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Research Article

Diabetic Cardiovascular Autonomic Neuropathy: Insulin Resistance, Lipids and Simvastatin

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ABSTRACT

Background: Treatment of diabetic cardiac autonomic neuropathy (CAN) is a complex process, that includes: lifestyle modification; reducing of insulin resistance (IR); optimal glycemic control; management of diabetic dyslipidemia; antioxidants; vitamins; treatment of myocardial metabolic abnormalities; thrombosis and others. The aim of study was to investigate the effects of simvastatin on insulin resistance and blood lipid profile parameters in patients with type 2 diabetes mellitus (DM) and the definite stage of cardiac autonomic neuropathy.

Methods: The study involved 107 patients with type 2 DM among them 16 patients without CAN, 19 with subclinical stage of CAN and 72 with definite CAN. Median age of patients was 53.6±0.41 years, disease duration - 4.12±0.24 years and median glycated hemoglobin (HbA1c) - 7.01%±0.09%. The control group included 14 almost healthy people without DM. Patients with definite CAN were allocated into two treatment groups: 1st group - 22 patients received standard hypoglycemic therapy and simvastatin 20 mg/day; 2nd group - control (n = 15). The duration of the study was 3 mos. The concentrations of glucose, HbA1c, immunoreactive insulin (IRI) in the blood were determined. Lipid metabolism was assessed by the concentration of total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), triglycerides (TG) measurements. The IR index (HOMA-IR), atherogenic coefficient (AC), TG/HDL-C parameters, and TG-glucose (TyG) index were calculated.

Result: It was established that in patients with type 2 DM with subclinical stage of CAN there was statistically significant increase in the parameters of HbA1c, IRI, TC, TG and HOMA-IR, AC, TyG indices and decrease of TG/HDL-C and HDL-C compared to control; increase of IRI, TG, TG/HDL-C and TyG indices compared to patients with type 2 DM without CAN. The definite stage of CAN is characterized by an increase of HbA1c, IRI, TC, LDL-C levels and HOMA-IR, AC indices and a significant decrease in the concentration of HDL-C (compared to patients with subclinical CAN). As a result of our study, we found out that prescription of simvastatin to patients with definite stage of CAN was accompanied by a statistically significant decrease in the concentration of TC, LDL-C, TG and an increase in the content of HDL-C (compared to 2nd, control group).

Conclusion: Obtained results justify the appropriateness of statins prescription to patients with type 2 DM and the definite stage of CAN.

Introduction

It was estimated that there were 415 million people with diabetes mellitus (DM) aged 20-79 years in 2015, and the number was predicted to rise to 642 million by 2040 [1]. The development of cardiovascular autonomic neuropathy (CAN) is associated with the lesion of the autonomic nervous system, and maybe accompanied by coronary vessels ischaemia, arrhythmias, 'silent' myocardial infarction, severe orthostatic hypotension (OH) and sudden death syndrome [2-4]. Cardiovascular autonomic neuropathy treatment is a complex process, that includes:

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lifestyle modification; reducing of insulin resistance (IR); optimal glycemic control; management of diabetic dyslipidemia (DLP); antioxidants; vitamins; treatment of myocardial metabolic abnormalities; thrombosis; management of OH; symptomatic treatment of concomitant diseases and others [5, 6].

The aim of this study was to investigate the effects of simvastatin on blood lipid profile, triglyceride-glucose index and insulin resistance parameters in patients with type 2 diabetes mellitus and the definite stage of the cardiovascular autonomic neuropathy.

Materials and Methods

I Study Population

The study involved 107 patients with type 2 DM among them 16 patients without CAN, 19 with subclinical stage of CAN and 72 with definite CAN. Median age of patients was 53.6±0.41 years, disease duration - 4.12±0.24 years and median glycated hemoglobin (HbA1c) - 7.01%±0.09%. The control group contained 14 almost healthy people without DM, that could be compared with the treatment group by demographic characteristics. CAN was diagnosed according to previously proposed criteria [2]. The work was done according to the principles of the Helsinki Declaration II and was approved by the Medical Ethics Committee of Danylo Halytsky Lviv National Medical University. All participants signed informed consent prior to their inclusion into the study. Patients with definite CAN were allocated into two treatment groups: 1st group- 22 patients received standard hypoglycemic therapy and simvastatin 20 mg/day; 2nd group - control (n = 15). The duration of the study was 3 months.

II Biochemical Methods

The concentration of glucose in the blood was determined by the glucose oxidase method while HbA1c level was assessed by using a highly sensitive method of ion-exchange liquid chromatography with D-10 analyzer and BIO-RAD reagents (United States). Determination of immunoreactive insulin (IRI) was performed using commercial kits from Immunotech insulin immunoradiometric assay reagents (Czech Republic). Lipid metabolism was assessed by the concentration of total cholesterol (TC), triglyceride (TG), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C); atherogenic coefficient (AC) and TG/HDL-C parameters. The TG-glucose (TyG) index was calculated by the Ln [fasting triglyceride (mg/dL) x fasting glucose (mg/dL)/2] [7]. The lipid fractions were determined by using HUMAN reagents (Germany) for the analyser HUMANLAYZER 2000. Homeostasis model assessment (HOMA) IR index (HOMA-IR) was calculated according to the formula: fasting IRI (mcIU/mL) x fasting glucose (mmol/L)/22.5 [8].

III ECG Recordings

Resting 12-lead surface electrocardiography (ECG) with a paper speed of 25 mm/s and a signal size of 10 mm/mV was recorded in the morning period. We performed resting ECG analysis included measurement of the following parameters: heart rhythm, heart rate, conduction intervals, and Holter-ECG [(ECG 'EC-3H' ('Labtech', Hungary)] analysis included measurement of 24 hours ECG, circadian indexes and heart rate variability parameters.

IV Statistical Analysis

Statistical analysis was based on the variational method using a statistical parametric t-test, nonparametric Wilcoxon t-test, and Fisher's Pearson correlation coefficient. Data are presented as mean ± standard error of the mean (SEM). All tests were performed using the ANOVA (Microcal Origin v. 8.0) software. Statistical significance was set at $p < 0.05$.

Results

As can be seen from the results, in patients with type 2 DM with subclinical stage of CAN, compared with the results obtained in the group of patients with type 2 DM without verified CAN, there was statistically significant increase in the content of IRI (+24.44%) in the absence of statistically significant changes in other parameters (Table 1).

Table 1: The IRI and HOMA-IR parameters in patients with type 2 DM and CAN (M±m).

Parameters	Patients with type 2 DM (n = 107)			
	Without CAN [group 1 (n = 16)]	Subclinical CAN [group 2 (n = 19)]	Definite CAN [group 3 (n = 72)]	Group 4 [control group (n = 14)]
HbA1c, %	6.58±0.16*	6.74±0.19†	7.18±0.11§**††	5.44±0.17
IRI, мcIU/L	16.12±1.15*	20.06±1.3†	27.31±1.1§♣**††	11.57±0.62
HOMA-IR	4.36±0.47*	5.73±0.6†	8.35±0.49§**††	2.45±0.14

IRI: Immunoreactive Insulin; HOMA-IR: Homeostasis Model Assessment Insulin Resistance; DM: Diabetes Mellitus; CAN: Cardiovascular Autonomic Neuropathy; HbA1c: glycated HbA1c.

The results are presented as mean ± standard error of the mean (SEM). Statistical significance was set at $p < 0.05$. * - significance of differences between type 2 DM without CAN and control; † - subclinical CAN and control; § - definite stage of CAN and control; ♣ - type 2 DM without CAN and subclinical CAN; ** - type 2 DM without CAN and definite stage of CAN; †† - type 2 DM with subclinical and definite stage of CAN.

The definite stage of CAN in patients with type 2 DM is characterized by an increase of IRI concentration (+136.04%) compared to control (p < 0.001); +69.42% compared to patients without CAN (p < 0.001); +36.14% compared to patients with subclinical CAN (p < 0.001), and HOMA-IR (+240.82% compared to control, p < 0.001); (+91.51% compared to patients without CAN, p < 0.01); +45.72% compared to patients with subclinical CAN. Therefore, the most statistically significant hyperinsulinemia (determined according to the IRI concentration) as well as IR (HOMA-IR) were verified in patients with type 2 DM and definite stage of CAN. The results of lipids concentrations, as well as atherogenic indices are presented in (Table 2).

Table 2: Lipid metabolism parameters and atherogenic indices in patients with type 2 DM with CAN (M±m).

Parameters	Patients with type 2 DM (n = 107)			Group 4 [control group (n = 14)]
	Without CAN [group 1 (n = 16)]	Subclinical CAN [group 2 (n = 19)]	Definite CAN [group 3 (n = 72)]	
TC, mmol/L	4.82±0.26	5.21±0.42†	6.27±0.11§**††	4.28±0.22
LDL-C, mmol/L	2.99±0.25*	3.08±0.34	4.33±0.1§**††	2.37±0.15
HDL-C, mmol/L	1.06±0.07*	0.91±0.05†	0.79±0.02§**††	1.26±0.06
TG, mmol/L	1.67±0.12*	2.65±0.35†	2.56±0.11§♣**††	1.26±0.15
AC	3.97±0.56*	5.34±0.82†	7.24±0.26§**††	2.45±0.13
TG/HDL-C	1.77±0.24*	3.38±0.6†	3.42±0.17§**	1.04±0.14
TyG index	8.89±0.08*	9.29±0.14†	9.41±0.05§**	8.37±0.12

DM: Diabetes Mellitus; CAN: Cardiovascular Autonomic Neuropathy; TC: Total Cholesterol; LDL-C: Low-Density Lipoprotein Cholesterol; HDL-C: High-Density Lipoprotein Cholesterol; AC: Atherogenic Coefficient; TG: Triglyceride; TG/HDL-C: Triglyceride/High-Density Lipoprotein Cholesterol Ratio; TyG: Triglyceride-Glucose Index.

The results are presented as mean ± standard error of the mean (SEM). Statistical significance was set at $p < 0.05$. * - significance of differences between type 2 DM without CAN and control; † - subclinical CAN and control; § - definite stage of CAN and control; ♣ - type 2 DM without CAN and subclinical CAN; ** - type 2 DM without CAN and definite stage of CAN; †† - type 2 DM with subclinical and definite stage of CAN.

As can be seen from the obtained results in patients with type 2 DM and subclinical CAN was a statistically significant increase of TG level (+110.32% compared to control group, $p < 0.01$); +58.68% compared to patients without CAN ($p < 0.05$); decrease in the concentration of HDL-C (-27.78%; compared to control group, $p < 0.001$) against the background of the absence of statistically significant changes in other parameters (Table 2). The definite stage of CAN is characterized by a further increase in the content of TC (+46.49% compared to the control group, $p < 0,001$); (+30.08% - to patients with type 2 DM without verified CAN, $p < 0,01$); (+ 20.34% to subclinical stage, $p < 0.001$); TG (approximately 2.03 times, $p < 0.001$); (+53.29% - for patients with type 2 DM without CAN, $p < 0.001$); LDL-C (+82.7% - to control, $p < 0.001$); (+44.82% - to patients with type 2 DM without CAN, $p < 0.01$); (+40.58% - to subclinical stage, $p < 0.001$), and a significant decrease in the concentration of HDL-C (-37.3% - to the control group, $p < 0.001$); (-25.47% - to patients with type 2 DM without CAN, $p < 0.001$), and (-13.19 % to subclinical stage, $p < 0.01$).

Changes in the values of some metabolism indicators in patients with type 2 DM with definite CAN after 3 months treatment with simvastatin are presented in the (Table 3).

Table 3: Changes in the values of some metabolism indicators in patients with type 2 DM and definite stage of CAN after 3 months of simvastatin treatment (Δ%, mean ± SEM).

Parameters	Simvastatin [group 1 (n = 22)]			Group 2 [control group (n = 15)]		
	Before treatment	After treatment	% change	Before treatment	After treatment	% change
TC	6.18±0.29	4.81±0.23	-21.83±1.55**	6.59±0.18	6.13±0.15	-6.73±1.09
LDL-C	4.27±0.27	2.72±0.14	-34.23±2.55**	4.59±0.16	4.25±0.17	-8.27±1.44
HDL-C	0.77±0.03	0.92±0.06	+12.14±2.32*	0.84±0.03	0.87±0.03	+4.09±0.97
TG	2.5±0.27	1.83±0.18	-22.57±1.2*	2.52±0.12	2.31±0.11	-8.28±1.17
IRI	29.53±1.99	26.16±1.6	-8.91±4.21	27.79±2.13	26.01±2.25	-6.43±3.11
HOMA-IR	8.99±0.94	7.43±0.6	-8.86±6.84	9.04±0.99	8.46±0.99	-6.12±4.06
TyG index	9.27±0.14	8.96±0.13	-3.29±0.32	9.53±0.06	9.44±0.05	-0.89±0.39

DM: Diabetes Mellitus; CAN: Cardiovascular Autonomic Neuropathy; TC: Total Cholesterol; LDL-C: Low-Density Lipoprotein Cholesterol; HDL-C: High-Density Lipoprotein Cholesterol; TG: Triglyceride; IRI: Immunoreactive Insulin; HOMA-IR: Homeostasis Model Assessment IR; TyG - Triglyceride-Glucose Index.

The results are presented as Δ%, mean ± standard error of the mean (SEM). Statistical significance was set at $p < 0.05$. * - $p < 0.05$; ** - $p < 0.001$. We found out that the HbA1c of patients with type 2 DM and definite CAN was not statistically significant influenced by the treatment ($p > 0.05$).

As a result of our study, we found out that the use of simvastatin was accompanied by a statistically significant decrease in the concentration of TC, LDL-C, TG, IRI and an increase in the content of HDL-C (compared with the control group) (Table 3).

Discussion

TyG index, the product of fasting glucose and TG in the blood, has been proposed as a simple method for determining IR in healthy subjects [9, 10]. The use of the HOMA-IR homeostasis model, the insulin suppression test, and the hyperinsulinemic -euglycemic clamp suggested that the TyG index correlates with IR [9, 11-13]. TyG indexes are reported to be elevated in type 2 DM patients compared to parameters in patients with prediabetes [13-16]. Despite its clinical convenience, the accuracy of the TyG index may be limited. For example, although TG has been suggested to be an independent risk factor for DM, the TG level is strongly influenced by a high carbohydrate diet, alcohol consumption, and lifestyles such as exercise. However, due to its convenience and

relatively low cost, TG measurements are widely conducted in clinical practice [13].

The 3-hydroxy-3-methylglutaryl-coenzyme (HMG-CoA) reductase inhibitors (lovastatin, pravastatin, fluvastatin, simvastatin, atorvastatin, rosuvastatin, pitavastatin). By inhibiting HMG-CoA reductase, statins block the pathway for synthesizing cholesterol in the liver. The reduction in cholesterol level induces an increased expression of the low-density lipoprotein receptor, which results in decreased concentration of LDL-C and other apolipoprotein B-containing lipoproteins [17]. Secondary prevention statin studies such as MRC/BHF Heart Protection Study (HPS) showed significant risk reduction among individuals with diabetes mellitus. Based on this, the primary prevention of cardiovascular diseases (CVD) with atorvastatin in type 2 DM in the Collaborative Atorvastatin Diabetes Study (CARDS) was designed to assess the effects of aggressive lipid-lowering on the primary prevention of atherosclerotic CVD in individuals with type 2 DM. In individuals with average or mildly elevated LDL-C at baseline (mean 117 mg/dL), an LDL-C reduction to a mean of 82 mg/dL was accompanied by a 37% reduction in major cardiovascular events compared with placebo. CARDS, which originally planned a mean follow-up of 4 years, was terminated 2 years early because of the significant benefit achieved in the statin group [18].

In summary, statins: decrease LDL-C $\approx$ 21-55% by competitively inhibiting rate-limiting step of cholesterol synthesis in the liver, leading to upregulation of hepatic LDL receptors (primarily); decrease TG $\approx$ 6-30% and increase HDL-C $\approx$ 2-10% [18].

Thus, hyperinsulinemia/IR, lipid metabolism disorders are accompanied by an increase the concentration of TG, TC and LDL-C, decrease HDL-C in patients with type 2 DM, in particular, with CAN [6, 19]. Therefore, the appointment of statins is necessary in the treatment of DLP in patients with type 2 DM with definite stage of cardiovascular autonomic neuropathy.

Conclusion

Development of hyperinsulinemia and insulin resistance among patients with type 2 DM and CAN are accompanied by atherogenic changes in lipid profile namely by an increase in TC, TG, LDL-C, AC, TyG index and decrease in HDL-C. The most pronounced atherogenic changes observed among patients with definite stage of cardiovascular autonomic neuropathy.

Obtained results could witness about the essential importance of hyperinsulinemia, IR and DLP in the pathogenesis of the cardiovascular autonomic neuropathy. Prescription of simvastatin to patients with definite stage of CAN contributed to a statistically significant decrease in the concentration of TC, LDL-C, TG, and an increase in the content of HDL-C (compared to the control group). Obtained results justify the appropriateness of statins prescription to patients with type 2 DM and definite stage of cardiovascular autonomic neuropathy.

Conflicts of Interest

None.

REFERENCES

1. K Ogurtsova, J D da Rocha Fernandes, Y Huang, U Linnenkamp, L Guariguata et al. (2017) IDF Diabetes Atlas: Global estimates for the prevalence of diabetes for 2015 and 2040. *Diabetes Res Clin Pract* 128: 40-50. [Crossref]

2. Vincenza Spallone, Dan Ziegler, Roy Freeman, Luciano Bernardi, Simona Frontoni, Rodica Pop Busui et al. (2011) Cardiovascular autonomic neuropathy in diabetes: clinical impact, assessment, diagnosis, and management. *Diabetes Metab Res Rev* 27: 639-653. [Crossref]

3. Victoria A Serhiyenko, Alexandr A Serhiyenko (2015) Diabetic cardiac autonomic neuropathy: Do we have any treatment perspectives? *World J Diabetes* 6: 245-258. [Crossref]

4. Standards of Medical Care in Diabetes-2017: Summary of Revisions (2017) *Diabetes Care* 40: S4-S5. [Crossref]

5. Nikhil Tandon, Mohammed K Ali, K M Venkat Narayan (2012) Pharmacologic prevention of microvascular and macrovascular complications in diabetes mellitus: implications of the results of recent clinical trials in type 2 diabetes. *Am J Cardiovasc Drugs* 12: 7-22. [Crossref]

6. Victoria A Serhiyenko, Alexandr A Serhiyenko (2018) Cardiac autonomic neuropathy: Risk factors, diagnosis and treatment. *World J Diabetes* 9: 1-24. [Crossref]

7. Luis E Simental Mendía, Martha Rodríguez Morán, Fernando Guerrero Romero (2008) The product of fasting glucose and triglycerides as surrogate for identifying insulin resistance in apparently healthy subjects. *Metab Syndr Relat Disord* 6: 299-304. [Crossref]

8. D R Matthews, J P Hosker, A S Rudenski, B A Naylor, D F Treacher et al. (1985) Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 28: 412-419. [Crossref]

9. Fahim Abbasi, Gerald M Reaven (2011) Comparison of two methods using plasma triglyceride concentration as a surrogate estimate of insulin action in nondiabetic subjects: triglycerides x glucose versus triglyceride/high-density lipoprotein cholesterol. *Metabolism* 60: 1673-1676. [Crossref]

10. Tingting Du, Gang Yuan, Muxun Zhang, Xinrong Zhou, Xingxing Sun et al. (2014) Clinical usefulness of lipid ratios, visceral adiposity indicators, and the triglycerides and glucose index as risk markers of insulin resistance. *Cardiovasc Diabetol* 13: 146. [Crossref]

11. Fernando Guerrero Romero, Luis E Simental Mendía, Manuel González Ortiz, Esperanza Martínez Abundis, María G Ramos Zavala et al. (2010) The product of triglycerides and glucose, a simple measure of insulin sensitivity. Comparison with the euglycemic-hyperinsulinemic clamp. *J Clin Endocrinol Metab* 95: 3347-3351. [Crossref]

12. Ana Carolina Junqueira Vasques, Fernanda Satake Novaes, Maria da Saúde de Oliveira, José Roberto Matos Souza, Ademar Yamanaka et

al. (2011) TyG index performs better than HOMA in a Brazilian population: a hyperglycemic clamp validated study. *Diabetes Res Clin Pract* 93: e98-e100. [Crossref]

13. Ho Jin Kim, Jun Sung Moon, Il Rae Park, Joong Hee Kim, Ji Sung Yoon et al. (2017) A novel Index using soluble CD36 is associated with the prevalence of type 2 diabetes mellitus: comparison study with triglyceride-glucose index. *Endocrinol Metab* 32: 375-382. [Crossref]

14. Mohsen Janghorbani, Siedeh Zinab Almasi, Masoud Amini (2015) The product of triglycerides and glucose in comparison with fasting plasma glucose did not improve diabetes prediction. *Acta Diabetol* 52: 781-788. [Crossref]

15. Seung Hwan Lee, Hyuk Sang Kwon, Yong Moon Park, Hee Sung Ha, Seung Hee Jeong et al. (2015) Predicting the development of diabetes using the product of triglycerides and glucose: the Chungju Metabolic Disease Cohort (CMC) study. *PLoS One* 9: e90430. [Crossref]

16. David Navarro González, Laura Sánchez Íñigo, Juan Pastrana Delgado, Alejandro Fernández Montero, J Alfredo Martinez (2016) Triglyceride-glucose index (TyG index) in comparison with fasting plasma glucose improved diabetes prediction in patients with normal fasting glucose: the Vascular-Metabolic CUN cohort. *Prev Med* 86: 99-105. [Crossref]

17. Alberico L Catapano, Ian Graham, Guy De Backer, Olov Wiklund, M John Chapman et al. (2016) 2016 ESC/EAS Guidelines for the Management of Dyslipidaemias. *Eur Heart J* 37: 2999-3058. [Crossref]

18. Paul S Jellinger, Yehuda Handelsman, Paul D Rosenblit, Zachary T Bloomgarden, Vivian A Fonseca et al. (2017) American Association of Clinical Endocrinologists and American College of Endocrinology Guidelines for management of dyslipidemia and prevention of cardiovascular disease - executive summary. *Endocr Pract* 23: 1-87. [Crossref]

19. Serhiyenko VA, Serhiyenko LM, Ajmi S, Serhiyenko AA (2019) Diabetic cardiac autonomic neuropathy: Effects of statins and omega-3 polyunsaturated fatty acids on lipid profile and insulin resistance parameters. *Clin Trials J* 5: 1-6.