IJPSR (2015), Vol. 6, Issue 11 **(Research Article)**

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 26 April, 2015; received in revised form, 03 June, 2015; accepted, 10 September, 2015; published 01 November, 2015

HUMAN CARTILAGE GLYCOPROTEIN 39 (YKL-40): A VIEW IN TYPE 2 DIABETES MELLITUS

Rekha Kumari D. [*1], Farid Babu M. [1] and Balu Mahendran K. [2]

Department of Biochemistry [1], Konaseema Institute of Medical Sciences & Research Foundation, Amalapuram, Andhra Pradesh, India.
Division of Biochemistry [2], Rajah Muthiah Medical College and Hospital, Annamalai University, Annamalainagar, Tamil Nadu, India.

Keywords:

Diabetes mellitus (DM), Human cartilage glycoprotein 39 (YKL-40), Glycated hemoglobin (HbA1c)

Correspondence to Author:
D. Rekha Kumari

Assistant professor
Department of Biochemistry, Konaseema Institute of Medical Sciences & Research Foundation, Amalapuram -533201, Andhra Pradesh, India.

E-mail:rekha.dulala@gmail.com

ABSTRACT: Background: Diabetes is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. Type 2 diabetes causes the combination of resistance to insulin action and an inadequate compensatory insulin secretory response. The degree of hyperglycemia sufficient to cause pathologic and functional changes in various target tissues, but without clinical symptoms, may be present for a long period of time before diabetes is detected. Long-term complications of diabetes include retinopathy with potential loss of vision; nephropathy leading to renal failure; peripheral neuropathy with risk of foot ulcers, amputations, and Charcot joints; and autonomic neuropathy causing gastrointestinal, genitourinary, and cardiovascular symptoms and sexual dysfunction. Patients with type 2 diabetes have an increased incidence of atherosclerotic cardiovascular, peripheral arterial, and cerebrovascular complications. **Aim**: The aim of this study was to determine the association between plasma human cartilage glycoprotein 39 (YKL-40) and glycated hemoglobin (HbA1C) in type 2 diabetic patients. **Materials and methods:** Thirty type 2 diabetic patients with the age group of 35 to 50 years were selected for this study and 30 age matched healthy individuals were selected as a control group. Plasma YKL-40 was assessed by ELISA method and routine investigations was done fully automated analyzer. **Results:** The mean levels of Plasma YKL-40 was significantly increased in diabetic patients compared with controls. Plasma YKL-40 was positively correlated with HbA1c. **Conclusion:** Plasma YKL-40 might be useful to detect early stages of inflammation and endothelial dysfunction in T2DM patients. Hence measurement of plasma YKL-40 could be useful diagnostic marker for the assessment of vascular complications in type 2 diabetic patients.

INTRODUCTION: Diabetes is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.6(11).4852-56

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.6(11).4856-56

Type 2 diabetes which accounts for 90–95% of those with diabetes, previously referred to as non–insulin-dependent diabetes, or adult-onset diabetes, encompasses individuals who have insulin resistance and usually have relative (rather than absolute) insulin deficiency.

An elevated rate of basal hepatic glucose production in the presence of hyperinsulinemia is the primary cause of fasting hyperglycemia; after a meal, impaired suppression of hepatic glucose production by insulin and decreased insulin-mediated glucose uptake by muscle contribute

almost equally to postprandial hyperglycemia [1]. The risk of each of the microvascular and macrovascular complications of type 2 diabetes associated with hyperglycemia [2]. In patients with type 2 diabetes previous prospective studies have shown an association between the degree of hyperglycemia and increased risk of microvascular complications, sensory neuropathy, myocardial infarction, stroke, macrovascular mortality, and all-cause mortality [3-10].

Human cartilageglycoprotein – 39 (YKL-40, chitinase-3-like-1 [CHI3L1], is a heparin-, chitin-, and collagen-binding lectin) produced by immunologically active cells such as macrophages [11] and neutrophils [12]. YKL-40 is a member of the mammalian chitinase-like proteins and is a phylogenetically highly conserved serum protein [13-15]. Other cells shown to produce YKL-40 are vascular smooth muscle and endothelial cells [16-18], arthritic chondrocytes [13], and cancer cells [19]. YKL-40 was found to act as an inflammatory marker in relation to both acute and chronic inflammation as well as participate in the processes during the early stages of atherosclerosis [20, 21, 22]. So in this view the aim of the present study was to determine association between plasma YKL-40 levels and HbA1c in subjects with type 2 diabetes mellitus compared with normal subjects.

MATERIALS AND METHODS:

A total of 30 type 2 diabetic patients of both sexes aged between 35-50 years on oral hypoglycemic drugs, attending diabetic out-patient department of Konaseema Institute of Medical Sciences &Research foundation, Amalapuram, Andhra Pradesh, India, were selected for our study after approval of Institutional Human ethics committee. We excluded the patients based on the following criteria: Patients on insulin, Smokers, Alcoholics, Tobacco chewers, abnormal urinary sediment, urinary tract infection, history of other active or chronic persistent infection or inflammatory disorders, neoplastic disorders, uncontrolled thyroid disorders, severe liver dysfunction, history of acute myocardial infarction, stroke, and occlusive peripheral vascular disease. Thirty healthy individual age, sex matched subjects were selected as control. Experiments were done in accordance with Helsinki declaration of 1975.

Biochemical analysis:

A fasting blood samples were obtained from the subjects immediately after enrolment. Blood samples were centrifuged at 2000×g for 10 min. Samples were analyzed for routine investigations blood sugar, lipid profile (Total Cholesterol, HDL,Triglycerides), glycosylated hemoglobin (HbA1C). Plasma YKL-40, insulin assessed by ELISA and the 2 hour post prandial venous plasma glucose (PPBS) estimation was also done.

Statistical analysis: Statistical analyses were carried out with SPSS 20.0. Values were expressed as mean ± standard deviation, p value < 0.05 was considered statistically significant. Normally distributed data were analyzed by using one-way ANOVA. The Pearson correlation test was used for correlation analysis.

RESULTS:

TABLE 1: COMPARISON OF BASELINE CHARACTERISTICS BETWEEN CONTROL AND TYPE 2 DIABETIC SUBJECTS.

Parameters	Control (n=30)	Study group(n=30)	p- value
Age	45.95±3.4	44.4±2.7	0.08
Body mass index	25.2±1.3	26.4±2.7	0.07
Waist/Hip ratio	0.90±0.04	0.92±0.06	0.21
Systolic BP (mm Hg)	114.4±6.9	126.4±17.1	0.004
Diastolic (mm Hg)	73.5±3.2	77.8±8.9	0.045

Data are expressed as mean ±SD, p value <0.05 was considered statistically significant.

TABLE 2: COMPARISON OF BIOCHEMICAL PARAMETERS BETWEEN CONTROL AND TYPE 2 DIABETIC SUBJECTS.

Parameters	Control (n=30)	Study group(n=30)	p- value
FBS(mg/dl)	82.7±6.5	138.7±57.1	0.001
PPBS(mg/dl)	112.2±11.6	217.8±45.3	0.001
HbA1C	5.7±1.1	7.5±1.2	0.001
Serum cholesterol (mg/dl)	164.5±7.1	170.1±12.8	0.533

Serum Triglycerides (mg/dl)	93±5.3	137.5±48.5	0.001
HDL cholesterol (mg/dl)	42.5±2.4	40.2±2.6	0.001
LDL cholesterol (mg/dl)	112.1±9.7	114.1±28.4	0.717
Urea (mg/dl)	23.6±5.8	26.7±6.4	0.78
Creatinine(mg/dl)	0.6±0.2	0.8±0.3	0.002
AST(IU/L)	27.2±3.2	28.9±4.5	0.78
ALT(IU/L)	28.9±4.0	30.1±6.2	0.55
ALP(IU/L)	90.0±9.9	94.5±8.7	0.60
Insulin (µIU/mL)	6.8±0.7	11.5±5.3	0.001
HOMA-IR	1.3±0.17	3.8±2.2	0.001
Plasma YKL-40 (ng/ml)	19.9±3.7	46.0±10.4	0.001

Data are expressed as mean ±SD, p value <0.05 was considered statistically significant.

TABLE 3: CORRELATION BETWEEN PLASMA YKL-40 &MEASURED PARAMETERS

Parameters	Correlation Coefficient(r)	p- value
FBS	0.461**	0.001
PPBS	0.603**	0.001
HbA1C	0.486**	0.001
HOMA-IR	0.621**	0.001
Cholesterol	0.043	0.743
TGL	0.422**	0.001
HDL	-0.375**	0.003
LDL	0.027**	0.835

**Correlation is significant at the 0.01 level (2-tailed).

DISCUSSION: Glycemic exposure, i.e. the extent and duration of hyperglycemia, is a major risk factor for both micro and macrovascular complications of diabetes. Glycemic variability comprises "glucose variability" and "HbA1c variability". Glucose variability relates to within day fluctuations of glycemia, especially as a consequence of post-prandial hyperglycemia [23], which may eventually reflect in HbA1c levels above the normal range. Conversely, HbA1c variability relates to changes in glycaemia over longer periods of time [24]. However, three retrospective analyses of the Diabetes Control and Complications Trial (DCCT) found that within-day glucose variability does not contribute to the development of microvascular complications [25-27]. Recently, microvascular complications were shown to be predicted by HbA1c variability from one visit to the next, independently of average HbA1c and known risk factors for microangiopathy, both in type 1 [28-30] and type 2 [31-33] diabetes.

Micro- and macrovascular complications are known to decrease the quality of life as well as shortens the life in diabetic patients. Despite intensive research in the pathological mechanisms resulting in improved and intensified treatment of diabetes and its vascular risk factors and complications, there is still a need for supplementary risk markers to understand the pathogenesis and predict the development of micro and macrovascular disease. So the measurement various inflammatory markers in biologic fluids might be useful for detection vascular complications in diabetic patients.

In the present study, we observed that plasma YKL-40 levels were significantly increased in type 2 diabetic patients. It therefore seems to be reasonable that it is the low-grade inflammation and endothelial dysfunction that account for the elevated YKL-40 levels in diabetic patients. And also our study shows that plasma YKL-40 levels show strong positive correlation with HbA1C and HOMA-IR. Chronic hyperglycemia produces reactive oxygen species (ROS), protein glycation reactions which leads to the formation of advanced glycation end products (AGEs) and activates inflammatory signaling cascades, that leads inflammatory changes [34, 35]. Consequently, AGEs have key roles in the pathogenesis of vascular complications [35].

In addition, our study shows that Plasma YKL-40 levels shows strong positive correlation with LDL, TGL and negative correlation with HDL. AGEs may promote atherogenesis by oxidizing low density lipoproteins (LDL) and causing changes in

the intimal collagen [36]. The cellular effects of AGEs are mediated by specific receptors, one of which is the receptor for AGE (RAGE). The presence of RAGE has been demonstrated in all cells relevant to the atherosclerotic process including monocytes, macrophages, endothelial cells, and smooth muscle cells [37]. These vascular cells do not express significant amounts of RAGE under physiological conditions but can be induced to express RAGE in situations where either ligands accumulate and/or various transcription factors regulating RAGE are activated [38]. In the diabetic vasculature, cells expressing high levels of RAGE are often proximal to or co-localized in areas in which AGEs are abundant [39, 40].

Aside from AGEs, tumor necrosis factor-α (TNF-α), S100/calgranulins, β-amyloid, amphoterin, and oxidative stress can upregulate RAGE expression, thus rendering these cells more susceptible to the effects of AGEs and chronic inflammation [38, 41, 42].

Cell and animal studies suggest that limiting RAGE expression in vascular cells could modulate expression of various proinflammatory mediators and prevent atherosclerosis development [43, 44]. So therefore all these factors along with chronic hyperglycemia, low-grade inflammation and endothelial dysfunction may contribute the elevated YKL-40 levels in diabetic patients. Some other studies shown that patients that YKL-40 levels are associated with the presence and extent of CAD, are even higher in patients with MI and are associated with all-cause as well as cardiovascular mortality [45, 46]. In this view, the precise role of YKL-40 remains elusive, but our study suggest that YKL-40 might be involved vascular complications of type 2 diabetic patients.

In conclusion plasma YKL-40 might be useful to detect early stages of inflammation and endothelial dysfunction in T2DM patients. Hence measurement of plasma YKL-40 could be potentially useful diagnostic marker for the assessment of vascular complications in type 2 diabetic patients.

REFERENCES:

1. American Diabetes Association .Diagnosis and Classification of Diabetes Mellitus. Diabetes Care .2014; 37:S81-S90.
2. Fox CS, Coady S, Sorlie PD, et al. Increasing cardiovascular disease burden due to diabetes mellitus: the Framingham Heart Study. Circulation 2007; 115:1544-1550.
3. Moss SE, Klein R, Klein BE. Cause-specific mortality in a population-based study of diabetes. Am J Public Health 1991; 81:1158-1162.
4. Klein R. Hyperglycemia and microvascular and macrovascular disease in diabetes. Diabetes Care 1995; 18:25868.
5. UKPDS Group. Risk factors for coronary artery disease in non-insulin dependent diabetes (UKPDS 23). BMJ 1998; 316:82328.
6. Kuusisto J, Mykkänen L, Pyörälä K, Laakso M. NIDDM and its metabolic control predict coronary heart disease in elderly subjects. Diabetes.1994; 43:96067.
7. Lehto S, Ronnemaa T, Pyörälä K, Laakso M. Predictors of stroke in middle-aged patients with non-insulin-dependent diabetes. Stroke 1996; 27:638.
8. Groeneveld Y, Petri H, Hermans J, Springer MP. Relationship between blood glucose level and mortality in type 2 diabetes mellitus: a systematic review. Diabet Med 1999; 116:213.
9. Uusitupa MI, Niskanen LK, Siitonen O, Voutilainen E, Pyörälä K. Ten-year cardiovascular mortality in relation to risk factors and abnormalities in lipoprotein composition in type 2 (non-insulin- dependent) diabetic and non-diabetic subjects. Diabetologia 1993; 36:117584.
10. Wei M, Gaskill SP, Haffner SM, Stern MP. Effects of diabetes and level of glycaemia on all-cause and cardiovascular mortality. Diabetes Care 1998; 21:116772.
11. Johansen JS: Studies on serum YKL-40 as a biomarker in diseases with inflammation, tissue remodelling, fibroses and cancer. Dan Med Bull 2006; 53:172-209.
12. Volck B, Price PA, Johansen JS, Sorensen O, Benfield TL, Nielsen HJ, Calafat J, Borregaard N: YKL-40, a mammalian member of the chitinase family, is a matrix protein of specific granules in human neutrophils. Proc Assoc Am Physicians 1998; 110:351-360.
13. Hakala BE, White C, Recklies AD: Human cartilage gp-39, a major secretory product of articular chondrocytes and synovial cells, is a mammalian member of a chitinase protein family. J Biol Chem 1993; 268:25803-810.
14. Rehli M, Krause SW, Andreesen R: Molecular characterization of the gene for human cartilage gp-39 (CHI3L1), a member of the chitinase protein family and marker for late stages of macrophage differentiation. Genomics 1997; 43:221-225.
15. Bussink AP, Speijer D, Aerts JM, Boot RG: Evolution of mammalian chitinase(-like) members of family 18 glycosyl hydrolases. Genetics 2007; 177:959-970.
16. Shackelton LM, Mann DM, Millis AJ: Identification of a 38-kDa heparin binding glycoprotein (gp38k) in differentiating vascular smooth muscle cells as a member of a group of proteins associated with tissue remodeling. J Biol Chem 1995;270:13076 -13083
17. Malinda KM, Ponce L, Kleinman HK, Shackelton LM, and Millis AJ: Gp38k, a protein synthesized by vascular smooth muscle cells, stimulates directional migration of human umbilical vein endothelial cells. Exp Cell Res 1999; 250:168 - 173
18. Nishikawa KC, Millis AJ: gp38k (CHI3L1) is a novel adhesion and migration factor for vascular cells. Exp Cell Res 2003;287:79 -87
19. Johansen JS, Jensen BV, Roslind A, Nielsen D, Price PA: Serum YKL-40, a new prognostic biomarker in cancer patients? Cancer Epidemiol Biomarkers Prev.2006; 15:194 - 202
20. Rathcke, C. N., Johansen, J. S., & Vestergaard, H. YKL-40, a biomarker of inflammation, is elevated in patients with type 2 diabetes and is related to insulin resistance. Inflammation Research .2006; 55:53-59.

21. Rathcke, C. N., & Vestergaard, H.YKL-40, a new inflammatory marker with relation to insulin resistance and
22. Nielsen, A. R., Erikstrup, C., Johansen, J. S., Fischer, C. P., Plomgaard, P., Krogh-Madsen, R., Taudorf, S., Lindegaard, B., & Pedersen, B. K. Plasma YKL-40: a BMI independent marker of type 2 diabetes. Diabetes.2008; 57: 3078-3082.
23. Home PD: Contributions of basal and post-prandial hyperglycaemia to micro- and macrovascular complications in people with type 2 diabetes. Curr Med Res Opin. 2005, 21:989-998.
24. Kilpatrick ES: The rise and fall of HbA1c as a risk marker for diabetes complications. Diabetologia. 2012; 55:2089-2091.
25. Kilpatrick ES, Rigby AS, Atkin SL: The effect of glucose variability on the risk of microvascular complications in type 1 diabetes. Diabetes Care 2006; 29: 1486-1490.
26. Kilpatrick ES, Rigby AS, Atkin SL: Effect of glucose variability on the long- term risk of microvascular complications in type 1 diabetes. Diabetes Care.2009; 32:1901-1903.
27. Siegelaar SE, Kilpatrick ES, Rigby AS, Atkin SL, Hoekstra JB, and Devries JH: Glucose variability does not contribute to the development of peripheral and autonomic neuropathy in type 1 diabetes: data from the DCCT. Diabetologia .2009; 52:2229-2232.
28. Kilpatrick ES, Rigby AS, Atkin SL: A1C variability and the risk of microvascular complications in type 1 diabetes: data from the Diabetes Control and Complications Trial. Diabetes Care. 2008; 31:2198-202.
29. Wadén J, Forsblom C, Thorn LM, Gordin D, Saraheimo M, Groop PH, Finnish Diabetic Nephropathy Study Group: A1C variability predicts incident cardiovascular events, microalbuminuria, and overt diabetic nephropathy in patients with type 1 diabetes. Diabetes 2009; 58:2649-2655.
30. Marcovecchio ML, Dalton RN, Chiarelli F, Dunger DB: A1C variability as an independent risk factor for microalbuminuria in young people with type 1 diabetes. Diabetes Care .2011; 34:1011–1013.
31. Sugawara A, Kawai K, Motohashi S, Saito K, Kodama S, Yachi Y, Hirasawa Shimano H, Yamazaki K, Sone H: HbA(1c) variability and the development of microalbuminuria in type 2 diabetes: Tsukuba Kawai Diabetes Registry 2. Diabetologia. 2012; 55:2128–2131.
32. Hsu CC, Chang HY, Huang MC, Hwang SJ, Yang YC, Lee YS, Shin SJ, Tai HbA(1c) variability is associated with microalbuminuria development type 2 diabetes: a 7-year prospective cohort study. Diabetologia .2012; 55:3163–3172.
33. Penno G, Solini A, Bonora E, Fondelli C, Orsi E, Zerbini G, Morano S, Cavalot F, Lamacchia O, Laviola L, Nicolucci A, Pugliese G, Renal Insufficiency and Cardiovascular Events (RIACE) Study Group: HbA 1cvariability as an independent correlate of nephropathy, but not retinopathy, in patients with type 2 diabetes: The Renal Insufficiency and Cardiovascular Events (RIACE) Italian Multicenter Study. Diabetes Care 2013. epub Mar 14.
34. Aidan Ryan, Madeline Murphy, Catherine Godson, Fionnuala B. Hickey. Diabetes mellitus and apoptosis: inflammatory cells. Apoptosis .2009; 14:1435-1450.
35. Shi Fang Yan, Ravichandran Ramasamy, Ann Marie Schmidt. Mechanisms of Disease: advanced glycation end-products and their receptor in inflammation and diabetes complications. Nature Reviews Endocrinology .2008;4: 285-293
36. Wendt T, Bucciarelli L, Qu W, et al. Receptor for Advanced Glycation Endproducts (RAGE) and vascular inflammation: insights into the pathogenesis of macrovascular complications in diabetes. Curr Athe- roscler Rep .2002; 4:228 -37.
37. Brett J, Schmidt AM, Yan SD, Zou YS, Weidman E, Pinsky D, Nowygrod R, Neeper M, Przysiecki C, Shaw A: Survey of the distribution of a newly characterized receptor for advanced glycation end products in tissues. Am J Pathol .1993;143 : 1699 -712
38. Bierhaus A, Humpert PM, Morcos M, Wendt T, Chavakis T, Arnold B, Stern DM, Nawroth PP: Understanding RAGE, the receptor for advanced glycation end products. J Mol Med. 2005;83 : 876 -886
39. Soulis T, Thallas V, Youssef S, Gilbert RE, McWilliam BG, Murray-McIntosh RP, Cooper ME: Advanced glycation end products and their receptors co-localise in rat organs susceptible to diabetic microvascular injury. Diabetologia. 1997; 40: 619 -628
40. Tanji N, Markowitz GS, Fu C, Kislinger T, Taguchi A, Pischetsrieder M, Stern D, Schmidt AM, D'Agati VD: Expression of advanced glycation end products and their cellular receptor RAGE in diabetic nephropathy and nondiabetic renal disease. J Am Soc Nephrol .2000; 11 : 1656 -1666
41. Schmidt AM, Yan SD, Yan SF, Stern DM: The biology of the receptor for advanced glycation end products and its ligands. Biochim Biophys. 2000; 1498: 99 -111.
42. Kim W, Hudson BI, Moser B, Guo J, Rong LL, Lu Y, Qu W, Lalla E, Lerner S, Chen Y, Yan SS, D'Agati V, Naka Y, Ramasamy R, Herold K, Yan SF, Schmidt AM: Receptor for advanced glycation end products and its ligands: a journey from the complications of diabetes to its pathogenesis. Ann N Y Acad Sci .2005; 1043: 553 -561.
43. Park L, Raman KG, Lee KJ, Lu Y, Ferran LJ Jr, Chow WS, Stern D, Schmidt AM: Suppression of accelerated diabetic atherosclerosis by the soluble receptor for advanced glycation end products. Nat Med .1998;4 : 1025 -1031
44. Bucciarelli LG, Wendt T, Qu W, Lu Y, Lalla E, Rong LL, Goova MT, Moser B, Kislinger T, Lee DC, Kashyap Y, Stern DM, Schmidt AM: RAGE blockade stabilizes established atherosclerosis in diabetic apolipoprotein E-null mice. Circulation .2001; 106 : 2827 -2835
45. Kastrup J, Johansen JS, Winkel P, Hansen JF, Hildebrandt P, Jensen GB, Jespersen CM, Kjoller E, Kolmos HJ, Lind I, and Nielsen H, Gluud C: High serum YKL-40 concentration is associated with cardiovascular and all-cause mortality in patients with stable coronary artery disease. Eur Heart J .2009, 30:1066-1072.
46. Nojgaard C, Host NB, Christensen IJ, Poulsen SH, Egstrup K, Price PA, and Johansen JS: Serum levels of YKL-40 increases in patients with acute myocardial infarction. Coron Artery Dis. 2008; 19:257-263.

How to cite this article:
Kumari DR, Babu MF and Mahendran KB: Human Cartilage Glycoprotein 39 (YKL-40): A View in Type 2 Diabetes Mellitus. Int J Pharm Sci Res 2015; 6(11): 4852-56.doi: 10.13040/IJPSR.0975-8232.6(11).4852-56.