

Original Article

Saphenous vein harvested by endoscopic and conventional open methods in coronary artery bypass grafting: a comparative study based upon histological analysis

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Abstract: Objective: This study aimed to compare the effects of open vein harvest (OVH) and endoscopic vein harvest (EVH) on injury of greater saphenous veins based upon histological analysis. Methods: 20 patients who underwent coronary artery bypass grafting (CABG) were enrolled during September and December in 2014. Greater saphenous vein was selected as Blood Vessel Bridge. Those patients were divided into OVH group and EVH group. All the harvested veins satisfied with the requirements of Blood Vessel Bridge. Postoperative wound complications and continuity, integrity and uniformity of the tissue were evaluated using different staining methods. Results: The incidence of postoperative wound complications in EVH group was less than that in OVH group. There were no obvious differences on vascular injury of the saphenous vein intimal layer, elastic fiber layer, smooth muscle layer and outer membrane layer as observed under the light microscope. Conclusion: EVH method significantly reduces postoperative complications compared with OVH method, without remarkable injury of the saphenous vein. EVH is a safe and effective method for saphenous vein harvest.

Keywords: Open vein harvest, endoscopic vein harvest, coronary artery bypass grafting, greater saphenous veins

Introduction

Coronary artery bypass grafting (CABG) is one type of the most common cardiac-vascular surgeries [1]. Although internal mammary artery, radial artery and other arterial bridge have excellent patency rate, saphenous vein is still the widely used vascular material in CABG due to the distinguished advantages, including simple harvest, abundance source, high diam-

eter, and enough length for use [2]. Conventional open vein harvest (OVH) used to be applied to harvest enough length of saphenous vein, however with long scar and related complications in the incision. Moreover, the surgeries are always not satisfactory due to those disadvantages [3].

Endoscopic vein harvest (EVH) was firstly reported to assist the saphenous vein har-

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est by Lumsden in 1996 [4], which largely shortened the incision length. With the development of endoscopic technique, especially the increase of image resolution, the vein is now under great protection. In USA, EVH has already been widely applied in CABG surgery with a rate of 70% [5].

Saphenous vein is thought to be an important factor in determining the vascular patency rate [6]. However, the time consumed and tissue injuries during vein harvest seriously affect the long-term patency rate. There were some reports indicating that EVH could not completely replace the OVH method. Especially, no-touch technique used in OVH is beneficial for membrane protection during the surgery [7-10]. However, the application of OVH and EVH were still in debate [11-13]. Histological analysis is one of the most straightforward ways to observe the tissue injury. Although there were some studies using histological analysis to compare the effects of OVH and EVH on tissue injury, the conclusion is still inconsistent [14, 15]. Therefore, the results might directly affect the application of endoscopic assisted or conventional open methods in the surgery of coronary artery bypass grafting. In this study, we comparatively investigated the histological difference of EVH and OVH in saphenous vein harvest.

Materials and methods

Study design

20 cases of patients who underwent off-pump coronary artery bypass grafting surgery were enrolled in Department of Cardiac Surgery, Beijing AnZhen Hospital, the Affiliated Hospital of Capital Medical University (Beijing, China) from Sep, 2014 to Nov, 2014. Greater saphenous vein was used as the grafting material. The patients were divided into two groups based upon surgery protocol: EVH group (n=10) and OVH (n=10). EVH group underwent saphenous vein harvest assisted by endoscopic technique. Patients in OVH groups underwent saphenous vein harvest using the conventional open method. During the surgery, the region of lower limb without saphenous varicose vein was selected. All the veins were harvested by the same surgeons and satisfied with the requirements for the materials for off-pump coronary artery bypass grafting sur-

gery. The written consent was obtained from each patient and the experiments were under the approval of ethics committee of Capital Medical University.

VH endoscopic vascular collection system and visual system (Beijing Midaosi Co., Ltd, Beijing, China) were applied in the vein harvest. Based on the requirement for the vein bridge length in the surgery, a 1.5-cm length incision up or down the knee were selected to expose the saphenous vein. A 5 cm length tunnel was formed to include the casing with balloon. 20 mL air was injected into the casing. CO₂ system was connected and approximately 4-6 L/min of CO₂ was continuously insufflated to maintain a pressure of 12-15 mmHg. The mean harvesting time of CT-EVH was 48 min. For the standard conventional open technique, saphenous vein was exposed by a longitudinal leg incision starting from the medial malleolus and ending at the upper medial thigh at the saphenofemoral junction. The saphenous vein was dissected free from its perivascular fat pedicle and visible side branches were ligated and divided. Direct contact by surgical instruments and distension was avoided to prevent endothelial damage and spasm. The mean harvesting time of OVH was also 48 min.

The surgery complications of the incision and histological analysis were evaluated.

Histological analysis

Different stains were applied to observe the pathological performance, in order to evaluate the injury degree of the vein intimal layer, elastic fiber, smooth muscle layer and the outer layer. After staining, the continuity, integrity and uniformity of each layer were evaluated. In brief, the harvested veins were fixed in 10% formaldehyde and underwent paraffin embedding, sectioning into 5 μm by a Leica microtome (Leica, Germany). 20 saphenous veins from each group were applied for haematoxylin and eosin (HE), CD31, CD34, CD68, Alcian blue/periodic acid-Schiff (AB/PAS), Desmin, SMA, elastic fibers-VG and Masson 9 staining. Vein intima layer, elastic fibers, smooth muscle layer and outer membrane layer were observed under light microscope. The injury of endothelium was quantified as: 1 was defined as injury

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Table 1. The risk factors for incision infection (n=10 in each group)

	EVH	OVH	P
Age (Mean ± SD, years)	62.6 ± 6.4	61.9 ± 8.4	0.836
Sex (F/M)	4/6	5/5	0.653
Hypertension (n)	4	6	0.369
Hyperlipemia (n)	5	3	0.359
Diabetes (n)	4	3	0.639
Peripheral angiopathy (n)	2	1	0.528
Smoking history (n)	2	1	0.528
Operation history (n)	2	3	0.605
Preoperative TG (Mean ± SD, mM)	1.24 ± 0.36	1.39 ± 0.52	0.485
Preoperative TCHO (Mean ± SD, mM)	3.83 ± 0.61	4.21 ± 1.50	0.467
Preoperative LDL-C (Mean ± SD, mM)	2.23 ± 0.73	2.69 ± 1.47	0.391
Preoperative HDL-C (Mean ± SD, mM)	1.20 ± 0.26	1.05 ± 0.25	0.195

F/M: female/male; TCHO: total choline compounds; LDL-C: low-density lipoprotein cholesterol; TG: triacylglycerol; HDL-C: high-density lipoprotein cholesterol.

less than 10%; 2 was injury less than 25% but higher than 10%; 3 was injury between 25% and 50%; 4 was injury higher than 50%. The quantification was completed by one researcher in Pathology Department.

Statistical analysis

Data were presented as mean and standard deviation (S.D.) and analyzed by student t test and Chi-square test. $P < 0.05$ was considered as significant difference. The data was analyzed by SPSS 19.0.

Results

The risk factors for incision infection

All patients underwent coronary artery bypass surgery without cardiopulmonary bypass. There was no surgery death and all patients had excellent recovery. As listed in **Table 1**, the age and sex in the two groups were comparable ($P > 0.05$). There was no significant difference between the two groups regarding the prevalence rates of hypertension, hyperlipidemia, diabetes mellitus, peripheral vascular disease, smoking and surgery history, preoperative blood lipid level ($P > 0.05$).

Postoperative incision complications

One case in OVH group has incision complication and underwent a second round debridement surgery. Scar was found in five cases from

OVH group. By contrast, no surgery complication was found in EVH group. All patients in EVH had excellent recovery.

Histological analysis

Haematoxylin and eosin (**Figure 1A**), CD31 (**Figure 2A**), CD34 (**Figure 2B**), CD68 (**Figure 2C**), alcian blue-periodic acid schiff (AB-PAS) (**Figure 1B**), Desmin (**Figure 1C**), SMA (**Figure 1D**), elastic fibers/ vein graft (VG) (**Figure 1E**) and Masson's trichrome staining (**Figure 1F**) were applied to evaluate the continuity, integrity and uniformity of saphenous vein. As shown in **Table 2**, there was no

significant difference regarding vein intima layer, elastic fibers, smooth muscle layer and outer layer in the two groups ($P > 0.05$).

Discussion

Many studies have shown that, OVH was beneficial to reduce wound related complications, improve patient satisfaction, shorten hospitalization days, and reduce the pain of the wound compared with EVH [16, 17]. Some scholars believe that EVH is an independent factor for vein-graft failure and adverse clinical outcomes in a one-year post surgery study [18] and even leads to endothelial cell damage [19], although coronary angiography showed that the venous patency rate and histological performance in both groups were comparable 6 month post-surgery [20, 21]. However, in a follow-up study, association between EVH and mid-term adverse outcomes was not found in EVH group. Moreover, EVH eliminated the rate of re-hospitalization due to acute coronary syndrome [22]. In the present study, we also found a significant reduction in the incidence of incision complications in EVH compared to OVH group.

The long-term patency rate of vascular bridge is the most important evaluation index for CABG surgery. In addition to the vascular anastomosis technique and coronary artery conditions, it has been clear that the vascular graft stenosis or occlusion is related to the venous acquisition and treatment process [23]. The

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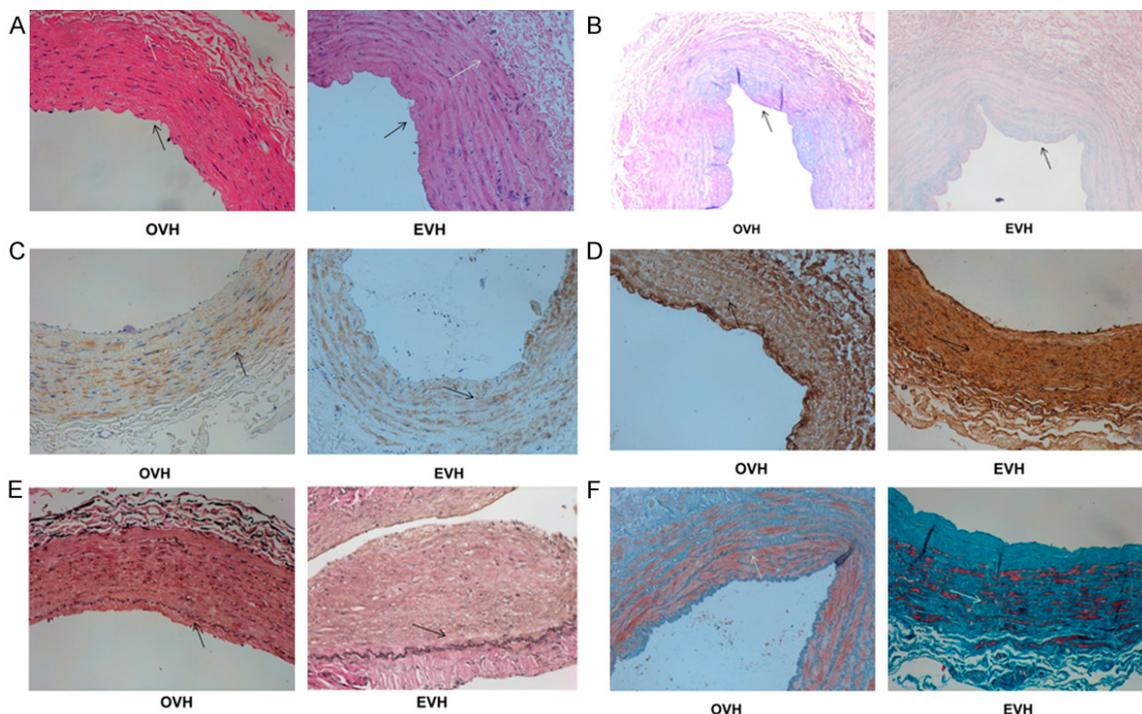


Figure 1. The pathological features of saphenous vein through different stains. A: Haematoxylin and eosin (HE). Black arrows indicate intima membrane layers. White arrows indicate outer membrane layers. B: AB-PAS staining. Black arrows indicate intima membrane layers. C: Desmin staining. Black arrows indicate smooth muscle layers. D: SMA staining. Black arrows indicate smooth muscle layers. E: Elastic fibers/vein graft (VG) staining. Black arrows indicate elastic fibers layer. F: Masson's trichrome staining. White arrows indicate smooth muscle layers.

injury of endothelial cells damages the original anticoagulant properties of normal blood vessels [24, 25]. Studies have shown that, after vascular endothelial injury, the exposure of endometrium tissue in the blood, promote the deposition of platelet and fibrin, leading to chronic inflammatory repair process, accompanied by fibrosis and myoepithelial cell proliferation [26]. Animal experiments have confirmed that the endothelial damage would activate the tissue factor of vascular wall [27]. Therefore, it is considered that the damage of intimal layer integrity is one of the important factors determining the thrombosis of the greater saphenous vein, intimal hyperplasia and vascular sclerosis [28].

Meyer and other scholars found that the effects of endoscopic assisted and traditional whole incision on endothelial injury were comparable through vWT and CD34 immunofluorescence staining [29]. Our present study stained the intima by HE, CD31 and CD34. CD31 is specifically expressed in the cytoplasm of vascular endothelial cells while CD34 is mainly

expressed in vascular endothelial cells. In addition, elastic fiber/VG staining mainly showed vascular elastic fiber layer [30]. Masson staining determined the vascular smooth muscle and connective tissue. SMA is equally distributed in vascular smooth muscle. Desmin is an intermediate filament protein, which plays an important role in the connection of nucleus and plasma membrane. The phenotype, proliferation and cytoskeletal protein expression of vascular smooth muscle play important roles in the maintenance of cell morphology, contractile function and vascular remodeling [31]. In addition, CD68 is an inflammatory factor, which originated from macrophages. It was reported that CD68 expression was increased in the cytoplasm of the blood vessel when the blood vessel was stimulated by inflammatory reaction [32, 33]. AB-PAS staining was positive in the tissue with mucus-like degeneration [34]. Through the above staining, our study observed saphenous vein intimal layer, elastic fibers, smooth muscle layer, outer layer of four structural levels, and expression of inflammatory factors and mucus degeneration in both two groups. The

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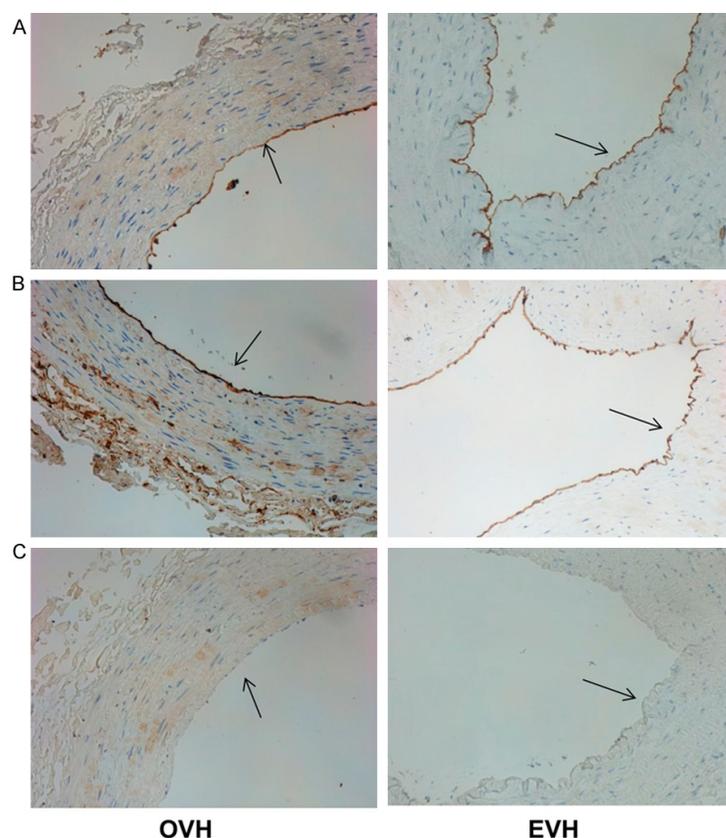


Figure 2. The saphenous vein intima layer pathological performance through different stains. CD31 (A), CD34 (B) and CD68 (C) staining. Black arrows indicate intima membrane layers.

Table 2. Comparison of the injury of vein intima layer, elastic fibers, smooth muscle layer and outer membrane layer in the two groups (mean and SD, N=10 in each group)

Groups	Intima layer	Elastic fibers	Smooth muscle layer	Outer membrane layer
EVH	1.9 ± 0.9	2.8 ± 0.6	1.8 ± 0.6	3.2 ± 0.6
OVH	1.6 ± 0.8	2.5 ± 0.5	1.6 ± 0.5	3.3 ± 0.5
P	0.445	0.264	0.449	0.696

results showed that there was no significant difference in two groups regarding the injury of saphenous veins. However, this study was restricted by the relatively small sample size. In order to further understand the effects of EVH and OVH on saphenous vein grafts, future large sample size study, in combination with new research methods is required.

Conclusion

Our study demonstrates that saphenous vein harvesting by endoscopic significantly decre-

ases postoperative wound related complications compared with traditional open method. Especially, there was no significant difference in the two kinds of acquisition of great saphenous vein. Endoscopic saphenous vein is feasible and safe in clinical practice.

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Disclosure of conflict of interest

None.

Authors' contribution

Tie Zheng and Xuejun Sun contributed to the conception and design of the study; Hao Ding, Jianfeng Shang and Hui Li contributed to the acquisition of data; Ming Gong, Haiyang Li, Shuai Zhu and Dong Chen contributed to the analysis of data; Tie Zheng wrote the manuscript; All authors reviewed and approved the final version of the manuscript.

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