diabetes. J ClinInvestig. 2005; 115:1111–1120.
4. Moinuddin K, Awanti SM. Evaluation of the relationship between glycemic parameters and serum uric acid level in type 2 diabetes mellitus patients, 2016;3:395-401.
5. Jagtap MW, Rohankar PH, Kale SA. The Relation between serum uric acid & HbA1c in geriatric patients of Type 2 Diabetes in Amravati, Maharashtra, India . Int. J. Bioassays. 2016 ;5(06):4630-32.
6. Arab, A. G., Zahedi, M., Nejad, V. K., Sanagoo, A., &Azimi, M. Correlation between Hemoglobin A1c and Serum Lipid Profle in Type 2 Diabetic Patients Referred to the Diabetes Clinic in Gorgan, Iran. JCBR, 2018;2(1):26-31.
7. Gardiner HM, Pasquini L, Wolfenden J, Kulinskaya E, Li W, Henein M. Increased periconceptual maternal glycated haemoglobin in diabetic mothers reduces fetal long axis cardiac function. Heart. 2006; 92(8):1125-30.
8. Ford ES. The metabolic syndrome and C-reactive protein, fibrinogen, and leukocyte count: findings from the Third National Health and Nutrition Examination Survey. Atheroscler. 2003;168(2):351–359.
9. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. JAMA. 2001;286(3):327–334.
10. Devaraj S, Xu DY, Jialal I. C-Reactive Protein Increases Plasminogen Activator Inhibitor-1 Expression and Activity in Human Aortic Endothelial Cells. Circulation. 2003;107(3):398–404.
11. Fichtlscherer S, Rosenberger G, Walter DH, Breuer S, Dimmeler S, et al. Elevated C-Reactive Protein Levels and Impaired Endothelial Vasoreactivity in Patients With Coronary Artery Disease. Circ. 2000;102(9):1000–1006.
12. Ridker PM, Buring JE, Cook NR, Rifai N. C-reactive protein, the metabolic syndrome, and risk of incident cardiovascular events. An 8-year follow-up of 14,719 initially healthy American women. ACC Curr J Rev. 2003;12(3):33–34.
13. Wellen KE, Hotamisligil GS. Inflammation, stress, and diabetes. J ClinInvestig. 2005; 115:1111–1119.
14. Moldovan L, Moldovan NI. Oxygen free radicals and redox biology of organelles. Histochem Cell Biol. 2004;122(4):395–412.
15. Cheeseman KH, Beavis A, Esterbauer H. Hydroxyl-radicalinduced iron-

catalysed degradation of 2-deoxyribose. Quantitative determination of malondialdehyde. Biochem J. 1988;252(3):649–653.
16. Mahfouz MO, Hariprasad CH, Shaffie IA, Sadasivudu B. Serum malondialdehyde levels in myocardial infarction and chronic renal failure. IRCS Med Sci. 1986; 14:1110-11.
17. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistanceand? Cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 1985; 28(7):412–19.
18. Asegaonkar SB, Marathe A, Tekade ML, Cherekar L, Bavikar J, Bardapurkar J and Ajay R: High-sensitivity C-reactive protein: A novel cardiovascular risk predictor in type 2 diabetics with normal lipid profile. J Diabetes Complications. 25:368–370. 2011.
19. Zhang P, Zhang X, Brown J, Vistisen D, Sicree R, et al. Global healthcare expenditure on diabetes for 2010 and 2030. Diabetes Res Clin Pract. 2010;87(3):293-301.
20. Derosa G, D'Angelo A, Bonaventura A, Bianchi L, Romano D, et al. Effects of berberine on lipid profile in subjects with low cardiovascular risk. Expert OpinBiolTher. 2013;13(4):475-82.
21. Meng J, Ge J, Yu C, Xu L and Gu Q: The analysis of dynamic glucose characteristics in type 2 diabetic patients with different concentration of HbA1c. Zhongguo Tang Niao Bing ZaZhi. 19:816–818. 2011.
22. Sarinnapakorn V and Wanicagool W: Association between hs-CRP and HbA1c in overweight type 2 diabetic female patients. J Med Assoc Thai. 96(Suppl 3): S54–S58. 2013.
23. Kilpatrick ES, Rigby AS and Atkin SL: Effect of glucose variability on the long-term risk of microvascular complications in type 1 diabetes. Diabetes Care. 32:1901–1903. 2009.
24. Sakurai M, Saitoh S, Miura K, Nakagawa H, Ohnishi H, Akasaka H, Kadota A, Kita Y, Hayakawa T, Ohkubo T, et al: HbA1c and the risks for all-cause and cardiovascular mortality in the general Japanese population: NIPPON DATA90. Diabetes Care. 36:3759–3765.
25. Chang CM, Hsieh CJ, Huang JC and Huang IC: Acute and chronic fluctuations in blood glucose levels can increase oxidative stress in type 2 diabetes mellitus. Acta Diabetol. 49 (Suppl 1): S171–S177.
26. Su G, Mi S, Tao H, Li Z, Yang H, Zheng H, Zhou Y and Ma C: Association of glycemic variability and the presence and severity of coronary artery disease in patients with type 2 diabetes. Cardiovasc Diabetol. 10:192011.
27. Mo Y, Zhou J, Li M, Wang Y, Bao Y, Ma X, Li D, Lu W, Hu C, Li M and Jia W: Glycemic variability is associated with subclinical atherosclerosis in Chinese type 2 diabetic patients. Cardiovasc Diabetol. 12:152013.
28. Zhang X, Xu X, Jiao X, Wu J, Zhou S and Lv X: The effects of glucose fluctuation on the severity of coronary artery disease in type 2 diabetes mellitus. J Diabetes Res. 2013:5769162013.
29. Schafer FQ, Buettner GR. Redox environment of the cell as viewed throughtheredoxstate of the glutathione disulfide/glutathione couple. Free Radical Biol Med. 2001;30(11):1191-12.
30. Pérez A. D., Valle D. M., Medina L. C. G., Burgos R. A. O., Reyes J. D. S., Solano O. I. A., Anguila J. J. M. & Rojas M. F. R. Assisted Therapy with Vacuum and Floating Stoma: A New Way to Treat a Periostomal Abscess. Journal of Medical Research and Health Sciences, 2021:4(12), 1629–1635.
31. Giugliano D, Ceriello A, Paolisso G. Oxidative Stress and Diabetic Vascular

Complications. Diabetes Care. 1996;19(3):257-67.
32. Pendyala G, Thomas B, Joshi S. Evaluation of total antioxidant capacity of saliva in type 2 diabetic patients with and without periodontal disease: A case-control study.North Am J Med Sci. 2013;5(1):51.
33. Misra DP, Das S and Sahu PK: Prevalence of inflammatory markers (high-sensitivity C-reactive protein, nuclear factor-κB and adiponectin) in Indian patients with type 2 diabetes mellitus with and without macrovascular complications. Metab Syndr Relat Disord. 10:209–213.
34. Yang Y, Du JL, Zhang XJ, Bai R, Ba Y, Xing Q, Sun LP, Sun GH and Li CC: Association between serum amyloid A and intima-media thickness of common carotid artery in patients with type 2 diabetes. Zhonghua Nei Fen Mi Dai XieZaZhi. 24:188–189.