Cardiovascular Pathology 52 (2021) 107328

Contents lists available at ScienceDirect

Cardiovascular Pathology

journal homepage: www.elsevier.com/locate/carpath

Original Article

Histopathologic evaluation of saphenous vein grafts in patients with type II diabetes mellitus undergoing coronary artery bypass grafting

Esin Ak[1,*], Koray Ak[2], Ahmet Midi[3], Elif Kervancıoğlu-Demirci[4], Sinan Arsan[2], Şule Çetinel[5], Rabia Pişiriciler[1]

[1] Department of Basic Medical Sciences, Histology and Embryology, Faculty of Dentistry, Marmara University, Istanbul, Turkey
[2] Department of Cardiovascular Surgery, Faculty of Medicine, Marmara University, Istanbul, Turkey
[3] Department of Pathology, Faculty of Medicine, Bahcesehir University, Istanbul, Turkey
[4] Department of Histology and Embryology, Istanbul University, Faculty of Medicine, Istanbul, Turkey
[5] Department of Histology and Embryology, Faculty of Medicine, Marmara University, Istanbul, Turkey

ARTICLE INFO

Article history:
Received 8 September 2020
Revised 13 February 2021
Accepted 13 February 2021

Keywords:
Type II Diabetes Mellitus
great saphenous vein
histopathology
immunohistochemistry
Caveolin-1
eNOS and VCAM-1

ABSTRACT

Introduction: Diabetes Mellitus (DM) has been known to be a risk factor for the development of more severe form of saphenous vein graft disease after coronary artery bypass grafting (CABG). We aimed to evaluate the impact of type II-DM on histopathological features of great saphenous vein grafts of patients undergoing CABG.

Patients and methods: Forty consecutive patients undergoing elective CABG were enrolled into the study. Patients were grouped into two; Diabetic group (n = 20); includes patients with preoperative diagnosis of type II-DM and Nondiabetic group (n = 20): those without type II-DM. In all patients, a short segment of the great saphenous vein graft at the level of medial malleolus was taken for light microscopy and transmission electron microscopy (TEM) evaluation. Moreover, immunoexpressions of Caveolin-1, Vascular cell adhesion protein 1 (VCAM-1) and endothelial nitric oxide synthase (eNOS) were studied.

Results: There were no differences in the demographics of patients between two groups. The magnitude of intimal fibrosis in diabetic group was slightly higher than in nondiabetics (1.95 ± 0.99 versus 1.3 ± 0.8, $P = .04$). In TEM, vacuolization in endothelial cells, substance accumulation along with coarse collagen fibers and cytoplasmic degeneration with vacuolization in muscle cells were detected in diabetic group. While there were no differences in Caveolin-1 and VCAM-1 immunostaining, the intensity of positive eNOS immunostaining was significantly higher in endothelium (2.10 ± 0.64 versus 1.55 ± 0.68, $P = .01$) and tunica media 1.75 ± 0.63 versus 1.2 ± 0.52, $P = .007$) in nondiabetic group, respectively) compared with diabetic group.

Conclusion: Type II DM might be a reason for decreased expression of eNOS and increased intimal fibrosis, vacuolization of endothelial and smooth muscle cells in saphenous vein grafts. The clinical implications of these alterations on the graft patency need to be evaluated.

© 2021 Elsevier Inc. All rights reserved.

1. Introduction

Recently great saphenous vein grafts (SVGs) continue to be one of the most commonly used conduits in coronary artery bypass grafting (CABG) despite their predisposition to failure. While the early patency rate has been shown to be satisfactory (around 75% after 12 to 18 months), more than 50% of the SVGs occlude within 10 years after CABG [1]. Many clinical risk factors have been shown to be crucial for the development of SVG failure like cigarette smoking, hyperlipidemia/triglyceridemia, female sex, lipoprotein (a) and diabetes mellitus (DM). The presence of DM has associated with more advanced and rapidly progressing vein graft disease after CABG [2–6]. The reasons for more severe nature of SVG disease in DM have not been clearly elucidated yet while some inherent vascular features related to DM such as chronic vessel wall inflammation driven mostly by proinflammatory cytokines have been claimed to be important in some studies [7].

The metabolic milieu of type-II DM alters vascular wall by a series of events including endothelial dysfunction, platelet hyperac-

* Corresponding author. Esin Ak, PhD, Marmara Universitesi Dis Hekimligi Fakultesi, Temel Bilimler Anabilim Dalı Saglik Bilimleri Basibuyuk Yerleskesi, 34854 Maltepe, Istanbul, Turkey
E-mail address: esinbir@yahoo.com (E. Ak).

https://doi.org/10.1016/j.carpath.2021.107328
1054-8807/© 2021 Elsevier Inc. All rights reserved.

Table 1
Demographics and preoperative data related to the patients.

	Nondiabetic group (n = 20)	Diabetic group (n = 20)	*P* value
Age (mean ± SD)	61,45 ± 11,1	63.8 ± 8.17	.43
Male sex (n, %)	14(70)	16 (80)	.72
Smoker (n, %)	7 (35)	9 (45)	.28
HT (n, %)	7 (35)	11 (55)	.14
COPD (n, %)	7 (35)	8 (40)	.73
Hyperlipidemia (n, %)	13 (65)	10 (50)	.21
PAD (n, %)	2 (10)	3 (15)	.63
HbA1c (g/dl, mean ± SD)	5.41 ± 0.5	9.3 ± 2.5	.000

DM: diabetes mellitus, HT: hypertension, COPD: chronic obstructive pulmonary disease, PAD: peripheral arterial disease, hemoglobin A1c: HbA1c.

tivity, oxidative stress and low-grade inflammation. Accumulation of advanced glycation end products (AGEs) and activation of AGEs receptors could accelerate the proliferation of smooth muscle cells (SMC) in SVGs from patients with DM. Even though blood glucose is well controlled, intimal hyperplasia is more prevalent in the vein grafts of diabetic patients compared to patients without DM, suggesting that additional factors contribute to the poor outcome [8]. In this study, we speculate that SVGs of diabetic patients might have some inherent histopathologic factors that could help explain the dismal fate of the SVGs after surgery. We aimed to compare the histopathologic features of SVGs of type-II diabetic patients undergoing CABG with those without DM.

2. Patients and methods

This study was approved by the local ethics committee (approval number: B.30.2.MAR.0.01.02/AEK/188,23.02.2012). All participants gave written informed consent. Forty consecutive patients with 3-vessel coronary artery disease undergoing elective first-time CABG were included. They were divided to 2 groups: diabetic (n = 20) and nondiabetic (n = 20). Inclusion criteria were as follows 1. Adult patients undergoing elective first time CABG, 2. Patients with type II DM (hemoglobin A1c (HbA1c) level was $\geq$ 6 g/dl) for diabetic group, 3. A normal left ventricle ejection fraction (EF), 4. No past medical history of thrombophelebitis or cellulitis of the lower limbs and 5. A healthy SVG that was judged via both preoperative duplex scanning and intraoperative direct macroscopic assessment were included. The patients with type II DM under control, presence of heart hailure and related statis dermatitis, peripheral arterial disease causing chronic critical limb ischemia were excluded. Uncontrolled DM was defined as HbA1c level was $\geq$ 6 g/dl on admission. Otherwise, patients with type II DM and HbA1c level $<$ 6 g/dl were discarded from the study. All patients were operated by the same surgical team. The data related to patients' demographic, preoperative routine biochemical analyses and perioperative variables were recorded to the institutional database prospectively (Table 1).

2.1. Harvesting of saphenous vein grafts

Under general anesthesia, median sternotomy was carried out in all patients for CABG. Concurrently, the harvesting of the SVG was started at the level of the medial malleolus and continued up to the thigh. Side branches were ligated with silk ligatures. All veins were kept in place until aortic cross clamping and then divided distally and proximally and preserved in small container containing patients own anticoagulated blood. Macroscopically, only the veins that were judged to be healthy were included. If the vein was ectatic/varicose or sclerotic, it was discarded. The harvested SVGs were preferred for the coronary arteries other than the left anterior descending coronary artery.

2.2. Light microscopy preparation and histopathological scoring

For histological evaluation, approximately 2 cm of the SVG at the level of the medial malleolus was taken and examined by light and transmission electron microscope (TEM) in all patients. For light microscopic investigations, samples were fixed with 10% neutral buffered formalin for 24 hours and routinely processed for paraffin embedding. Five micrometer thick sections were stained with Hematoxylin-Eosin (HE) for general morphology, Masson's Trichrome with Aniline Blue for collagen distribution and Verhoeff's Van Gieson stain for demonstration of elastic structures. All histopathological evaluations were performed by two observers who were blinded to the study groups independently.

A scoring system described by Thiene et al [9] was used. Intimal fibrous, thickening and sclerosis of the medial longitudinal muscular layer were defined as degeneration in the vessel wall and scored as; Intimal fibrosis consisted of an a cellular connective tissue proliferation, causing luminal narrowing and scored semiquantitatively as 0 (normal intima), 1 (focal proliferation with minimal narrowing), 2 (concentric proliferation with mild narrowing), 3 (concentric proliferation with moderate narrowing-less than 50% of the lumen), and 4 (marked narrowing-more than 50% of the lumen). Likewise, the sclerosis of the medial longitudinal muscular layer was scored semiquantitatively as score 0 (none), 1 (minimal), 2 (mild), 3 (moderate), and 4 (severe).

2.3. Caveolin-1 (Cav-1), vascular cell adhesion molecule-1 (VCAM-1) and endothelial nitric oxide synthase (eNOS) immunohistochemistry

Three µm thick sections were mounted on positive charged slides and dried overnight at 37^0 C. After tissue sections were deparaffinized, rehydrated in a graded ethanol series, they were exposed to 3% H_2O_2 in methanol to block endogenous peroxidase activity. Antigen retrieval was performed by heating the sections in a 10 mM citrate buffered solution at the microwave for 20 min and cooled in the room temperature. Following washing in phosphate buffered saline (PBS), tissue sections were incubated with protein blocking solution (Thermo Scientific, UltraVision Plus Large Volume Detection System, TP-060-HLX, USA) for nonspecific binding of antibodies. Primary antibodies for Cav-1 (dilution 1:200; Cav-1 (N: 20), sc-894, Santa Cruz Biotechnology, USA) were incubated for 1h at the room temperature and primary antibodies for VCAM-1 (ready to use, Thermo Scientific, MS-1101-R7, USA) and eNOS (ready to use, Thermo Scientific, RB-9279-R7, USA) were incubated for overnight at $+4^o$ temperature. Following washing with PBS, tissue sections were incubated with biotinylated secondary antibody and streptavidin–peroxidase (Thermo Scientific, UltraVision Plus Large Volume Detection System, TP-060-HLX, USA). Staining was visualized by incubation with a freshly prepared solution of 3.3′-diaminobenzidine tetrhydrochloride (DAB) solution. (Thermo Scientific, DAB Plus Substrate Staining System, TA-060-HDX, USA). The sections were counterstained with Mayer's hematoxylin, de-

hydrated and mounted with entellan. To test the specificity of the staining, the primary antibody was replaced with nonimmune serum in negative control sections. The intensity of staining of eNOS, Cav-1 and VCAM-1 was scored semiquantitatively as 0 (negative), 1 (mild), 2 (moderate) and 3 (severe).

Integrity of the endothelium has been evaluated by VCAM-1 immunohistochemical staining and classification ~~was~~ defined by Kendler et al [10]. In accordance with this classification, complete depiction of the endothelium (score 1), depiction of the endothelium-between 50 % and <100 % (score 2), depiction of the endothelium between 10% and 50% (score 3), 10 % endothelium or none (score 4) were used. All stained tissue sections were investigated by a light microscope (Olympus BX50, Olympus, Tokyo, Japan) and photographed with a digital camera (Olympus DP72, Tokyo, Japan) attached to a photomicroscope (Olympus BX51, Tokyo, Japan). To minimize the impact of confounding factors on the immunohistochemical data, we applied strict protocols of quantification of staining intensity. Also, proper masking of observers and well-defined scoring systems that clearly define each grade were used as described [11].

2.4. Transmission electron microscopy (TEM)

The samples were fixed with 2.5% glutaraldehyde in PBS (0.1 M, pH 7.2) for 4 h at 4^0 C, post fixed in 1% osmium tetroxide (OsO4) in PBS (0.1 M, pH 7.2) for 1 h, dehydrated in a graded series of alcohols, and embedded in Epon 812. Ultrathin sections (60 nm) were stained with uranyl acetate and lead citrate and investigated by using a JEOL 1200 EXII TEM (Tokyo, Japan) and photographed with a digital camera (Olympus Morada Soft Imaging System) attached to the TEM.

2.5. Statistical analysis

Statistical analysis of all data was carried out using Graph Pad Prism 6.0 (GraphPad Software, San Diego, CA, USA). Categorical variables were expressed as percentages and the chi-square test was used to analyze relationship between categorical data. Continuous variables were expressed as mean ± standard deviation (SD). Data were analyzed by Mann Whitney U test. Differences were considered to be significant at $P < 0.05$.

3. Results

The mean ages of nondiabetic and diabetic patients were 61.4 ± 11.1 and 63.8 ± 8.1 years, respectively (Table 1). Similarly, two groups were homogeneous in terms of major comorbidities. The mean percentages of HbA_1c of diabetic patients were significantly higher compared to the one in nondiabetic group. In diabetic group, the diagnosis of DM was made on admission for CABG in 2 patients. In the remaining 18 patients with DM, the mean duration of diabetes was 7.1 ± 2.9 (min-max: 3-12 years) and duration of disease was ≥ 5 years in 14 patients and ≥10 years in 5 patients. Of 20 patients with DM, 2 patients had never taken any treatment for DM, 8 were under divided doses of insulin regimen and 10 were under oral hypoglycemic agents. Two groups were comparable in terms of intraoperative variables and postoperative co-morbidities. The early mortality was zero for both groups.

3.1. Histopathologic results

In histopathologic evaluation, scores of intimal fibrosis and sclerosis of the medial longitudinal muscular layer were shown in Table 2. The mean score of intimal fibrosis in diabetic group was significantly higher than in nondiabetic group (1.95 ± 0.99 versus 1.3 ± 0.8, $P = .04$) (Fig. 1G). Sclerosis of the medial longitudinal muscular layer did not show a significant difference between diabetic and nondiabetic groups ($P > .05$) (Fig. 1G).

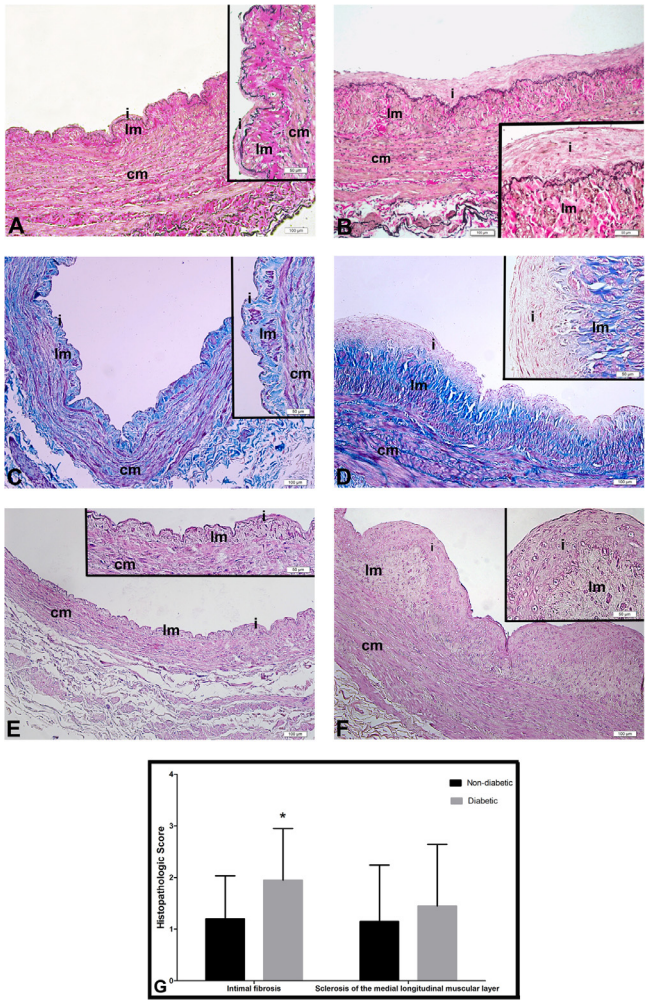


Fig. 1. Representative light microscopy of great saphenous veins in nondiabetic (A, C, E) and diabetic groups (B, D, F) using Verhoeff's Van Gieson (A, B), Masson's Trichrome with Aniline Blue (C, D) and HE (E, F) staining. Nondiabetic Group: Focal intimal fibrosis (score 1) and minimal sclerosis of the medial longitudinal muscular layer (score 1) were shown (A). Normal intima (score 0) and minimal sclerosis of the medial longitudinal muscular layer (score 1) were detected (C). Normal intima and medial longitudinal muscular layer were observed (E). Diabetic group: Intimal fibrosis (score 3) and moderate sclerosis of the medial longitudinal muscular layer (score 3) were shown (B, D and F). Intima: i; longitudinal layer of media (lm), circular layer of media (cm). Graph of histopathologic score of intimal fibrosis and sclerosis of the medial longitudinal muscular layer of diabetic and nondiabetic groups (G). *$P < 0.05$ compared to nondiabetic group.

3.2. Immunostaining results

In nondiabetic group eNOS immunoreactivity score was significantly higher in endothelium and tunica media (2.1 ± 0.64 and 1.55 ± 0.68, respectively) compared with diabetic group (1.75 ± 0.63 and 1.2 ± 0.52, respectively) ($P = .01$ and $P = .007$, respectively) (Fig. 2 A-C). There was no significant difference in Cav-1 immunoreactivity score in endothelium and tunica media between two groups ($P > .05$) (Fig. 2 D-F). Also, the intensity of VCAM-1 staining did not show a significant difference between two groups ($P > .05$) (Fig. 2 G-I). The integrity of endothelium was evaluated by VCAM-1 immunohistochemical staining. There were no significant differences in the integrity of endothelium between nondiabetic and diabetic groups (3 ± 1.12 versus 2,9 ± 0.96 $P > .05$) (Fig. 3).

Table 2
Scores of intimal fibrosis and sclerosis of the medial longitudinal muscular layer.

	Intimal fibrosis		Sclerosis of the medial longitudinal muscular layer	
Score	Non diabetic n =20 (%)	Diabetic n = 20 (%)	Nondiabetic n =20 (%)	Diabetic n = 20 (%)
0	3 (15)	1 (5)	7 (35)	5 (25)
1	9 (45)	7 (35)	6 (30)	7 (35)
2	7 (35)	4 (20)	4 (20)	2 (10)
3	1 (5)	8 (40)	3 (15)	6 (30)
4	none	none	none	None

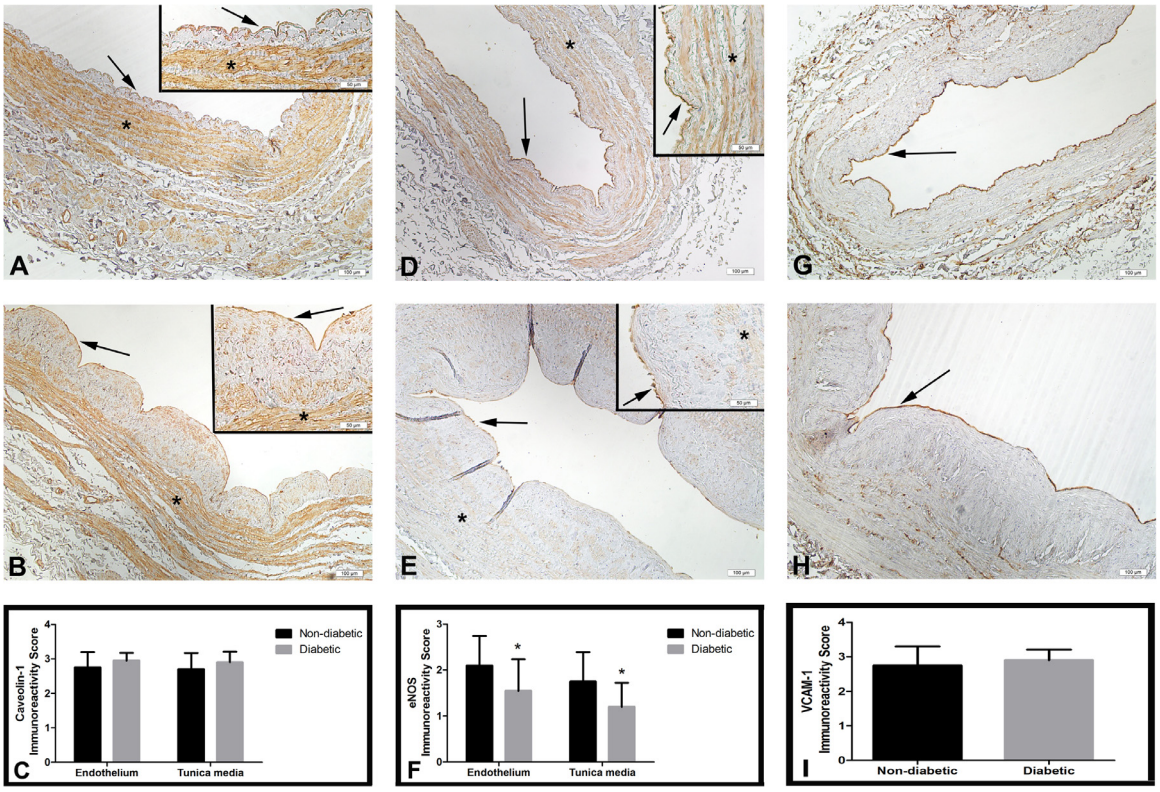


Fig. 2. Representative light microscopy of great saphenous veins in nondiabetic (A, D, G) and diabetic groups (B, E, H) using Cav-1 (A, B), eNOS (D, E) and VCAM-1 (G, H) immunohistochemical staining. Nondiabetic group: Strong Cav-1 (A), eNOS (D) immunoreactivity in endothelium (arrows) and in muscle cells (*) and strong VCAM-1 (G) immunoreactivity in endothelium (arrows) were shown. Diabetic group: Strong Cav-1 (B), and VCAM-1 (H) immunoreactivity in endothelium (arrows), strong Cav-1 (B) immunoreactivity in muscle cells (*) and moderate eNOS (E) immunoreactivity in endothelium (arrow) and weak eNOS (E) immunoreactivity in muscle cell (*) were shown. Graphs of immunoreactivity score of Cav-1 (C), eNOS (F) and VCAM-1 (I) staining. $^{*}P < 0.05$ compared to nondiabetic group.

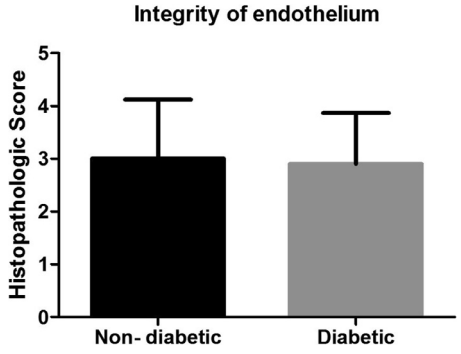


Fig. 3. Histopathologic score of the integrity of endothelium in nondiabetic and diabetic groups.

3.3. TEM findings

In nondiabetic group, in general the morphology of endothelial cells and collagen fibers was normal. A slight dilation in perinuclear area of the muscle cells was observed (Fig. 4A). In diabetic group, vacuolization in endothelial cells, substance accumulation along with coarse collagen fibers and cytoplasmic degeneration with vacuolization in muscle cells were detected in the most of the samples of diabetic group (Fig. 4B).

4. Discussion

Our results demonstrate that even though the SVGs from diabetic patients were macroscopically normal, they were found to carry some histopathological substrates of accelerated atherosclerosis like intimal fibrosis and the reduced expression of eNOS. These substrates might pave the way for the development of a higher rate of accelerated atherosclerosis of the SVG in patients with DM compared to the nondiabetic patients in the long term.

The pathophysiologic process of postoperative SVG failure has been shown to happen in a time-related fashion as 1. acute graft thrombosis within the first month, 2. Neointimal hyperplasia (NIH) at anastomotic sites (1 month to 2 years) (hyperplastic stage) and (3) accelerated atherosclerotic degeneration (after 2 years) [2]. Patients with DM are more vulnerable to SVG failure immediately

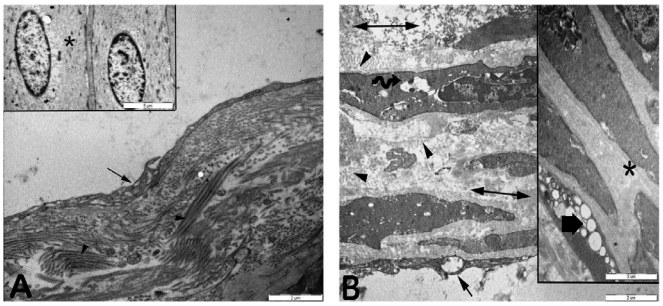


Fig. 4. . Represantative TEM microscopy of great saphenous veins in nondiabetic (A) and diabetic (B) groups. (A): Endothelial cell (arrow), collagen fibers (arrowheads) and slight dilation in perinuclear area of the muscle cell (asterix). (B): Endothelial cell with vacuole (arrow), collagen fibers (arrowheads), cytoplasmic degeneration (broken arrow) of fibroblast, substance accumulation in the connective tissue (double-headed arrow), cytoplasmic degeneration with vacuolization (thick arrow) in the muscle cell, connective tissue between two muscle cells (asterix).

after CABG as the presence of diseased runoff vessels and multiple derangements in coagulation and fibrinolysis increase the risk of acute graft thrombosis. Likewise, endothelial impairment, growth factor dysregulation and the presence of AGEs have been shown to be potential factors promoting the process of SMC migration and proliferation and extracellular matrix activation that characterize the NIH. The reasons behind more aggressive nature of the atherosclerotic degeneration in patients with type II DM were shown to be closely related with insulin dysregulation, lipid metabolism and abnormal expression of cytokines in SVGs [12].

Several studies revealed that SVGs of patients with type II DM might have some inherent substrates related to endothelial cells (ECs) and SMCs of SVGs facilitating the development of NIH and accelerated atherosclerosis. In studies by Gur et al [13] and Larusso et al [14], eNOS mediated vasodilatation response of SVGs was depressed significantly in diabetic patients compared to those from the nondiabetics. Roberts et al [15] found that ECs of SVGs from type II DM patients show inhibition of Akt and eNOS pathways compared to the nondiabetic ECs in vitro. Hu et al [16] revealed that as seen in diabetic atherosclerotic disease, expression of the transcription factor called bone morphogenetic protein 4 (BMP4) was considerably higher in venous conduits of diabetes patients. Moreover, the increased expression of BMP4 causes more oxidative stress in the wall of the SVGs from diabetic patients.

In the present study, we found that eNOS immunostaining of the endothelium and media of diabetic SVGs significantly decreased compared to the nondiabetics that supports the presence of already existing EC dysfunction in diabetic SVGs. This finding supports the previous results of the studies showing the presence of the already existing EC dysfunction in SVGs in diabetics. On the other hand, no differences were found in Cav-1 and VCAM-1 expression in the vessel wall between the diabetics and nondiabetics. Walker et al [17] showed that activation of the ECs of SVGs causes expression of various adhesion molecules like ICAM-1, VCAM-1 and E-selectin that are claimed to be the reason for the enhanced vessel wall inflammation and related vein graft disease after cardiac surgery. In the present study, while the eNOS expression of diabetic SVGs depressed in comparison, we could not detect a significant difference in the expression of EC Cav-1. Endothelial pathways connecting Cav-1, eNOS, and reactive oxygen species (ROS) are closely interrelated and overexpression of Cav-1 reduces endothelial cell proliferation, migration, and nitric oxide production and increases expression of VCAM -1 [18-19].

Similarly, SMCs of diabetic SVGs have some inherent morphological differences like rhomboid phenotype and are more migratory, but less proliferative SMCs compared to the nondiabetics in vitro [20]. Moreover, in a study by Sun et al [21] SVGs exposed to diabetic conditions demonstrated a prominent increase in the expression of matrix metallopeptidase (MMP)-2 and MMP-9 but a notable decrease in the expression of tissue metallopeptidase inhibitor (TIMP)-2 and TIMP-3 in protein concentration compared with control group. An imbalance in the MMPs/TIMPs activity ratio was claimed to be one of the leading factors in the pathogenesis of several vascular diseases such as abdominal aortic aneurysm, varicose veins, preeclampsia and NIH [22].

It has been shown that a certain proportion of SVGs in patients undergoing CABG has ultrastructural abnormalities. The grafts with low endothelial cell coverage, stenotic lumen, and thick walls were found to be at a higher risk of developing intrawall lesions that lead to early graft failure at postoperative 6 months [23]. Apart from EC dysfunction, diabetic SVGs have been shown to harbor some ultrastructural alterations that are claimed to facilitate the development of vein graft disease after CABG. Experimentally, it was shown that type 2 DM causes a significant increase in extracellular matrix and SMC deposition in surgical vein grafts [24]. Also, AGEs were claimed to promote a simultaneous increase in proliferation and apoptosis of SMCs of the vein grafts and, thereby, accelerating arterializations or atherosclerosis of vein grafts [25]. Lorusso et al [14] found that SVGs from diabetic patients had severe intimal hyperplasia and a higher incidence of atherosclerotic calcified plaques compared to those from the nondiabetics. Moreover, high levels of proteinuria and glycosylated hemoglobin were found to be good predictors of the degree of structural derangements of the SVCs.

In the present study, marked intimal fibrosis in diabetic SVGs was the only significant finding on light microscopic evaluation. This finding was consistent with the finding reported by Lorusso et al [14]. The most of the patients with DM were in an uncontrolled situation in terms of blood glucose levels (mean glycosylated Hemoglobin (HbA1c) around 8,3 g/dl) and exposed to the higher blood levels of AGEs for a long time. In a study by Friedl et al [26], increased expression of TGF-β1 in SVGs was the main reason of the preexisting intimal hyperplasia and fibrosis. Our TEM findings were also parallel with those seen in light microscopy. The increased number of vacuoles in the EC and VSM of diabetic SVGs could be attributed to the mitochondrial dysfunction and activation of the cellular apoptosis [27].

Our study has some apperent limitations. First, as type II DM is a well-known factor for inflammation and oxidative stress in the vessel wall, comparison of the tissue oxidative stress markers and serum levels of proinflammatory cytokines would have increased the power of the present study. Second, semiquantitative analyses of the histopathological findings are not without limitations. Some level of observer bias and variability that could contribute to reproducibility issues are inevitable to some extent. The use of quantitative analyses like measurement of protein level or enzyme activity would further strengthen the results of the study. Third, discarding the macroscopically unhealthy veins from both groups could have resulted in selection bias. Last, the long-term clinical data about graft failure and related recurrent angina and myocardial infarction are missing.

5. Conclusion

In conclusion, our results show that patients undergoing CABG with type II DM have some pathologic substrates in the SVGs like intimal fibrosis, vacuolization of the endothelial cells and SMCs. The impact of these substrates on the patency of SVGs has yet to be elucidated. Further studies are warranted to figure out the clinical implications of the already existing pathologic changes of diabetic SVGs in both short and long term after CABG.

Acknowledgments

We gratefully acknowledge the assistance of Dr. Erol Nargileci (Colombus, IN) for checking grammar and spelling errors.

Funding

This study was supported by Marmara University Scientific Research Projects Commission, Project No: SAG-B-110412-0074.

References

[1] Moshkovitz Y, Raanani E. The art of saphenous vein grafting and patency maintenance. J Thorac Cardiovasc Surg 2016;151:300–2. doi:10.1016/j.jtcvs.2015.09.004.

[2] Harskamp RE, Lopes RD, Baisden CE, de Winter RJ, Alexander JH. Saphenous vein graft failure after coronary artery bypass surgery: pathophysiology, management, and future directions. Ann Surg 2013;257:824–33. doi:10.1097/SLA.0b013e318288c38d.

[3] Motwani JG, Topol EJ. Aortocoronary saphenous vein graft disease: pathogenesis, predisposition, and prevention. Circulation 1998;97:916–31. doi:10.1161/01.cir.97.9.916.

[4] Cox JL, Chiasson DA, Gotlieb AI. Stranger in a strange land: the pathogenesis of saphenous vein graft stenosis with emphasis on structural and functional differences between veins and arteries. Prog Cardiovasc Dis 1991;34:45–68. doi:10.1016/0033-0620(91)90019-i.

[5] Deb S, Singh SK, Moussa F, Tsubuota H, Une D, Kiss A, et al. The long term impact of diabetes on graft patency after coronary artery bypass grafting surgery: a substudy of the multicenter Radial Artery Patency Study. J Thorac Cardiovasc Surg 2014;148:1246–53 6. doi:10.1016/j.jtcvs.2014.06.057.

[6] Kapur A, Bartolini D, Finlay MC, Qureshi AC, Flather M, Strange JW, et al. The bypass angioplasty revascularization in type 1 and type 2 diabetes study: 5-year follow up of revascularization with percutaneous coronary intervention versus coronary artery bypass grafting in diabetic patients with multivessel disease. J Cardiovasc Med (Hagerstown) 2010;11:26–33. doi:10.2459/JCM.0b013e328330ea32.

[7] Lowe G, Woodward M, Hillis G, Rumley A, Li Q, Harrap S, et al. Circulating inflammatory markers and the risk of vascular complications and mortality in people with type 2 diabetes and cardiovascular disease or risk factors: the advance study. Diabetes 2013;63:1115–23. doi:10.2337/db12-1625.

[8] Sun Y, Kang L, Li J, Liu H, Wang Y, Wang C, et al. Advanced glycation end products impair the functions of saphenous vein but not thoracic artery smooth muscle cells through RAGE/MAPK signaling pathway in diabetes. J Cell Mol Med 2016;20:1945–55. doi:10.1111/jcmm.12886.

[9] Thiene G, Miazzi P, Valsecchi M, Valente M, Bortoloti U, Casarotto D, et al. Histological survey of the saphenous vein before its use as autologous aorto-coronary bypass graft. Thorax 1980;35:519–22. doi:10.1136/thx.35.7.519.

[10] Kendler M, Averbeck M, Simon JC, Ziemer M. Histology of saphenous veins after treatment with the ClariVein® device - an ex-vivo experiment. J Dtsch Dermatol Ges 2013;11:348–52. doi:10.1111/ddg.12022.

[11] Libard S, Cerjan D, Alafuzoff I. Characteristics of the tissue section that influence the staining outcome in immunohistochemistry. Histochem Cell Biol 2019;151:91–6. doi:10.1007/s00418-018-1742-1.

[12] Gleissner CA, Galkina E, Nadler JL, Ley K. Mechanisms by which diabetes increases cardiovascular disease. Drug Discov Today Dis Mech 2007;4:131–40. doi:10.1016/j.ddmec.2007.12.005.

[13] Gür DÖ, Gür Ö, Gürkan S, Cömez S, Gönültaş A, Yılmaz M. Comparison of endothelial function of coronary artery bypass grafts in diabetic and nondiabetic patients: Which graft offers the best? Anatol J Cardiol 2015;15:657–62. doi:10.5152/akd.2014.5613.

[14] Lorusso R, Pentiricci S, Raddino R, Scarabelli TM, Zambelli C, Villanacci V, et al. Influence of type 2 diabetes on functional and structural properties of coronary artery bypass conduits. Diabetes 2003;52:2814–20. doi:10.2337/diabetes.52.11.2814.

[15] Roberts AC, Gohil J, Hudson L, Connolly K, Warburton P, Suman R, et al. Aberrant phenotype in human endothelial cells of diabetic origin: implications for saphenous vein graft failure? J Diabetes Res 2015:409432 2015. doi:10.1155/2015/409432.

[16] Hu J, Liu J, Kwok MW, Wong RH, Huang Y, Wan S. Bone morphogenic protein-4 contributes to venous endothelial dysfunction in patients with diabetes undergoing coronary revascularization. Ann Thorac Surg 2013;95:1331–9. doi:10.1016/j.athoracsur.2012.12.028.

[17] Walker T, Müller I, Raabe C, Nohe B, Zanke C, al Ziemer Get. Effective silencing of adhesion molecules on venous endothelial cells for protection of venous bypass grafts. Eur J Cardiothorac Surg 2011;40:1241–7. doi:10.1016/j.ejcts.2011.01.076.

[18] Wong DJ, Lu DY, Protack CD, Kuwahara G, Bai H, Sadaghianloo N, et al. Ephrin type-B receptor 4 activation reduces neointimal hyperplasia in human saphenous vein in vitro. J Vasc Surg 2016;63:795–804. doi:10.1016/j.jvs.2014.09.036.

[19] Shiroto T, Romero N, Sugiyama T, Sartoretto JL, Kalwa H, Yan Z, et al. Caveolin-1 is a critical determinant of autophagy, metabolic switching, and oxidative stress in vascular endothelium. PLoS One 2014;9:e87871. doi:10.1371/journal.pone.0087871.

[20] Madi HA, Riches K, Warburton P, O'Regan DJ, Turner NA, Porter KE. Inherent differences in morphology, proliferation, and migration in saphenous vein smooth muscle cells cultured from nondiabetic and Type 2 diabetic patients. Am J Physiol Cell Physiol 2009;297:C1307–17. doi:10.1152/ajpcell.00608.2008.

[21] Sun Y, Ding W, Wei Q, Sheng WC. Disorder gene expression of extracellular matrix and adhesion molecules in saphenous vein conduits of diabetic patients. Interact Cardiovasc Thorac Surg 2012;14:279–82. doi:10.1093/icvts/ivr053.

[22] Raffetto JD, Khalil RA. Matrix metalloproteinases and their inhibitors in vascular remodeling and vascular disease. Biochem Pharmacol 2008;75:346–59. doi:10.1016/j.bcp.2007.07.004.

[23] Kouzi-Koliakos K, Kanellaki-Kyparissi M, Marinov G, Knyazhev V, Tsalie E, Batzios C, et al. Prebypass histological and ultrastructural evaluation of the long saphenous vein as a predictor of early graft failure. Cardiovasc Pathol 2006;15:336–46. doi:10.1016/j.carpath.2006.07.005.

[24] Salzberg SP, Filsoufi F, Anyanwu A, von Harbou K, Karlof E, Carpentier A, et al. Increased neointimal formation after surgical vein grafting in a murine model of type 2 diabetes. Circulation 2006;114(1 Suppl):I302–7. doi:10.1161/CIRCULATIONAHA.105.001339.

[25] Ping S, Liu S, Zhou Y, Li Z, Li Y, Liu K, et al. Protein disulfide isomerase-mediated apoptosis and proliferation of vascular smooth muscle cells induced by mechanical stress and advanced glycosylation end products result in diabetic mouse vein graft atherosclerosis. Cell Death Dis 2017;8:e2818. doi:10.1038/cddis.2017.213.

[26] Friedl R, Li J, Schumacher B, Hanke H, Waltenberger J, Hannekum A, et al. Intimal hyperplasia and expression of transforming growth factor-beta1 in saphenous veins and internal mammary arteries before coronary artery surgery. Ann Thorac Surg 2004;78:1312–18. doi:10.1016/j.athoracsur.2004.02.066.

[27] Mishiro K, Imai T, Sugitani S, Kitashoji A, Suzuki Y, Takagi T, et al. Diabetes mellitus aggravates hemorrhagic transformation after ischemic stroke via mitochondrial defects leading to endothelial apoptosis. PLoS One 2014;9:e103818. doi:10.1371/journal.pone.0103818.