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The impact of insulin induced lipohypertrophy on carotid intima-media thickness in patients with type 2 diabetes mellitus

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Abstract

Lipohypertrophy (LH) is a major localized complication of insulin therapy. We aimed to investigate the association between insulin-induced LH and carotid intima-media thickness (CIMT) in patients with type 2 diabetes mellitus (DM). A total of 75 patients with DM treated with insulin were included in this study. The insulin injection sites of the patients were evaluated by inspection and palpation and then radiologically with ultrasound. The CIMT of the patients was evaluated using ultrasonography. According to the guideline recommendation, the CIMT cutoff value was taken as 0.9 mm, and the patients were categorized into 2 groups according to the CIMT value and evaluated statistically. The presence of LH (CI: 1.379–30.000; OR = 6.432; $P < .05$), age (CI: 1.036–1.149; OR = 1091; $P < .05$), BMI (CI: 1.003–1.262; OR = 1.125; $P < .05$) and duration of DM (CI: 1.001–1.300; OR = 1.141; $P < .05$) were independent risk factors for high-CIMT in patients with DM. The most interesting result of this study was that the presence of LH was an independent risk factor for increased CIMT. According to this result, we think that LH may increase the risk of cardiovascular disease as well as being a complication that disrupts the blood glucose regulation of patients with DM and increases the cost of treatment.

Abbreviations: BMI = body mass index, CIMT = carotid intima-media thickness, CVD = cardiovascular disease, DM = diabetes mellitus, LH = lipohypertrophy, USG = ultrasonography.

Keywords: CIMT, diabetes mellitus, insulin injection, lipid profile, lipohypertrophy

1. Introduction

Cardiovascular diseases (CVD), including coronary artery, cerebrovascular, and peripheral artery diseases, are the most important causes of morbidity and mortality due to diabetes mellitus (DM).[1] The estimated annual amount spent on diabetes-related cardiovascular complications was $37.3 billion.[2] In patients with DM, in addition to preventing dyslipidemia and hypertension, smoking cessation, healthy diet, and physical activity, the most important factor in preventing CVD is glycemic control.[3] In the Diabetes Control and Complications Trial, at milestone in the management of patients with DM, glycemic control has been shown to reduce microvascular complications.[4] The importance of glycemic control in DM has been mentioned in many international guidelines.[5,6] Many oral antidiabetic agents and injectable treatment options are available to achieve glycemic control. Pancreatic beta cell destruction is known to occur in type 1 DM and, the only treatment option is intensive insulin therapy. Many patients with type 2 DM eventually remain insulinopenic and require insulin therapy.[7] Although insulin therapy is a life-saving treatment for patients with DM, it has some side effects. The most common side effects of insulin therapy are hypoglycemia, weight gain, and fluid retention.[8] Lipohypertrophy (LH) is a leading localized complication of insulin therapy.[9] In addition to the increase in daily insulin needs, and treatment costs, the frequency of hypoglycemia is 6 times higher and glycemic fluctuations are 7 times higher in patients with LH.[10] The etiopathogenesis of LH is thought to be due to the lipogenic effect of insulin. However, the presence of macrophages and T lymphocytes in pathological examinations of tissue with LH suggests that there may be an inflammatory response to insulin.[11]

Carotid intima-media thickness (CIMT) measurement is one of the methods used to evaluate subclinical atherosclerosis.[12] CIMT is not only a predictor of CVD but is also associated with atherosclerosis risk factors.[13–15] As both LH and CVD develop on an inflammatory basis, we evaluated the relationship between these 2 conditions in this study. For this purpose, we used CIMT, a noninvasive method. As far

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The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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as we know, this study is the first study of this subject in the literature.

2. Methods

Our study was approved by the Ethics Committee of Sutcu Imam University Faculty of Medicine dated 12.01.2022 and numbered 02. All clinical investigations were conducted in accordance with the principles of the Declaration of Helsinki.

Patients with type 2 DM who applied to the endocrinology outpatient clinic of our hospital between January 2022 and December 2022 were evaluated for inclusion in the study. Written informed consent was obtained from all eligible patients and included in the study.

2.1. Inclusion criteria

•Patients aged 18 to 65 years.
•Patients who receive 4 or more insulin injections per day for at least 1 year.

2.2. Exclusion criteria

- Patients with skin lesions such as dermatitis, infection at the insulin injection sites.
- Patients with AIDS.
- Patients with known history of coronary artery disease, cerebrovascular disease, peripheral artery disease.
- Patients with hypertension or dyslipidemia.
- Patients using statins, angiotensin-converting enzyme inhibitors or angiotensin II receptor blockers.
- Tobacco use.
- Patients with chronic renal failure or chronic liver disease.
- Patients with malignancy.
- Pregnancy.

The duration of DM, insulin doses used, and time of starting insulin therapy were recorded. During the examination of the patients, height, weight, and blood pressure were measured and recorded. Body mass index (BMI) was calculated by dividing the weight in kilograms by height in meters squared. Blood samples were collected from the patients for HbA1c, total cholesterol, HDL-c, LDL-c, triglyceride, fasting blood glucose, fasting C-peptide, and creatinine levels for routine DM follow-up.

2.3. Ultrasound technique

Insulin injection sites (abdomen, both arms, and legs) of the patients included in the study were evaluated first by inspection and palpation and then radiologically with an B-mode ultrasound (USG) device in the endocrinology outpatient clinic (Biosound Esaote MyLab 60 USG System, 6–18 MHz, linear probe). Patients with LH were divided into 3 groups: diffuse hyperechoic, nodular hyperechoic, and hypoechoic LH.[16] Normal areas near the affected areas were considered as references. Then, with the linear probe of the same USG device, carotid artery imaging was performed while the patient was lying in the supine position, at an angle of approximately 20° to the opposite side of the neck. CIMT was determined by the average result of 3 measurement of the thickness of the tunica intima media on the right common carotid artery and left common carotid artery, which appeared as a double-line sign longitudinally on the walls of the common carotid artery. USG measurements were performed by an experienced endocrinologist in USG.

According to the European Society of Hypertension/European Society of Cardiology guideline recommendation, the CIMT cutoff value, which is accepted as an indicator of subclinical organ damage, was taken as 0.9 mm,[17] and the patients were categorized into 2 groups according to the CIMT value as high-CIMT and low-CIMT and evaluated statistically.

2.4. Statistical analysis

Statistical analyzes were performed using SPSS version 22.0 (SPSS Inc., Chicago, IL). Mean, standard deviation, and median (interquartile range) were used to describe continuous variables, and count (n) and percent were used for categorical variables. Chi-square test, Fisher exact test, Mann–Whitney test, and Student test were used for data analysis as appropriate. Correlation coefficients of numeric variables that did not fit at least one normal distribution measure were calculated using Pearson test and statistical significance using Spearman test. Multivariate logistic regression models were performed to determine independent predictors adjusted for pre-established baseline covariates. Variables with $P < .1$ in univariate analysis were included in multivariate analysis. Receiver operating characteristic curve analysis was used to determine the sensitivity and specificity of the predictive value of the CIMT for LH. The optimal cut-off value of CIMT was derived from Youden index. A two-tailed value < 0.05 was considered statistically significant.

3. Results

The characteristics of patients included this prospective study are shown in Table 1. Of the 75 included patients, 34 (45.3%) were male. The mean values at baseline were age 48.73 ± 16.03 years. The low-CIMT group included 47 patients and the high-CIMT group included 28 patients. The groups had similar characteristics in terms of sex distribution ($P > .05$). When the groups were stratified by age, the high-CIMT group consisted of older individuals ($P < .05$). The median BMI ($P < .05$), time to insulin initiation, total daily insulin dose ($P < .05$), and mean DM duration were higher in the high-CIMT group and no differences were found in FBG, HbA1c, C-peptide, triglycerides, LDL-c, HDL-c, and creatinine levels ($P > .05$). When the groups were analyzed according to their LH status, the incidence of LH was higher in the high-CIMT group ($P < .05$). Although nodular hyperechoic type LH (51.3%) was the most common in subgroup analyzes for LH, the LH distribution of the subgroups was similar ($P > .05$).

Correlations between CIMT levels, clinical parameters, and blood markers were examined using Spearman and Pearson analyses. The results showed that the CIMT levels were positively correlated with age ($R = 0.613$, $P < .001$), BMI ($R = 0.381$, $P < .001$), and diabetes duration ($R = 0.412$, $P < .001$). No statistically significant association was found between CIMD and C-peptide levels. ($R = 0.220$, $P > .05$) (Table 2).

To investigate potential risk factors for high-CIMT in patients with DM, univariate and multivariate regression analyzes were performed using the 7 potential risk factors (Table 3). Univariate analysis showed that the presence of LH, BMI, time to insulin initiation, total daily insulin dose, duration of DM (years), and C-peptide levels were the 6 potential risk factors for patients with DM (Table 3). All variables with a value < 0.1 in univariate analyses could be included in the multivariate analysis. The presence of LH (CI: 1.379–30.000; OR = 6.432; $P < .05$), age (CI: 1.036–1.149; OR = 1091; $P < .05$), BMI (CI: 1.003–1.262; OR = 1.125; $P < .05$) and duration of DM (CI: 1.001–1.300; OR = 1.141; $P < .05$) were independent risk factors for high-CIMT in patients with DM. As shown in Figure 1, baseline CIMT was a significant predictor of LH status with an area under the curve of 0.835, cutoff value of ≥0.77, Youden index = 0.57, sensitivity of 79%, and specificity of 78%.

Table 1

Baseline characteristics of patients according to carotid intima-media thickness.

Variables	Total (n = 75)	Low-CIMT (n = 47)	High-CIMT (n = 28)	*P*
Sex (male, %)	34 (45.3%)	19 (40.4%)	15 (53.6%)	.340
Age (years)	48.73 ± 16.03	41.06 ± 15.35	59.71 ± 11.93	**<.001**
BMI (kg/m^2)	30.5 (27–34)	30 (26–33)	32.75 (29–36)	**.001**
CIMT (cm)	0.79 ± 0.25	0.65 (0.50–0.97)	1.1 (0.75–1.1)	**–**
LH status (yes, %)	39 (52%)	16 (34%)	23 (82.1%)	**<.001**
LH subgroups	**–**	**–**	**–**	.435
Diffuse hyperechoic	15 (38.5%)	8 (50%)	7 (30.4%)	
Nodular hyperechoic	20 (51.3%)	7 (43.8%)	13 (56.5%)	
Hypoechoic	4 (10.3%)	1 (6.3%)	3 (13%)	
Duration of DM (years)	12.75 ± 7.28	10.51 ± 6.09	16.50 ± 7.66	**<.001**
Time to insulin initiation (years)	10 (5–15)	6 (2–12)	10 (8.25–15)	**.004**
Total daily insulin dose (U/day)	66 (50–94)	64 (48–88)	77 (54–98)	**.049**
FBG (mg/dL)	203 (157–250)	196 (144–250)	212 (179–270)	.255
HbA1c (%)	9 (7.6–10.5)	9 (7.4–10.4)	9.2 (8.2–10.5)	.126
C-peptide (ng/ mL)	1.2 (0.7–2.6)	1.9 (0.7–2.83)	1 (0.65–1.75)	.072
Triglycerides (mg/dL)	167 (111–233)	150 (110–228)	180 (127–251)	.384
LDL-c (mg/dL)	110 (88–140)	105 (88–138)	121 (88–165)	.261
HDL-c (mg/dL)	45 (40–52)	46 (40–54)	42 (40–49)	.167
Creatinine (mg/dL)	0.9 (0.7–1)	0.8 (0.7–1)	0.9 (0.72–1)	.254

Bold text shows that the values are less than <0.05.

ALT = alanine aminotransferase; BMI = body-mass index; CIMT = carotid intima-media thickness; DM =diabetes mellitus; FBG = fasting blood glucose; HbA1c = glycated haemoglobin; HDL-c = high-density lipoprotein; LDL-c = low-density lipoprotein; LH: lipohypertrophy.

Table 2

Correlations of carotid intima-media thickness with clinical parameters and blood markers.

Variables	Correlation r value	*P*-value
Age	0.613	<.001
BMI	0.381	<.001
Duration of DM	0.412	<.001
C-peptide	0.220	.058

Bold text shows that the values are less than <0.05.

BMI = body-mass index; DM = diabetes mellitus.

4. Discussion

In our study, we found that the CIMT increased significantly in patients with LH. LH is a common complication in patients with diabetes who are undergoing injection therapy. The prevalence of LH differs according to the examination methods used in the studies and ranges from 23% to 73.4%.[18,19] Beyond being a cosmetic problem, LH not only causes glycemic fluctuations, and increases the frequency of hypoglycemia, but also increases the cost of treatment by increasing the daily insulin dose requirement.[10] Recent studies have reported that glycemic fluctuations are strongly associated with CVD. Some studies have even shown that glycemic fluctuations increase the risk of CVD more than chronic hyperglycemia.[20,21] In a multicenter study by Gentile et al, insulin injection training was administered to patients with LH, who were followed up for CVD risk. The study showed that the incidence of LH decreased in patients and that the 10-year CVD risk score decreased significantly.[9] In this study, the HbA1c and lipid levels of the patients were normalized, and glycemic fluctuations decreased. It can be assumed that the reduction in CVD risk is due to the improvement of these metabolic parameters. However, in our study, LH was an independent risk factor for CVD. In addition to glycemic fluctuations, LH pathophysiology may increase the risk of CVD. The pathogenesis of insulin-dependent LH has not yet been fully elucidated. The most important reason for this is thought to be the local reaction of adipocytes to insulin injection.[22] In addition, some studies have suggested that immunological and inflammatory responses also play a role in the pathophysiology of LH.[11,22] Inflammation is known to play a prominent role in the pathogenesis of CVD, and inflammatory markers, such as high-sensitive C-reactive protein are significantly increased in patients with CVD.[23,24] For these reasons, it can be considered that the increase in CIMT in patients with LH may be due to a common pathophysiology. Another association between LH and increased CIMT may be due to hyperinsulinism. The daily dose of insulin administered to patients with LH is higher than that in LH-negative patients.[10] Hyperinsulinemia overstimulates the extracellular signal-regulated mitogen-activated protein kinase pathway, promoting the development and progression of atherosclerosis.[25] In our study, it was observed that patients with high CIMT were using much higher daily insulin doses, which was consistent with previous studies. We also observed that the duration of insulin use was longer in patients with high CIMT values. This result suggests that the duration of hyperinsulinism may have a negative effect on CIMT.

Bertuzzi et al, in their study of the ultrasonographic classification of LH, divided LH lesions into 2 classes: hypoechoic and hyperechoic. The authors considered that hypoechoic lesions are the first phase of LH, and such lesions are due to subcutaneous edema. It is thought that hypoechoic lesions may heal rapidly when the injection site is changed. Hyperechoic lesions are considered to be the second phase of LH, due to fibrosis caused by repetitive micro-trauma.[26] Although these lesions differ from each other pathologically, no significant difference was observed between the LH subgroups in terms of CIMT in our study.

In a review published by Inge et al, there was a significant relationship between CIMT and age in both healthy individuals and those with CVD disease.[27] The Tromsø study also reported that CIMT has a linear relationship with age, but age does not affect plaque formation like the traditional risk factors for CVD (blood pressure, cholesterol, and smoking).[28] In our study, age correlated with CIMT and was an independent risk factor for CIMT, which is consistent with the literature.

Obesity is a serious health problem not only for patients with DM but also worldwide. Between 39% and 49% of the world's population are overweight and obese.[29] According to the data

Table 3

Univariate and multivariate logistic regression models to assess which variables were independently associated with increased carotid intima-media thickness.

	Univariate odds ratios			Multivariate odds ratios		
Variables	**OR**	**95% CI**	***P***	**OR**	**95% CI**	***P***
Male	1.700	0.662–4.370	.270	–	–	–
Age (years)	1.091	1.047–1.137	**<.001**	1.091	1.036–1.149	**.001**
BMI (kg/m^2)	**1.154**	1.055–1.263	**.002**	1.125	1.003–1.262	**.044**
LH status (yes, %)	7.104	2.399–21.040	**<.001**	6.432	1.379–30.000	**.018**
Duration of DM (years)	1.139	1.051–1.234	**.002**	1.141	1.001–1.300	**.048**
Total daily insulin dose (U/day)	1.017	1.002–1.033	**.031**	1.006	0.985–1.028	.581
C-peptide (ng/mL)	0.684	0.465–1.005	**.053**	0.834	0.447–1.556	.569

Bold text shows that the values are less than <0.05.

BMI = body-mass index; CI = confidence interval; DM = diabetes mellitus; LH = lipohypertrophy; OR = odds ratio.

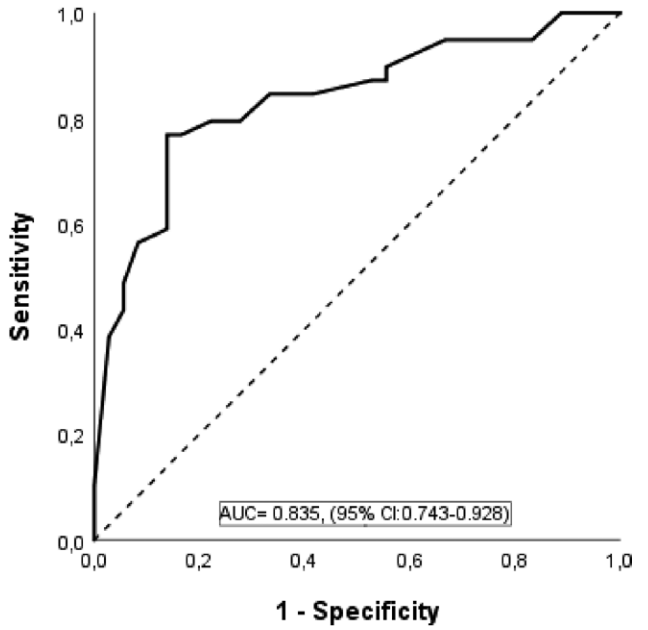

Figure 1. The predictive value of carotid intima-media thickness for lipohypertrophy status in patients with diabetes mellitus by ROC analysis. ROC = receiver operating characteristic.

of the Global Burden of Disease study, approximately 4 million deaths occurred due to increased BMI in 2015, and it was reported that the cause of two-thirds of these deaths was due to CVD.[30] In a study conducted by Pillay et al on 40,000 data, it was found that there was a linear and positive correlation between BMI and CIMT.[31] In our study, an increased BMI was found to be an independent risk factor for CIMT.

When we evaluated the relationship between sex and CIMT, we found no significant difference between the 2 genders in our study. This result is inconsistent with the literature. Studies have found that CIMT is generally higher in males than females.[32–34] In a study by Su et al, it was observed that CIMT is higher in men, but the advantage of CIMT decreases rapidly due to the increase in CVD risk factors in women after the age of 55.[35] The difference between the literature and our study may be due to the small sample size. In addition, since all subjects included in the study were patients with DM, the CIMT values depending on gender may have been higher in both groups.

DM has been considered equivalent to CVD in some studies.[36] Evans et al stated that the duration of DM, rather than its presence, may have a greater effect on CVD-related mortality.[37] In our study, the duration of DM was significantly longer in the patient group with high CIMT. This finding seems to be compatible with those of previous studies.[38,39] The increase in CIMT with the duration of diabetes is thought to be the result of increased oxidative stress, impaired endothelial function and monocyte adhesion to endothelial cells as a result of high postprandial glucose levels independent of HbA1c.[38]

When we evaluated previous studies on blood glucose regulation and CIMT, we found conflicting results. In a study by Kumar et al on patients with DM, a positive correlation was observed between fasting and postprandial blood glucose levels and CIMT, but no correlation was found between HbA1c level and CIMT. The authors considered that this result was due to the adequate glycemic control of the patient group and the small variation in HbA1c levels.[40] In another study conducted in China, no relationship was found between HbA1c levels and fasting, and postprandial blood glucose levels with CIMT. The authors stated that this result might depend on the design of the study, patient population, or antihypertensive and lipid-lowering treatments used by the patients.[41] In our study, no correlation was found between HbA1c levels, fasting blood glucose levels and CIMT. We observed that the variation in HbA1c level was small between the high and low CIMT groups in our study.

Previous studies have reported that C-peptide causes atherosclerosis in diabetic patients by stimulating smooth muscle cell proliferation.[42,43] A study conducted by Liu et al showed a positive correlation between postprandial C-peptide level and CIMT in diabetic patients.[44] In a study by Zhou et al, while there was no relationship between fasting C-peptide level and CIMT, a significant relationship was observed between postprandial C-peptide and CIMT.[41] In our study, we did not evaluate postprandial C-peptide levels. However, similar to the literature, no significant relationship was found between fasting C-peptide level and CIMT.

Previous studies have reported conflicting results regarding the relationship between lipid parameters and CIMT. In this study, we evaluated the relationship between CIMT and total cholesterol, LDL-c, HDL-c and triglycerides. We found no significant relationship between these 4 parameters and the CIMT. However, while evaluating this result, it should be noted that we selected the patient population from patients without dyslipidemia. In a study by Liu et al on patients with DM, similar results were found in our study, and no significant relationship was observed between lipid parameters and CIMT.[44] In this study, unlike our study, dyslipidemia was not considered an exclusion criterion. In the study of Zhou et al on patients with DM, only low HDL-c levels were found to be associated with increased CIMT and no significant correlation was found between the other 3 lipid parameters and CIMT.[41] As in our study, patients with CVD were not included, but this study was conducted with a large sample size of 1372 patients. In a study by Kota et al on patients with DM, a relationship was observed between high total cholesterol, LDL-c, and triglyceride levels and increased

CIMT, but no significant relationship was found between low HDL-c levels and CIMT.[45] This study was performed only in patients with both DM and ischemic cerebrovascular disease. As can be seen, the relationship between lipid parameters and CIMT may differ depending on the study design and the number of patients included in the studies.

Our study has some limitations. First, there was a difference in the number of patients in the low and high CIMT groups. We determined as exclusion criteria the diseases such as hypertension and dyslipidemia, which affect the CIMT value. Owing to the high prevalence of these comorbidities in patients with DM, the sample size of the study remained low. The second limitation was the cutoff value determined for CIMT. According to the European Society of Hypertension/European Society of Cardiology guideline recommendations, we used at CIMT cutoff value of 0.9 mm. However, studies have shown that CIMT may vary according to age, gender, CVD risk factors, and race.

5. Conclusions

The most interesting result of this study was that the presence of LH was an independent risk factor for increased CIMT. According to this result, we think that LH may increase the risk of CVD as well as being a complication that disrupts the blood glucose regulation of the patients with DM and increases the cost of treatment.

We need to work harder to reduce the frequency of LH in order to regulate blood glucose, reduce the cost of treatment, and prevent CVD, as we have shown in our study. The most effective way to achieve this is patient education.

Author contributions

Conceptualization: Cem Onur Kirac.
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References

[1] American Diabetes Association. 10. Cardiovascular disease and risk management: standards of medical care in diabetes-2021. Diabetes Care. 2021;44(Suppl 1):S125–150.
[2] American Diabetes Association. Economic costs of diabetes in the U.S. in 2017. Diabetes Care. 2018;41:917–28.
[3] Rosengren A, Dikaiou P. Cardiovascular outcomes in type 1 and type 2 diabetes. Diabetologia. 2023;66:425–37.
[4] Nathan DM, Cleary PA, Backlund JY, et al. Intensive diabetes treatment and cardiovascular disease in patients with type 1 diabetes. N Engl J Med. 2005;353:2643–53.
[5] Cosentino F, Grant PJ, Aboyans V, et al. 2019 ESC Guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the EASD. Eur Heart J. 2020;41:255–323.
[6] Davies MJ, Aroda VR, Collins BS, et al. Management of hyperglycemia in type 2 diabetes, 2022. A consensus report by the American Diabetes Association (ADA) and the European Association for the Study of Diabetes (EASD). Diabetes Care. 2022;45:2753–86.
[7] ElSayed NA, Aleppo G, Aroda VR, et al. 9. Pharmacologic approaches to glycemic treatment: standards of care in diabetes-2023. Diabetes Care. 2023;46(Suppl 1):S140–57.
[8] Gerstein HC, Miller ME, Byington RP, et al. Effects of intensive glucose lowering in type 2 diabetes. N Engl J Med. 2008;358:2545–59.
[9] Gentile S, Guarino G, Corte TD, et al. Insulin-induced skin lipohypertrophy in type 2 diabetes: a multicenter regional survey in Southern Italy. Diabetes Ther. 2020;11:2001–17.
[10] Blanco M, Hernández MT, Strauss KW, et al. Prevalence and risk factors of lipohypertrophy in insulin-injecting patients with diabetes. Diabetes Metab. 2013;39:445–53.
[11] Fujikura J, Fujimoto M, Yasue S, et al. Insulin-induced lipohypertrophy: report of a case with histopathology. Endocr J. 2005;52:623–8.
[12] Takase M, Nakaya N, Nakamura T, et al. Carotid intima media thickness and risk factor for atherosclerosis: Tohoku Medical Megabank Community-based cohort study [published online ahead of print February 11, 2023]. J Atheroscler Thromb. doi: 10.5551/jat.64039.
[13] O'Leary DH, Polak JF, Kronmal RA, et al. Carotid-artery intima and media thickness as a risk factor for myocardial infarction and stroke in older adults. Cardiovascular Health Study Collaborative Research Group. N Engl J Med. 1999;340:14–22.
[14] Rosvall M, Persson M, Östling G, et al. Risk factors for the progression of carotid intima-media thickness over a 16-year follow-up period: the Malmö Diet and Cancer Study. Atherosclerosis. 2015;239:615–21.
[15] Naya T, Hosomi N, Ohyama H, et al. Smoking, fasting serum insulin, and obesity are the predictors of carotid atherosclerosis in relatively young subjects. Angiology. 2008;58:677–84.
[16] Perciun R. Ultrasonographic aspect of subcutaneous tissue dystrophies as a result of insulin injections. Med Ultrason. 2010;12:104–9.
[17] Mancia G, De Backer G, Dominiczak A, et al. 2007 guidelines for the management of arterial hypertension: the task force for the management of arterial hypertension of the European society of hypertension (ESH) and of the European society of cardiology (ESC). Eur Heart J. 2007;28:1462–536.
[18] Frid AH, Hirsch LJ, Menchior AR, et al. Worldwide injection technique questionnaire study: injecting complications and the role of the professional. Mayo Clin Proc. 2016;91:1224–30.
[19] Hauner H, Stockamp B, Haastert B. Prevalence of lipohypertrophy in insulin-treated diabetic patients and predisposing factors. Exp Clin Endocrinol Diabetes. 1996;104:106–10.
[20] Poznyak A, Grechko AV, Poggio P, et al. The diabetes mellitus-atherosclerosis connection: the role of lipid and glucose metabolism and chronic inflammation. Int J Mol Sci . 2020;21:1835.
[21] Torimoto K, Okada Y, Mori H, et al. Relationship between fluctuations in glucose levels measured by continuous glucose monitoring and vascular endothelial dysfunction in type 2 diabetes mellitus. Cardiovasc Diabetol. 2013;12:1.
[22] Xu XH, Carvalho V, Wang XH, et al. Lipohypertrophy: prevalence, clinical consequence, and pathogenesis. Chin Med J (Engl). 2020;134:47–9.
[23] Libby P, Ridker PM, Hansson GK, et al. Inflammation in atherosclerosis: from pathophysiology to practice. J Am Coll Cardiol. 2009;54:2129–38.
[24] Stone NJ, Robinson JG, Lichtenstein AH, et al. 2013 ACC/AHA guideline on the treatment of blood cholesterol to reduce atherosclerotic cardiovascular risk in adults: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. Circulation. 2014;129(25 Suppl 2):S1–45.
[25] Reusch JE, Draznin BB. Atherosclerosis in diabetes and insulin resistance. Diabetes Obes Metab. 2007;9:455–63.
[26] Bertuzzi F, Meneghini E, Bruschi E, et al. Ultrasound characterization of insulin induced lipohypertrophy in type 1 diabetes mellitus. J Endocrinol Invest. 2017;40:1107–13.
[27] van den Munckhof ICL, Jones H, Hopman MTE, et al. Relation between age and carotid artery intima-medial thickness: a systematic review. Clin Cardiol. 2018;41:698–704.
[28] Herder M, Johnsen SH, Arntzen KA, et al. Risk factors for progression of carotid intima-media thickness and total plaque area: a 13-year follow-up study: the Tromsø Study. Stroke. 2012;43:1818–23.
[29] Powell-Wiley TM, Poirier P, Burke LE, et al. Obesity and cardiovascular disease: a scientific statement from the American Heart Association. Circulation. 2021;143:e984–e1010.
[30] Afshin A, Forouzanfar MH, Reitsma MB, et al. Health effects of overweight and obesity in 195 countries over 25 years. N Engl J Med. 2017;377:13–27.
[31] Pillay P, Carter J, Taylor H, et al. Independent relevance of different measures of adiposity for carotid intima-media thickness in 40 000 adults in UK biobank. J Am Heart Assoc. 2023;12:e026694.
[32] Kim HJ, Lim CW, Lee JH, et al. Gender-based differences in the relationship between fatty liver disease and atherosclerosis. Cardiovasc J Afr. 2016;27:281–6.

[33] Łoboz-Rudnicka M, Jaroch J, Bociąga Z, et al. Impact of cardiovascular risk factors on carotid intima-media thickness: sex differences. Clin Interv Aging. 2016;11:721–31.
[34] Kasliwal RR, Bansal M, Desai N, et al. A study to derive distribution of carotid intima media thickness and to determine its COrrelation with cardiovascular risk factors in asymptomatic nationwidE Indian population (SCORE-India). Indian Heart J. 2016;68:821–7.
[35] Su TC, Chien KL, Jeng JS, et al. Age- and gender-associated determinants of carotid intima-media thickness: a community-based study. J Atheroscler Thromb. 2012;19:872–80.
[36] Hajar R. Diabetes as "Coronary artery disease risk equivalent": a historical perspective. Heart Views. 2017;18:34–7.
[37] Evans JM, Wang J, Morris AD. Comparison of cardiovascular risk between patients with type 2 diabetes and those who had had a myocardial infarction: cross sectional and cohort studies. BMJ. 2002;324:939–42.
[38] Sainani G, Khan NR. Carotid artery intima media thickness—a correlative study of diabetics with postprandial hyperglycemia versus controls with euglycemia. Med Updat. 2012;22:298–303.
[39] Butt M, Zakira M. Association of common carotid intimal medial thickness (CCA-IMT) with risk factors of atherosclerosis in patients with type 2 diabetes mellitus. J Pak Med Assoc. 2009;59:590–3.
[40] Kumar A, Rathore A, Singh AK. Study of carotid intima media thickness (CIMT) in type 2 diabetes mellitus and its correlation with glycaemic control and inflammatory marker HsCRP. Int J Med Res Prof. 2020;6:81–6.
[41] Zhou YY, Qiu HM, Yang Y, et al. Analysis of risk factors for carotid intima-media thickness in patients with type 2 diabetes mellitus in Western China assessed by logistic regression combined with a decision tree model. Diabetol Metab Syndr. 2020;12:8.
[42] Kim ST, Kim BJ, Lim DM, et al. Basal C-peptide level as a surrogate marker of subclinical atherosclerosis in type 2 diabetic patients. Diabetes Metab J. 2011;35:41–9.
[43] Li Y, Zhao D, Li Y, et al. Serum C-peptide as a key contributor to lipid-related residual cardiovascular risk in the elderly. Arch Gerontol Geriatr. 2017;73:263–8.
[44] Liu M, Ao L, Hu X, et al. Influence of blood glucose fluctuation, C-peptide level and conventional risk factors on carotid artery intima-media thickness in Chinese Han patients with type 2 diabetes mellitus. Eur J Med Res. 2019;24:13.
[45] Kota SK, Mahapatra GB, Kota SK, et al. Carotid intima media thickness in type 2 diabetes mellitus with ischemic stroke. Indian J Endocrinol Metab. 2013;17:716–22.