

Original Article

A novel rat model combining surgical vein grafting with type 2 diabetes

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Abstract: Background: Diabetes is a risk factor for vein graft failure after bypass surgery with coronary artery disease (CAD). We first developed a vein grafting model in rats with a type 2 diabetes mellitus-like (T2DM) condition to provide animal models for relevant clinical research. Methods: Sixty male Sprague-Dawley rats were fed high-fat (T2DM group) or normal food (CON group) for one month and then injected (T2DM group) with streptozotocin (35 mg/kg) to induce T2DM. Insulin or glucose tolerance tests were performed to verify the T2DM models. All rats subsequently underwent surgical vein grafting, and the morphology of their grafts at 0, 14, and 28 days was analyzed under microscope. Results: The T2DM group exhibited significantly increased glucose, insulin, free fatty acids, and cholesterol levels (all, $P < 0.05$), and also exhibited significant hyperglycemic and hyperinsulinemic responses to an intraperitoneal glucose tolerance test compared to that in the CON group. Furthermore, the insulin sensitivity of the T2DM group was significantly reduced as the CON group gained little change according to an insulin tolerance test ($P < 0.05$). Additionally, the number of cell nuclei of the intima and media on the 14th and 28th day in the T2DM group approximately doubled as compared to that in the CON group (both, $P < 0.05$). Conclusion: We successfully developed a model of surgical vein grafting in type 2 diabetic rats. This novel rat model may facilitate in vivo mechanistic studies of the increased neointimal formation in type 2 diabetes mellitus-related vein graft failure.

Keywords: Coronary artery disease, type 2 diabetes, rat model, neointima, surgery

Introduction

Coronary artery disease (CAD) is a leading cause of death and disability in individuals around the world, and the incidence and mortality of CAD in China has increased each year [1, 2]. Coronary artery bypass surgery (CABG) remains the gold standard for treating CAD, making it the most common form of heart surgery for CAD. Although arterial grafts have superior long-term patency rates, especially for the left internal mammary artery to the left anterior descending artery [3], saphenous vein grafts remain a cornerstone of daily coronary revascularization practice [4]. As for the pathological changes of CAD, intimal hyperplasia is the most common lesion that underlies vein graft stenosis, which is defined as excessive smooth muscle cell (SMC) migration and proliferation in the intima of the vein graft wall [5].

Unfortunately, there is no effective approach to prevent this clinical condition.

Diabetes mellitus (DM) is also an important cause of global mortality and morbidity and is the most prevalent metabolic disorder worldwide [6, 7]. Furthermore, scientists have demonstrated that patients with diabetes and no history of myocardial infarction (MI) have a similar risk of MI compared to patients without diabetes and with a history of MI, which could lead to serious clinical consequences [8].

Type 2 diabetes mellitus (T2DM) is a condition characterized by hyperglycemia caused by insulin resistance. T2DM accounts for 90-95% of all diabetes prevalence [9]. T2DM can cause a variety of vascular changes and lead to more extensive neointimal hyperplasia, plaque formation, altered hemodynamics, and inade-

Table 1. High-fat diet composition

Ingredients	Diet (g/kg)
Lard	320
Powder	200
Glutinous rice flour	100
Corn starch	100
Peanut powder	75
Sesame powder	50
Milk Powder	90
Egg	60
Salt	5
Total	1,000

Table 2. Caloric composition

	Normal protein diet (kcal)	High fat diet (kcal)
Fat	39.87 (10%)	325.8 (56%)
Carbohydrates	228.76 (64%)	185.6 (32%)
Protein	93.68 (26%)	69.6 (14%)
Total	362.31	580.4

quate compensatory remodeling [10]. Furthermore, these vascular changes are found to be closely related to CAD. Extensive neointimal hyperplasia and rapid plaque formation in patients with diabetes can lead to more serious restenosis and less stable atherosclerotic plaques. Moreover, compared with non-diabetic individuals, patients with diabetes who undergo CABG have worse outcomes, and vein graft occlusive disease occurs in nearly half of the conduits within 10 years [11]. As a result, patients with diabetes have a three- to seven-fold higher risk of death due to a cardiovascular event as compared to the general population [12].

A recent study by Oikawa S et al. indicated that 25% of the patients who underwent coronary revascularization had diabetes, highlighting the clinical importance of understanding the underlying cellular and molecular mechanisms of vein graft stenosis [13]. To date, only one animal model exists to investigate the precise molecular and cellular pathways involved in diabetic vein graft failure [14]. However, that model is in syngeneic and mutant mice, and it cannot replicate the natural pathogenesis by creating diabetic characteristics that parallel the human pathogenesis of T2DM. Therefore, we sought to develop a model of T2DM using fat-fed strepto-

zotocin (STZ)-treated rats and then performed vein graft operations to evaluate the molecular mechanisms of the graft failure. We hoped that this model would facilitate further research regarding the pathophysiology of vein graft neointimal formation in T2DM patients.

Materials and methods

Animals

Sixty healthy male Sprague-Dawley rats (aged 7-8 weeks; weighing 160-180 g) were obtained from the Laboratory Animal Centre of Guangxi Medical University (Nanning, Guangxi, China). The animals were housed in standard polypropylene cages (one rat per cage), and the cages were maintained under controlled room temperature ($22 \pm 2^\circ\text{C}$) and humidity ($55 \pm 5\%$), with a 12:12 h light and dark cycle. Rats were provided with free access to food (a standard ad libitum laboratory diet) and water, with a 1-week adaptive feeding period. All animal care and experimental protocols were performed according to the Ethical Principles in Animal Research (as adopted by the Guangxi Medical University for Animal Experimentation) and the Guide for the Care and Use of Laboratory Animals.

Development of fat-fed and STZ-treated type 2 diabetic rats

We randomized the rats using Excel's randomization function ("Rand.") into the control group ($n = 30$, CON) and the diabetic model group ($n = 30$, T2DM). All the rats were allocated into two ad libitum dietary regimens: a normal protein diet (12% fat, 21% protein, and 67% carbohydrate as a percentage of the total kcal) or a high-fat diet (58% fat, 25% protein, and 17% carbohydrate) for 4 weeks [15]. The composition and preparation of the high-fat diet is listed in **Tables 1** and **2**. The weight and food intake of each rat were evaluated each week.

After the 4 weeks of dietary manipulation, STZ (Sigma, St. Louis, MO, USA) was dissolved in a citrate buffer (pH 4.4) at a concentration of 15 mg/mL, and filter-sterilized by a 0.2- μm filter. All animals fasted overnight. The high-fat-fed animals were then injected intraperitoneally with a low dose of STZ (35 mg/kg), and the control rats received a 1 mL/kg dose of the vehicle

Surgical vein grafting model in diabetic rats

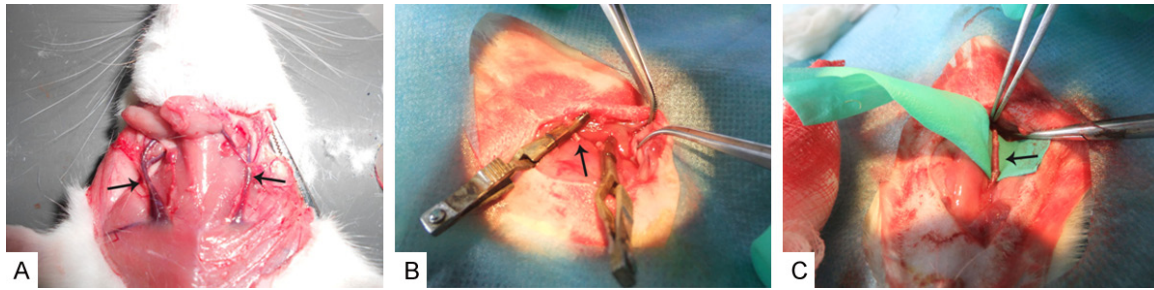


Figure 1. Photographs of the surgical procedure. (A) A 1-1.2-cm segment of the right common carotid artery was mobilized, (B) the vessel was ligated with microhemostatic clamps and a 0.5-cm vein segment was removed, and (C) the vein segment was grafted between the two ends of the carotid artery.

(citrate buffer, pH 4.4). After the injections, all animals were allowed to continue with their respective diets until the end of the study.

The following experiments were started on the third day after the STZ injection. On experimental days, the food was removed at 7 AM, and blood samples were collected via tail snips 4 h later. The serum samples were used to evaluate the levels of glucose, insulin, free fatty acid (FFA), and cholesterol. Rats that had a fasting blood glucose level of ≥ 7.8 mmol/L were considered to be the diabetic group.

Insulin tolerance test (ITT)

Insulin (0.75 IU/kg) was administered via intraperitoneal injection, and blood samples were collected at 0, 30, 60 and 120 post-injection to evaluate the plasma glucose response. Insulin sensitivity was evaluated with the glucose disappearance rate within 120 min, and the value was reported as the percentage of the initial plasma glucose level.

Intraperitoneal glucose tolerance test

After an overnight fast, the rats received an intraperitoneal injection of 40% glucose (2 g/kg body weight). Blood samples were collected from the tail at 15, 30, 45, 60, 120, 180 and 240 post-injection to evaluate the glucose and insulin responses.

Development of the vein graft model

One week after the STZ injection, we created a graft model as previously described [16]. In brief, the rats were anesthetized by 10% chloral hydrate (300 mg/kg body weight, intraperitoneally), injected with heparin sodium (200 U/kg body weight, tail vein injection), and the

operation was performed under a dissecting microscope (XT-X-4A, Zhenjiang Xintian Medical Devices Co., Ltd, China). The right external jugular vein was initially exposed, and a 1-cm vein segment was removed. A 1-1.2-cm segment of the right common carotid artery was then mobilized (**Figure 1A**), and a diluted papaverine solution was sprayed on the surface of the artery. Afterwards, the vessel was ligated with microhemostatic clamps, and a 0.5-cm vein segment was removed (**Figure 1B**). Later, the vein graft and both lumens were washed with a saline solution that contained 100 U/mL of heparin and the vein segment was subsequently grafted between the two ends of the carotid artery by the “cuff sleeve technique” (**Figure 1C**) with a 20 G 1.1-mm arterial cannula (Becton Dickinson Critical Care Systems Pte Ltd, Singapore). After surgery, the animals received an intramuscular injection of penicillin (400,000 U/kg, body weight) and fed as normal. All procedures were conducted in a pathogen-free environment.

Collection of the implanted grafts

Prior to the procedure, we collected preoperative metabolic data for all animal groups, the rats were fasted for 6 h, tail vein blood samples were collected to evaluate plasma glucose and insulin levels, and the rats were subsequently sacrificed. Previous studies have demonstrated that rats in this state exhibit steady neointimal hyperplasia [17, 18]. Prior to the surgery, the rats were pre-medicated with high-rate isoflurane and perfusion-fixed with fresh 4% paraformaldehyde in a phosphate-buffered saline solution. To prepare the frozen sections, the rats were sacrificed via cervical dislocation, and the vein grafts were harvested via en bloc resection and were immediately frozen in liquid nitrogen and stored at -80°C .

Table 3. Effect of streptozotocin in type 2 diabetic rats

Variable	CON (n = 18)	T2DM (n = 18)	P
Weight (g)	445 ± 31	436 ± 28	NS
Glucose (mmol/L)	6.9 ± 0.6	24.8 ± 2.9	< 0.05
Insulin (pmol/L)	0.58 ± 0.08	1.00 ± 0.22	< 0.05
FFA (μg/L)	22.18 ± 5.83	90.91 ± 17.16	< 0.05
Cholesterol (μmol/L)	2.43 ± 0.42	9.44 ± 1.71	< 0.05

Abbreviations: CON, Control; T2DM, Rats with a type 2 diabetes-like condition; NS, Not significant.

Morphology analysis

The vein grafts were harvested in the 0th, 2nd, and 4th week after their insertion (from seven randomly selected rats for each time point) by cutting the transplanted segments from the native vessels at the cuff end. The high-fat diet was provided until they were sacrificed. The levels of glucose in the rats were detected to confirm that the animals were diabetic throughout the entire experiment. From each animal, 5 sections within the vein graft were cut and used for the morphometric assessment. We measured four regions of a section in the shape of a cross and recorded the neointimal growth in micrometers (mean ± standard error). Intimal and medial morphometric examinations were performed with a hematoxylin and eosin staining kit (Jiancheng Biotechnology Co., Ltd. Nanjing, China) and the thickness of the intima and media was measured. Cell counts in the intima and media were performed in two opposite regions of each section and expressed as the number of nuclei per 100-μm vessel wall section [16]. Each of the stained sections was measured five times, and the average was calculated. All measurements and data analysis were performed by two independent researchers.

Statistical analysis

All results were analyzed with SPSS software (version 17.0, SPSS Inc., Chicago, IL), and differences with a *P*-value of < 0.05 were considered statistically significant. All results were expressed as the mean ± standard error, unless otherwise stated. Because the data were normally distributed, a one-way analysis of variance was used to find the effect of streptozotocin in T2DM rats by comparing the glucose, insulin, FFA, and cholesterol levels in two groups at each time point. Curve estimation and area under the curve were developed

to analyze the glucose tolerance and insulin sensitivity by comparing the results of IPGTT and ITT, respectively. Column-value histogram statistics were generated to analyze the difference of vein restenosis by comparing the thickness of the intima and media and the cell nuclei content in vein grafts of both groups by one-way ANOVA analysis.

Results

The first aim of this study was to create a rat model of T2DM, while the second aim was to assess neointimal formation after surgical vein grafting in a clinically and physiologically relevant manner on the rat model.

Glucose, insulin, FFA, and cholesterol levels after STZ injection

After 4 weeks of the test diets, the T2DM animals were injected with STZ, and most of the experiments were conducted 3 days after that injection. In the T2DM group, significant increases in plasma glucose, insulin, FFA, and cholesterol were observed compared to the control animals, although no significant difference was observed in the weight (**Table 3**).

Intraperitoneal glucose tolerance test results

As shown in **Figure 2A**, the T2DM group exhibited hyperglycemia over the full 240 min after the glucose injection. In the first 15 min, plasma glucose levels in the CON and T2DM groups increased rapidly, although the CON group levels decreased significantly after that point. The area under the glucose curve was significantly greater in the T2DM group than in the CON group (6,402.11 ± 405.84 mmol/L·min vs. 2,115.66 ± 192.46 mmol/L·min, *P* < 0.05, **Figure 2B**).

In addition, treating high-fat-fed rats with a low dose of STZ damaged their insulin-producing β cells, which resulted in hyperglycemia despite insulin levels that were similar to or even higher than those in normal rats. After the glucose injection, the plasma insulin levels of the T2DM group remained high throughout the entire test (**Figure 3A**). In contrast, the insulin levels in the CON group increased quickly in the first 15 min and then subsequently declined similar to the plasma glucose findings. The area under the insulin curve was significantly greater in the

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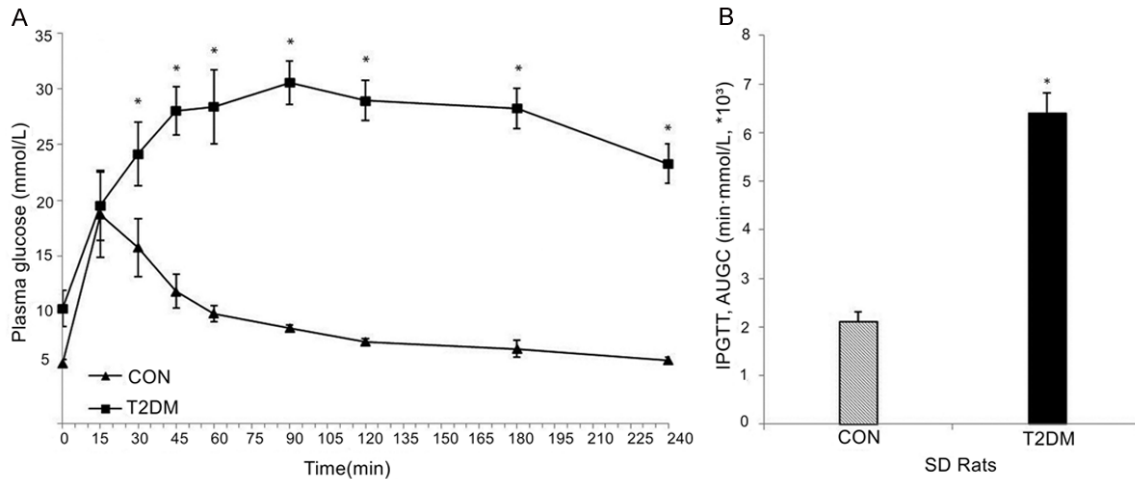


Figure 2. A. Plasma glucose levels during an intraperitoneal glucose tolerance test in the diabetic (T2DM) and control (CON) groups at 3 days after a streptozotocin injection. B. Areas under the glucose curves for the two groups. The data are shown as the mean \pm standard error ($n = 7$ rats per group per time point). *Significant difference relative to the controls, $P < 0.05$.

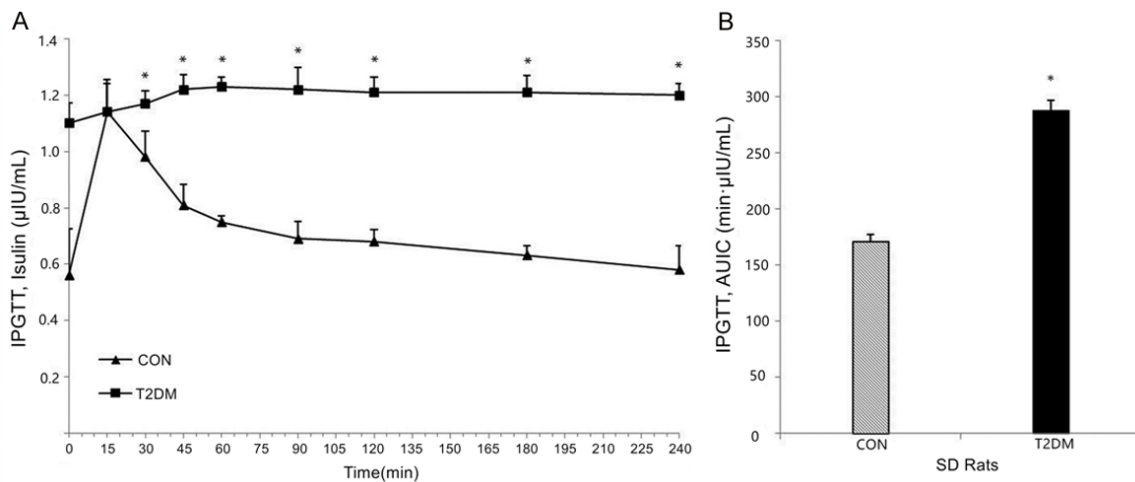


Figure 3. A. Plasma insulin levels during an intraperitoneal glucose tolerance test in the diabetic (T2DM) and control (CON) groups at the 3rd day after a streptozotocin injection. B. Areas under the insulin curves for the two groups. The data are shown as the mean \pm standard error ($n = 9$ rats per group per time point). *Significant difference relative to the controls, $P < 0.05$.

T2DM group than that in the CON group (288.23 ± 9.18 min· μ U/mL vs. 171.22 ± 6.61 min· μ U/mL, $P < 0.05$) (Figure 3B).

ITT results

To investigate insulin sensitivity, we also performed an ITT at different time points (Figure 4). Insulin was administered intraperitoneally, and blood samples were collected to evaluate the glucose response. After the insulin

administration, we found that the glucose concentrations in the CON group significantly decreased at 30 min ($P < 0.05$). However, the glucose concentrations in the T2DM group increased slightly and then declined slowly over the first 30 min. After the first 30 minutes, the slopes of the curves in two groups presented alike. Thus, rats that had received an STZ injection developed a condition that was comparable to T2DM with insulin resistance and hyperglycemia. In conclusion, according to the afore-

Surgical vein grafting model in diabetic rats

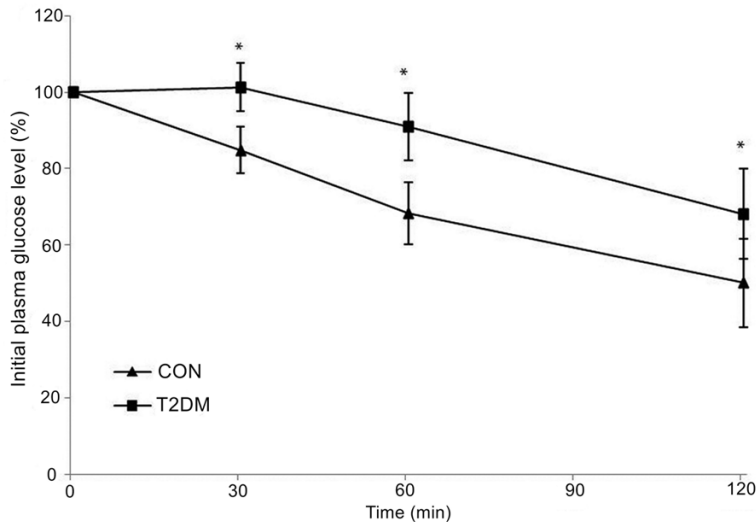


Figure 4. Glucose response during an insulin tolerance test in the diabetic (T2DM) and control (CON) groups at the 3rd day after a streptozotocin injection. The data are shown as the mean \pm standard error ($n = 7$ rats per group per time point). *Significant difference relative to the controls, $P < 0.05$.

mentioned data, we successfully developed a rat model with T2DM.

Surgical outcomes

Forty-four of the 60 animals that received a vein graft survived until their designated time of harvest. In the T2DM group, four rats did not establish the graft model, two rats died from perioperative bleeding at the anastomotic site, two rats died due to anesthetic accidents, and another four rats died after the operation from infection and ankyleron. In the CON group, four rats died during the study. Therefore, 36 surviving animals were included in our analysis ($n = 18$ each in the CON and T2DM groups). The duration of diabetes for the rats that were used in these experiments was approximately four weeks.

Thickness of the intima and media in the vein grafts

Representative histological sections of external jugular veins and vein grafts from the CON group were shown in **Figure 5**. The thickness of the vessel wall increased significantly in the 14- and 28-day vein grafts as compared to that in the 0-day, and neointima hyperplasia progressed throughout all time points in the CON and T2DM groups. However, the thickness of the neointima in the vessel walls of the T2DM rats was significantly greater than that in the

CON group ($P < 0.01$; **Figure 5B, 5C, 5E, 5F**). We also microscopically evaluated neointima thickness (**Figure 6**). Thickening of the vein grafts began as early as the day of the surgery, although this was not significantly greater than that observed in the CON group ($P = 0.101$, $n = 18$). During the vein graft remodeling, 14.7% (14-day) and 15.5% (28-day) increases in the vessel wall thickness were observed in T2DM rats compared to the controls (**Figure 6A**) (14-day: $386.29 \pm 126.67 \mu\text{m}$ vs. $336.77 \pm 114.94 \mu\text{m}$, $P < 0.05$, $n = 12$; 28-day: $463.60 \pm 200.19 \mu\text{m}$ vs. $401.37 \pm 173.05 \mu\text{m}$, $P < 0.05$, $n = 6$; respectively).

Cell nuclei content of the intima and media in vein grafts

We then evaluated 100- μm sections of the grafted vessels and counted the number of nuclei in the intima and media. In the CON group, the total cell numbers had increased significantly in the 14-day grafts and were markedly higher in 28-day grafts (**Figure 6B**); similar results were observed in the T2DM group. Comparable nuclei contents were observed in both groups at day 0 (T2DM: 32 ± 10 , CON: 29 ± 9 ; $P = 0.159$, $n = 18$) (**Figure 6B**). However, when we compared the 14-day and 28-day grafts for both groups, a significantly larger number of cells was observed in the T2DM group as compared to that in the CON group (14-day: 145 ± 52 vs. 78 ± 17 , $P < 0.05$, $n = 12$; 28-day: 205 ± 79 vs. 122 ± 36 , $P < 0.05$, $n = 6$, respectively).

Discussion

In the present model, the rats developed a condition that could parallel the development of T2DM in humans where the decline in hyperinsulinemia was not able to compensate for insulin resistance and subsequently resulted in hyperglycemia. In the second part of this study, we evaluated whether this model could be used to assess neointimal formation after surgical vein grafting in a clinically and physiologically

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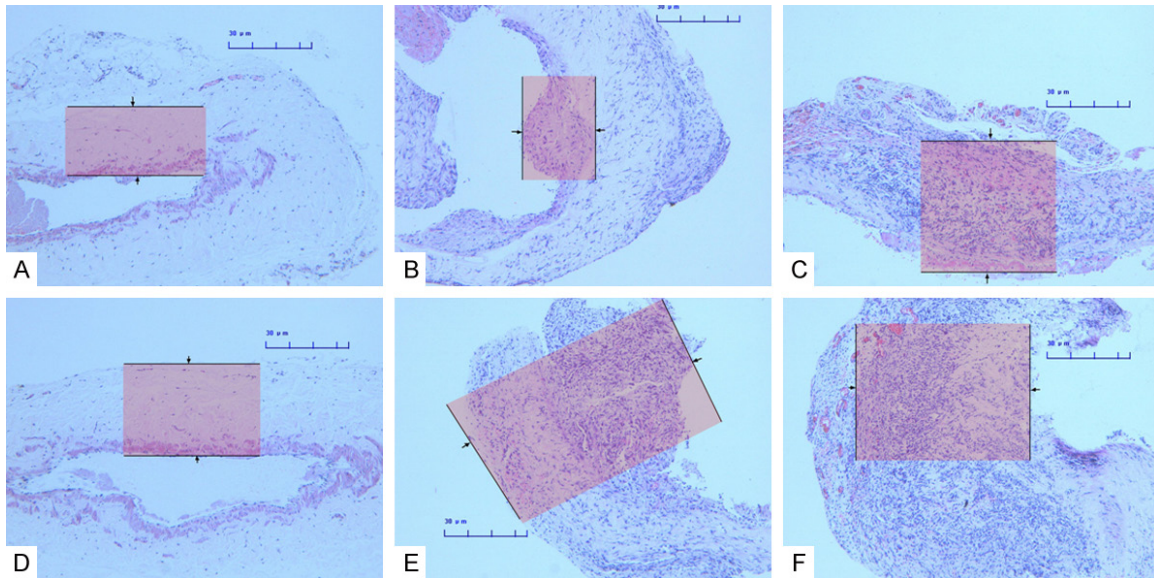


Figure 5. Representative photomicrographs of the control veins and vein grafts. Under anesthesia, the vena cava segments were grafted into the carotid arteries of (A-C) the control and (D-F) diabetic rats. The animals were sacrificed on (A, D) 0th day, (B, E) 14th day, and (C, F) 28th day after surgery, and the grafted veins were fixed in 4% phosphate-buffered (pH 7.2) formaldehyde, embedded in paraffin, sectioned, and stained with hematoxylin and eosin. Arrowheads indicate the wall thickness and lumens, respectively, of the (A, D) control vessels and (B, C, E, F) vein grafts. Bar = 30 µm, original magnification: 100×.

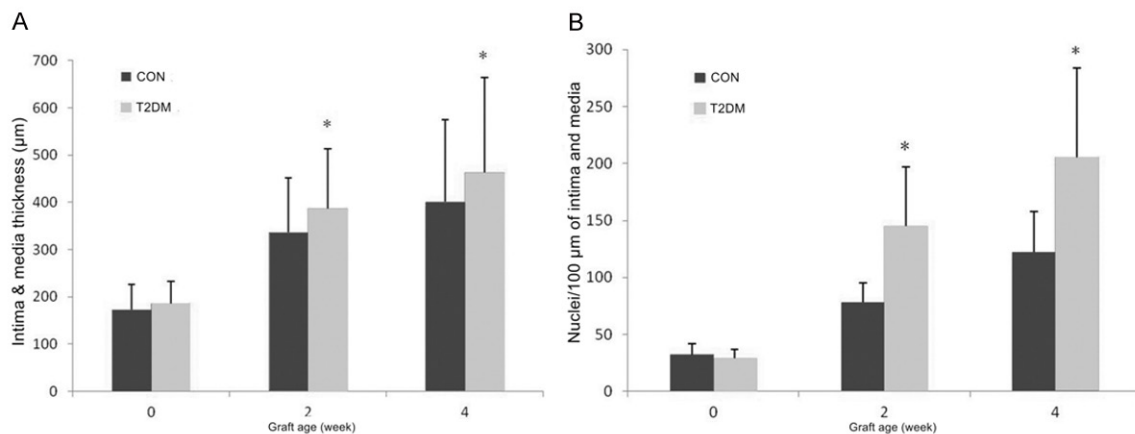


Figure 6. Neointima thickness and nuclei counts in the vein grafts. A. The intima and media thickness in the grafts from the control and diabetic rats (mean ± standard error). B. The total number of nuclei in 100-µm segments of the vein grafts (mean ± standard error). The data were obtained from 6-8 animals per time point. Four regions from each section were counted, and five sections per animal were selected. *Significant difference relative to the controls; $P < 0.05$.

relevant manner. However, according to recent research, Koshizaka M. et al. found that it was not clear whether diabetes had an influence on graft stenosis or failure [19]. On the contrary, our results strongly suggested that this model could accurately parallel the development of T2DM and that significant neointimal formation could be observed in 4 weeks after the vein graft.

Autologous vein grafts remain the only surgical alternative for many types of vascular reconstruction, although the 1-year failure rate is approximately 20% [20]. Various researchers have demonstrated that vessel restenosis contributed mostly to the failure of surgery, and several specific cellular mechanisms have been proposed [21-23]. For example, Davies et al. have reported that mononuclear

cell infiltration, smooth muscle cell proliferation, and extracellular matrix deposition are the hallmarks of neointima lesions, which can eventually lead to vessel restenosis [24]. However, the pathogenesis of vessel restenosis remains poorly understood, and no successful clinical interventions have yet been identified. Therefore, several autologous vein graft animal models have been specifically developed to generate lesions that resemble human vein graft restenosis.

Interestingly, T2DM can result in hyperglycemia, insulin resistance, hyperinsulinemia, hyperlipidemia, and hyperhomocysteinemia, and these conditions can subsequently give rise to endothelial and vascular dysfunction. Therefore, specific studies of a pre-clinical model that combines both DM and vein graft restenosis are urgently required, preferably in a way that closely simulates the natural progression and metabolic characteristics of T2DM and vessel disease. To date, an extensive range of T2DM animal models has been developed, including non-human primates, pigs, dogs, rabbits, and rats. Furthermore, the well-defined and easily manipulated genetic systems in mice have generated additional interest in creating murine models of the T2DM condition. To date, only one murine model has been established to study the mechanisms of neointimal formation in type 2 diabetic mice [14]. However, though this available C57BL/6J strain model could develop insulin resistance, it is suboptimal due to the lack of β cell failure that should be observed in type 2 diabetic patients and animal models. In contrast, even proliferation of β cells could be found in the C57BL/6J strain model in the report of Salzberg et al. [14]. Moreover, the cost of the aforementioned murine model is relatively high as compared to standard laboratory rats. Therefore, we sought to develop a rat model of surgical vein grafting in T2DM that requires no genetic manipulation and also attempted to find the relationship between T2DM and neointima formation. We indeed developed a novel rat model to combine surgical vein grafting and T2DM together in the current study.

Reduced insulin sensitivity and impaired pancreatic function are two major factors in the pathogenesis of type 2 diabetes. In the present study, we treated the rats with a low dose of STZ, which induced a characteristic increase in insulin resistance, with subsequent mild β cell impairment [25]. After the STZ injection,

we observed a 3-fold greater hyperglycemic response in the T2DM group compared to the controls, and this response rapidly increased to a peak at approximately 90 min. These findings imply that that insufficient insulin secretion in our model cannot compensate for the failure of the pancreatic β cells. Furthermore, the peak insulin levels were reached at approximately 45 min in the T2DM group, and these insulin levels remained elevated throughout the test. Therefore, these findings indicate that the decreased insulin secretion was exclusively related to the lack of β cells. To determine the insulin sensitivity of the model, we also subjected the rats to an ITT. In that test, the CON rats exhibited a significant reduction in their glucose levels within the first 30 min, while there was a slow or non-existent decrease over this period in the T2DM rats; however, the slopes of both glucose response curves were similar after 30 min. It appears that the T2DM group could not respond quickly to the insulin injection due to their reduced insulin sensitivity. Therefore, our findings confirm that the rat model closely mimics the natural course (from insulin resistance to β cell dysfunction) and metabolic features of T2DM.

Ideally, a rat model of vein graft disease will reproduce the flow and pressure characteristics that are found in the clinical setting. Therefore, we sought to evaluate the suitability of the T2DM model, and we selected to perform the insertion of the external jugular vein segment into the common carotid artery to ensure maximum simplicity and reproducibility. In addition, we have established a model in which a hemodynamic steady state was achieved by subjecting the vein graft to a continuous high-pressure arterial flow that would ensure that the cellular and molecular responses were solely responsible for the neointimal formation.

Surgical, traumatic, or ischemic injury may contribute to the lesion formation in the first post-operative month, and this pathophysiology is primarily responsible for the altered hemodynamics that lead to endothelial damage and dysfunction [26]. Therefore, in many respects, the morphological features of our vascular graft model resemble those of human venous bypass graft disease [27, 28]. In this context, medial thickening is a complex and dynamic process that involves endothelial cell dysfunction, leukocyte infiltration, vascular smooth muscle cell apoptosis, migration, and proliferation [29, 30].

Our results confirm this mechanism, as intima-media thickness of the vein grafts increased significantly in both the T2DM and CON groups and analogous results were observed when we evaluated the total cell numbers in both groups. However, the thickness of the vein graft was greater in the T2DM group as compared to the CON group, and a larger number of cells were also observed in the T2DM group. Therefore, we suspect that the increased levels of the plasma glucose in type 2 diabetic rats may lead to increased neointimal formation and that this mechanism also seems to be responsible for the neointimal increases that we observed in our rat model. Therefore, we conclude that the T2DM-like condition may have contributed to the subsequent vein graft restenosis.

This study contains an important limitation that should be considered in developing future research. We currently cannot establish an ideal model to simulate the process of graft-induced arteriosclerosis in humans with T2DM, as foam cell accumulation and necrotic core formation in the intima can be found in human vein grafts beyond the 1st year after bypass surgery [28, 30]. Regrettably, the life span of our model is only 3 months, which precludes any analysis of late-stage atherosclerosis.

Taken together, we successfully created a novel model of surgical vein grafting in type 2 diabetic rats. This model is cheap and simple to create, and it can provide an accurate recreation of T2DM and vein graft disease. Furthermore, future research regarding the pathogenesis and treatment of vein graft disease will likely benefit greatly from the current rat model.

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Disclosure of conflict of interest

None.

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