Glycated Hemoglobin [HbA1C] As a Dual Marker for Glycemic Status and Dyslipidemia in Diabetics: A Cross Sectional Analysis of 450 Cases

Lavanya Rajagopal*, Balaji Ramraj**, Sundaram Arunachalam***, Veena Raja*, Shivashekar Ganapathy****

*Assistant Professor ***Dean (Medical) Professor ****Professor and HOD, Department of Pathology **Associate Professor, Department of Community Medicine, SRM Medical College Hospital and Research Centre, Kattankulathur, Chennai 603203. India.

Abstract

Background: The prevalence of dyslipidemia in diabetes mellitus [DM] is 95%. Diabetic patients with accompanied dyslipidemia are soft targets of cardiovascular deaths. Glycated Hemoglobin [HbA1C] the gold standard for assessing the glycemic status, has been regarded as an independent risk factor for Coronary Heart Disease [CHD] and stroke. Various studies have contradicting results on the impact of HbA1C levels on lipid profile in diabetics. *Aim:* To find out association between glycemic control (HbA1C), age, gender and serum lipid profile in type 2 diabetic patients and non-diabetic subjects. *Materials and Methods:* A cross-sectional study carried out in SRM Hospital and Research Centre, Chennai. Total 450 subjects (150 controlled & 150 poorly controlled diabetics, 150 non-diabetics) with equal number of males and females in each group and investigated for HbA1C and lipid profile. *Statistical Analysis:* The data were analyzed by SPSS version 21 using Independent samples student 't' test. 'p' value <0.05 was considered as statistically significant. *Results:* There was a significant increase in mean Total Cholesterol [TC], Triglyceride[TG], Low Density Lipoprotein-Cholesterol [LDL-C], Fasting Blood Glucose [FBG] levels and HbA1C between non-diabetics, controlled and poorly controlled diabetics. But High Density Lipoprotein-Cholesterol [HDL-C] was not significant between these study groups. TC, TG, LDL-C levels between males and females in each group were not statistically significant. But mean HDL-C was non-significantly lower in female diabetics. Subjects with worse glycemic control (HbA1C >9%) possessed significantly high values of TC, TG, LDL-C, FBG except HDL-C. No significant correlation for age with respect to serum lipid profile. *Conclusion:* We conclude, HbA1C predicts dyslipidemia. This highlights the usefulness of HbA1C as dual biomarker (glycemic control and lipidemic state) for screening high risk diabetic patients.

Keywords: Glycated Hemoglobin (HbA1C); Dyslipidemia; Diabetes.

Introduction

Diabetic patients with accompanied (but often unnoticed) dyslipidemia are soft targets of cardiovascular deaths [1].The prevalence of dyslipidemia in diabetes mellitus is 95% and is characterized by increased Triglyceride [TG], High Density Lipoprotein - Cholesterol [HDL-C] and small dense Low Density Lipoprotein – Cholesterol [LDL-C] [2]. An early intervention to normalize circulating lipids in diabetics has been shown to reduce cardiovascular complications and mortality [1].

Glycated Hemoglobin [HbA1C] is widely used as an indicator of glycemic status over previous three months. Recently, elevated HbA1C has been regarded as an independent risk factor for Coronary Heart Disease [CHD]and stroke in subjects with and without diabetes [3]. Estimated risk of Cerebrovascular Disease [CVD] has shown to be increased by 18% for each 1% increase in absolute HbA1C value in diabetics [4].

Corresponding Author: Lavanya Rajagopal, Door no. 902, B-Block, SRM Medical Staff Quarters, SRM Medical College Hospital and Research Centre, Kattankulathur, Pincode: 603203, Chennai India.

E-mail: drrlavan@yahoo.co.in

A few studies have previously tried to find the correlation between HbA1C and lipid profile. Some of these have shown that all the parameters of lipid profile have significant correlation with glycemic control [5].

Giansanti et al observed significantly higher levels of hypercholesterolemia and hyperlipidemia in type 2 diabetic patients with CVD as compared to patients without CVD [6]. Ravipati G et al observed a direct correlation between HbA1C and the severity of CHD in diabetic patients [7].

On the other hand some studies do not report significant correlation between glycemic control and all the parameters of lipid profile [8]. Even in non-diabetic cases with HbA1C levels within normal range, positive relationships between HbA1C and CVD has been demonstrated [9].

These controversies inspired us to take forward this study which was aimed to find out association between glycemic control (HbA1C), age, gender and serum lipid profile in type 2 diabetic patients and non-diabetic subjects.

Materials and Methods

This is a descriptive analytical cross-sectional study carried out in SRM Medical College Hospital and Research Centre, Chennai (February 2016 to December 2016) and approved by institutional ethical committee. Total 450 subjects were divided into 3 groups which included 150 controlled diabetics [known diabetics with Fasting Plasma Glucose (FPG) < 126 mg/dl since last 6 months], 150 poorly controlled diabetes [FPG > 126 mg/dl since last 6 months], 150 non-diabetics [no history of Diabetes Mellitus (DM) and two FPG values < 126 mg/dl performed close to the date of the complete blood count]. All study subjects were investigated for dyslipidemia.

Venous blood samples were collected from all the subjects after atleast 8 hours fasting. Blood samples were collected into EDTA tubes for HbA1C and then serum separated tube for fasting blood glucose and lipid profile measurement.

Reference Values

For serum lipid reference level, National Cholesterol Education Programme [NCEP], Adult Treatment Panel III [ATP III] [NCEP – ATP III] guideline was referred. Hypercholesterolemia defined as Total Cholesterol [TC] > 200 mg/dl, LDL-C > 100 mg/dl, Hypertriglyceridemia > 150 mg/dl and HDL-C < 40

mg/dl. Dyslipidemia defined by presence of one or more than one abnormal serum lipid concentration [4].

Measurements

HbA1C assay was done by High Performance Liquid Chromatography [HPLC]. FBG, TC, TG, HDL-C was measured by enzymatic method by using OLYMPUS AU400 autoanalyzer on the same day of collection.

The levels of LDL-C calculated by using Friedwald's formula [10]

$$LDL\text{-}C = TC - HDL\text{-}C - (TG/5)$$

The impact of glycemic control on various parameters was evaluated by categorizing all the patients (diabetic and non-diabetic) into 3 categories on the basis of HbA1C levels.

HbA1C <6% - good glycemic control

HbA1C 6-9% - poor glycemic control

HbA1C >9% - worse glycemic control.

Inclusion Criteria

1. Both males and females >30 years
2. Known type 2 diabetic patients and non-diabetics.

Exclusion Criteria

1. Patients with type I diabetes
2. Patients on anti-lipidemic therapy, steroids
3. Family history of hypercholesterolemia
4. Body Mass Index >30

Statistical Analysis

The data were analyzed by SPSS version 21. Independent samples student 't' test was used to compare the means of different parameters between males and females. One way analysis of variance (ANOVA) to examine the significance levels of various biochemical parameters in study groups. Univariate analysis was performed to evaluate the effects of gender, age and glycemic control on serum lipid profile. 'p' value <0.05 was considered as statistically significant.

Results

In our study total 450 diabetic and non-diabetic subjects were evaluated for lipid profile. The sex

distribution showed an equal number of males and females in all the groups. There was a linear and significant increase in the mean TC, TG, LDL-C, FBG levels and HbA1C between non-diabetics, controlled diabetics and poorly controlled diabetics. But HDL-C was not significant between these study groups [Table 1].

Table 2 shows the comparison between the mean biochemical parameters with respect to gender in 3 study groups. We found that TC, TG, LDL-C levels between males and females in each group were not statistically significant. Also our results showed that the mean HDL-C concentration was non-significantly lower in the female diabetics as compared to that in the male diabetics.

Table 1: Comparison of Lipid profile, FBG and HbA1C in diabetic and control groups

Parameter	Non-Diabetics (Mean ± SD)	Controlled Diabetics (Mean ± SD)	Poorly Controlled Diabetics (Mean ± SD)	ANOVA p value
Total cholesterol	158.61 ± 3.24	158.85 ± 3.03	174.67 ± 4.05	7.040, 0.001
Triglycerides	114.96 ± 4.84	116.98 ± 5.43	172.62 ± 10.12	20.736, 0.0001
HDL-C	39.81 ± 1.45	37.77 ± 0.80	37.62 ± 1.08	1.151, 0.317
LDL-C	105.83 ± 3.03	109.73 ± 2.81	124.27 ± 3.38	9.929, 0.0001
FBG	98.47 ± 1.10	100.33 ± 1.20	200.42 ± 5.94	269.251, 0.0001
HbA1C	5.98 ± 0.08	7.33 ± 0.14	9.72 ± 0.17	188.786, 0.0001

Table 2: Frequency of Lipid parameters in the Diabetic and Control Groups according to the ATP III classification. [M: Male, F: Female, T: Total]

Parameter		Non-Diabetics			Controlled Diabetics			Poorly Controlled Diabetics		
		M n=75	F n=75	T n=150	M n=75	F n=75	T n=150	M n=75	F n=75	T n=150
Total cholesterol (mg/dl)	Desirable (<200)	68	68	136	65	65	130	56	52	108
	Borderline high (200-239)	4	4	8	7	8	15	15	15	30
	High (≥240)	3	3	6	3	2	5	4	8	12
Triglycerides (mg/dl)	Normal (<150)	58	65	123	59	66	125	37	41	78
	Borderline high (150-199)	8	8	16	9	3	12	12	14	26
	High (≥200)	9	2	11	7	6	13	26	20	46
HDL-C (mg/dl)	Low (<40)	32	21	53	29	19	48	44	57	101
	Borderline High (40-59)	38	50	93	44	52	96	26	18	44
	High (≥60)	5	4	9	2	4	6	2	3	5
LDL-C (mg/dl)	Optimal(<100)	29	39	68	33	31	64	21	17	38
	Near optimal (100-129)	24	24	48	22	27	49	24	21	45
	Borderline high(130-159)	17	6	23	17	11	28	19	20	39
	High(160-189)	4	4	8	1	5	6	7	13	20
	Very high(≥190)	1	2	3	2	1	3	4	4	8

Table 3: Correlation between Lipid profile and HbA1C

Parameter	HbA1C% <6% (n=156)	6-9% (n=184)	>9% (n=110)	ANOVA p value
Total cholesterol	153.32 ± 2.73	165.12 ± 3.10	180.46 ± 4.97	14.602, 0.0001
Triglycerides	112.44 ± 3.80	125.32 ± 5.99	185.98 ± 13.08	25.932, 0.0001
HDL-C	39.16 ± 0.99	38.72 ± 1.27	36.78 ± 0.90	1.013, 0.364
LDL-C	103.97 ± 2.31	115.19 ± 3.12	126.12 ± 4.25	12.267, 0.0001
FBG	97.79 ± 0.95	120.97 ± 2.57	203.37 ± 8.47	154.952, 0.0001

HbA1C also demonstrated direct and significant correlation with TC, TG, LDL-C, FBG except HDL-C. Also we observed that worse glycemic control (HbA1C >9%) possessed significantly high values of TC, TG, LDL-C, FBG except HDL-C as compared to patients having poor(HbA1C 6-9%) and good(HbA1C <6%) glycemic control [Table3/Figure 1].
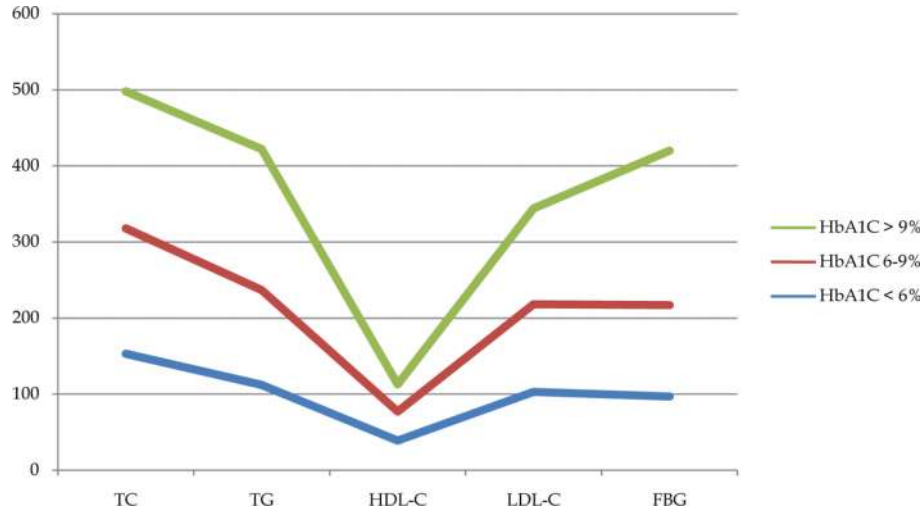
TC - Total Cholesterol, TG – Triglyceride, HDL-C – High Density Lipoprotein Cholesterol, LDL-C – Low Density Lipoprotein Cholesterol, FBG – Fasting Blood Glucose We categorized the study subjects into 4 groups based on ageviz < 50 years, 51-70 years, 61-70 years, and >70 years and then compared the mean values of TC, TG, LDL-C, FBG, HDL-C and HbA1C.

We observed that there was no significant correlation for age with respect to serum lipid profile [Table 4].

Table 4: Correlation between patients Age and lipid parameters

Parameter	Age in Years				
	≤50 (n=225) Mean ±SE	51-60 (n=91) Mean ±SE	61-70 (n=101) Mean ±SE	>70 (n=33) Mean ±SE	ANOVA p value
Total cholesterol	166.00 ± 2.67	161.24 ± 4.93	162.47 ± 4.47	163.27 ± 8.15	0.330, 0.804
Triglycerides	134.14 ± 6.98	136.97 ± 8.40	141.17 ± 7.82	113.36 ± 10.23	0.785, 0.503
HDL-C	37.83 ± 0.98	40.44 ± 1.79	37.33 ± 0.95	39.94 ± 1.86	1.099, 0.349
LDL-C	114.83 ± 2.42	109.48 ± 4.55	113.33 ± 3.76	113.00 ± 7.27	0.415, 0.742
FBG	128.97 ± 4.57	137.59 ± 6.02	135.34 ± 5.99	141.67 ± 11.87	0.687, 0.560
HbA1C	7.51 ± 0.16	7.97 ± 0.24	7.84 ± 0.22	7.54 ± 0.28	1.136, 0.334

Discussion

Diabetes Mellitus [DM] is a hereditary chronic and endocrine metabolic disorder which causes deaths worldwide [10]. The International Diabetes Federation [IDF] reported that total number of diabetic subjects in India is 41 million in 2006 and that this would raise to 70 million by the year 2025 [11].

Lipid abnormalities are common in diabetics and frequently seen in type 2 DM. Dyslipidemia make diabetics prone to develop CHD and other complications of atherosclerosis [1]. In the present study, the pattern of lipid profile parameters in diabetic subjects and its correlations with FBG, HbA1C, age, gender are seen.

In this study, we observed a significant correlation between glycemic status and lipid profile. High levels of TC, TG and LDL-C were seen in more number of poorly controlled diabetics when compared with non-

diabetics and controlled diabetics which was statistically significant. This agrees with the results of Petitti et al who observed significant increase in TC, TG, LDL-C in type 2 diabetes with poor glycemic control [12]. Also similar results were observed by Rosediani et al who reported that increasing non-HDL cholesterol concentration had significant curvilinear relationships with CVD and CHD risk [13]. Moreover NCEP ATP III has recommended using non-HDL cholesterol in assessing CVD risk in patients with diabetes [13,14].

Our study results confirm that glycemic control is directly related to lipid metabolism. This coincides with the research results of ZE Onal et al, who suggested that TG rich lipoproteins accumulate in the insulin resistant state, causing the decreased activity of lipoprotein, increased lipolysis in adipose tissue and increased output of Very Low Density Lipoprotein [VLDL] particles from the liver. They also concluded that poor glycemic control group correlated with

higher TC, LDL-C levels compared to both control and optimal glycemic control group which goes in hand with our results [15].

Our study results contradicts with the results of Zhe Yan et al, Khaw KT who reported no significant correlation between glycemic control and all parameters of lipid profile [8,9].

Insulin regulates the enzymatic activity of Lipoprotein Lipase [LPL] and cholesterol ester transport protein. All these factors are likely cause of dyslipidemia in diabetes. Moreover insulin deficiency reduces the activity of hepatic lipase and several steps in the production of biologically active LPL may be altered in DM [16].

In our study,non-significantly lower levels of HDL-C was more frequently observed among poorly controlled diabetics when compared with controlled diabetics and non-diabetics. Similar results were observed in study done by Khan et al [1]. Recent studies have shown that many factors contribute to the HDL dysfunction that is witnessed in T2DM. Oxidation and glycation of HDL-associated proteins were shown torender them inactive. By changing the gene expression and activity of HDL-metabolizing enzymes, metabolic abnormalities associated with DM compromise the efficiency of reverse cholesterol transport. Finally, the constantly increased inflammatory state in DM results in majorchanges in the HDL proteome; changes that not only devoid the HDL of its normal function,but also turn it into a proatherogenic particle [17]. HDL levels were found to be decreased in T2DM, with a predominance of small dense HDL particles that undergo rapid catabolism [18,19]. This alteration in HDL is the result of an augmentation in the activity of several lipolytic and HDL-modifying enzymes, alongside an increment in triglyceride levels [17].

Individuals with type 2 DM and CHD tend to have small HDL particles. Hypertriglyceridemia and hyper-insulinemia are independently associated with low levels of HDL-2 and small HDL particles. It is well documented that reduced HDL-C levels are associated with an increased risk of CHD. A number of functions of HDL particles may contribute to direct cardio-protective effects including promotion of cellular cholesterol efflux and direct anti-oxidative and anti-inflammatory properties [20].

Our study results show more number of male and female diabetic patients with uncontrolled diabetes had increased TC, TG and LDL-C levels. Female patients demonstrated non-significant decrease in HDL-C levels. The glycemic status worsens in females when compared to males.

Hyperlipidemia in females may be attributed to the effect of sex hormones on body fat distribution, leading to difference in altered lipoproteins [21]. Diabetic females are subjected to more adverse changes in coagulation, vascular function than diabetic men [1]. This explains why diabetes eliminated or attenuates a women's protection against IHD [1,22].

We observed the severity of dyslipidemia increases in patients with higher HbA1C value. Similar results were observed by Khan et al and Selvin et al who reported that elevated HbA1C and dyslipidemia are independent risk factors of CVD, diabetic patients with elevated HbA1C and dyslipidemia can be considered as a very high risk group for CVD [1,3]. Improving glycemic control can substantially reduce the risk of cardiovascular events in diabetics. It has been estimated that reducing the HbA1C levels by 0.2% could lower the mortality by 10% [3].

There was a slight increase in HbA1C in patient age 51-70 years as compared to patients aged <50 years. Older patients (>70 years) had HbA1C levels similar to the younger ones. However, this difference in HbA1C was not statistically significant among these age categories [1,24].

We also observed no significant correlation between age and lipid profile. This goes in hand with study results of Kalofoutis C et al but contradicts with the results of Khan et al who found a significant decrease in various lipid components including TC, TG and LDL-C with advancing age [1,24].

The above discussion clearly indicates the clinical significance of various lipid parameters in diabetic patients leading to various complications.

Conclusion

We conclude, HbA1C predicts dyslipidemia. This highlights the usefulness of HbA1C as dual biomarker (glycemic control and lipidemic state) for screening high risk diabetic patients. The better the glycemic control as reflected by HbA1C the better would be the lipidemic state. Achieving the target in HbA1C (< 6%) will contribute in improving the lipid state, and hence may lessen the diabetic complications in type 2 DM.

Conflicts of Interest: None

References

1.   Khan HA. Clinical significance of HbA1c as a marker of circulating lipids in male and female type 2 diabetic

patients. Actadiabetologica. 2007 Dec 1; 44(4):193-200.

2. Shankarprasad DS, Gundalli S, Kashinakunti SV, Sunitha P. Lipid profile in Diabetes Mellitus. Indian Journal of Pathology and Oncology. 2015; 2(4):290-4.

3. Selvin E,Coresh J, Golden SH et al Glycemic control and coronary heart disease risk in persons with and without diabetes: the atherosclerosis risk in communities study. Arch Intern Med. 2005; 165: 1910–1916.

4. Meenu J, Jayendrasinh MJ, Neeta M. Correlation between HbA1c values and lipid profile in type 2 diabetes mellitus. TC (mg/dl). 2013; 147:35-40.

5. Gligor R et al. Relationship between glycosylated hemoglobin and lipid metabolism in patients with type 2 diabetes. StudiaUniversitatis "VasileGoldiᵒ", Seriaᵃtiinþele Vieþii. 2011; 21(2):313-318.

6. Giansanti R, Rabini RA, Romagnoli F et al Coronary heart disease, type 2 diabetes mellitus and cardiovascular disease risk factors: a study on a middle-aged and elderly population. Arch GerontolGeriatr. 1999; 29:175–182.

7. Ravipati G, Aronow WS, Ahn C et al Association of hemoglobin A1c level with the severity of coronary artery disease in patients with diabetes mellitus. Am J Cardiol. 2006; 97:968–969.

8. Zhe Yan, Liu Y, Huang H. Association of glycosylated hemoglobin level with lipid ratio and individual lipids in type 2 diabetic patients. Asian Pacific Journal of Tropical Medicine 2012; 469-471.

9. Khaw KT, Wareham N, Bingham S, Luben R, Welch A and Day N. Association of hemoglobin A1c with cardiovascular disease and mortality in adults: the European Prospective Investigation into Cancer in Norfolk. Ann Intern Med. 2004; 141: 413-420.

10. Samatha P, Venkateswarlu M, Siva Prabodh V. Lipid profile levels in type 2 diabetes mellitus from the tribal population of Adilabad in Andhra Pradesh, India. Journal of clinical and Diagnostic Research. 2012 May; 6(4):590-2.

11. Sicree R, Shaw J, Zimmet P. Diabetes and impaired glucose tolerance. In: Gan D, editor. Diabetes Atlas. International Diabetes Federation. 3rd ed. Belgium: International Diabetes Federation 2006; 15:103.

12. Petitti DB, Imperatore G, Palla SL, Daniels SR, Dolan LM,Kershnar AK et al. Serum lipids and glucose control: the search for Diabetes in Youth study. Arch PediatrAdolesc Med. 2007; 161:159–165.

13. Rosediani M, Azidah AK, Mafauzy M. Correlation between fasting plasma glucose, post prandial glucose and Glycated haemoglobin and fructosamine. Med J Malaysia. 2006; 61:67–71.

14. Ito C, Maeda R, Ishida S, Sasaki H, Harada H. Correlation among fasting plasma glucose, two-hour plasma glucose levels in OGTT and HbA1c. Diabetes Res ClinPract. 2000; 50:225-230.

15. Önal ZE, Atasayan V, Akici N, Gürbüz T, Nuhoðlu Ç. The Relationship between Glycosylated Haemoglobin (HbA1c) Levels and Serum Lipid Profiles in Insulin Resistant Children HK J Paediatr (new series). 2014; 19(1):22-7.

16. VinodMahato R, Gyawali P, Raut PP, Regmi P, Singh KP, Pandeya DR et al. Association between glycaemic control and serum lipid profile in type 2 diabetic patients: Glycated haemoglobin as a dual biomarker. Biomedical research. 2011.

17. Farbstein D, Levy AP. HDL dysfunction in diabetes: causes and possible treatments. Expert review of cardiovascular therapy. 2012 Mar 1; 10(3):353-61.

18. Adiels M, Olofsson SO, Taskinen MR, Boren J. Diabetic dyslipidaemia. CurrOpinLipidol. 2006; 17(3):238–246.

19. Verges B, Florentin E, Baillot-Rudoni S, et al. Rosuvastatin 20 mg restores normal HDL-apoA-I kinetics in Type 2 diabetes. J Lipid Res. 2009; 50 (6):1209–1215.

20. Krauss RM. Lipids and lipoproteins in patients with type 2 diabetes. Diabetes care. 2004 Jun 1; 27(6): 1496-504.

21. Sibley SD, Thomas W, de Boer I, Brunzell JD, Steffes MW. Gender and elevated albumin excretion in the Diabetes Control and Complications trial/ Epidemiology of Diabetes Interventions and Complications (DCCT/EDIC) cohort: role of central obesity. Am J Kidney Dis. 2006; 47:223–232.

22. Firdous S, Khan MZ. Comparison of patterns of lipid profile in type-2 diabetics and non-diabetics. Ann. King. Edward. Med. Coll. 2007; 3(1): 84-7.

23. Doruk H, Mas MR, Ateskan U, Isik AT, Saglam M, Kutlu M. The relationship between age ad carotid artery intimamedia thickness, hemoglobin A1c in nondiabetic, healthy geriatric population. Arch GerontolGeriatr. 2005; 41:113–119.

24. Kalofoutis C, Piperi C, Zisaki A, Singh J, Harris F, Phoenix D, et al. Differences in expression of cardiovascular risk factors among type 2 diabetes mellitus patients of different age. Ann N Y Acad Sci. 2006; 1084:166–177.