Journal of Visualized Experiments

Synergizing antegrade endoscopic with bridging vein harvesting may advance great saphenous vein graft quality from the lower leg

--Manuscript Draft--

Article Type:	Invited Methods Article - JoVE Produced Video
Manuscript Number:	JoVE59009R2
Full Title:	Synergizing antegrade endoscopic with bridging vein harvesting may advance great saphenous vein graft quality from the lower leg
Keywords:	Coronary artery bypass graft, peripheral bypass grafting, long-term patency, graft thrombosis, tissue damage, endoscope, optical dissector, vein harvest technique.
Corresponding Author:	Christian Klopsch Universitatsmedizin Rostock Rostock, GERMANY
Corresponding Author's Institution:	Universitatsmedizin Rostock
Corresponding Author E-Mail:	christian.klopsch@med.uni-rostock.de
Order of Authors:	Christian Klopsch
	Alexander Kaminski
	Friedrich Prall
	Pascal Dohmen
Additional Information:	
Question	**Response**
Please indicate whether this article will be Standard Access or Open Access.	Standard Access (US$2,400)
Please indicate the **city, state/province, and country** where this article will be **filmed**. Please do not use abbreviations.	Rostock, Mecklenburg Western Pomerania, Germany

Christian Klopsch, MD.
Rostock University Medical Center
Schillingallee 35, 18057 Rostock, Germany
phone: +49 381 494 61 09, fax: +49 381 494 61 02
E-mail: christian.klopsch@med.uni-rostock.de

Editorial Office of the Journal of Visualized Experiments

February 18th, 2019

Ms. No.: JoVE59009R1 (invited manuscript)
Title: Synergizing antegrade endoscopic with bridging vein harvesting may advance great saphenous vein graft quality from the lower leg

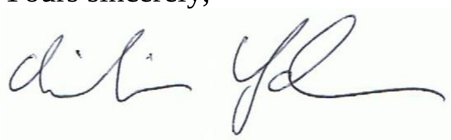
Manuscript re-submission

Dear Prof. Dr. Phillip Steindel,

Thank you for considering our revised manuscript for publication in Journal of Visualized Experiments. The manuscript was revised following the editorial and reviewer comments.

We hope that the paper will now find your approval for publication in the Journal of Visualized Experiments. All authors have read and approved the manuscript.

Yours sincerely,



Christian Klopsch, MD.

TITLE:

Synergizing Antegrade Endoscopic with Bridging Vein Harvesting for Improvement of Great Saphenous Vein Graft Quality from the Lower Leg

AUTHORS & AFFILIATIONS

Christian Klopsch[1], Alexander Kaminski[1], Friedrich Prall[2], Pascal Dohmen[1,3]

[1]Department of Cardiac Surgery, Heart Center Rostock, Rostock University Medical Center, University of Rostock, Rostock, Germany
[2]Institute of Pathology, Rostock University Medical Center, University of Rostock, Rostock, Germany
[3]Department of Cardiothoracic Surgery, Faculty of Health Science, University of the Free State, Bloemfontein, South Africa

Corresponding Author:

Christian Klopsc (christian.klopsch@med.uni-rostock.de)

Email Addresses of Co-Authors:

Alexander Kaminski (alexander.kaminski@med.uni-rostock.de)
Friedrich Prall (friedrich.prall@med.uni-rostock.de)
Pascal Dohmen (pascal.dohmen@med.uni-rostock.de)

KEYWORDS:

coronary artery bypass graft, peripheral bypass grafting, long-term patency, graft thrombosis, tissue damage, endoscope, optical dissector, vein harvest technique.

SUMMARY:

Presented here is a protocol for antegrade endoscopic vein harvesting from the lower leg, which can safely be introduced in routine coronary artery bypass grafting. Vein grafts present excellent graft quality following this standardized protocol with positioning of the legs, minimally invasive access to the vein, and antegrade endoscopic vein harvesting.

ABSTRACT:

Antegrade endoscopic harvesting of autografts for bypass grafting may be an optimal strategy addressing excellent graft quality and reduced post-operative wound complications. This standardized protocol for antegrade endoscopic vein harvesting (EVH) from the lower leg has the potential to be introduced to routine coronary artery bypass grafting (CABG). Patients undergoing CABG surgery are positioned on a surgical table with two additional foam rollers below the extended legs, enabling antegrade EVH from the lower leg. Following minimally invasive surgical access through a bridging vein harvest technique, an endoscopic optical dissector is inserted antegrade into the wound. The main vessel and side branches are dissected under continuous optical control of vein quality status and the working channel. After, an endoscopic optical retractor is inserted with an internal bipolar electrocoagulation device for precise, safe, and tissue-protective interruption of side branches. After release of the vein, the vessel is cut off at the proximal and distal ends under optical control, retrieved from the wound, then cannulated and flushed with heparinized saline. Finally, all side branches of the vein graft are double-clipped. Vascular histology is analyzed in a randomized selection of vein samples. After applying this standardized EVH protocol, the learning curve was shown to be steep, and graft quality was sufficient for coronary artery bypass grafting in every case. There was no conversion to surgical harvesting and low risks for tissue damage and bleeding. Leg positioning and synergizing EVH with bridging vein harvesting improved procedural success and vein graft quality. In our hands, antegrade EVH from the lower leg was feasible, demonstrating straightforward graft dissection as well as adequate macroscopic and microscopic graft quality with preserved endothelial integrity. In conclusion, the introduced technique is safe, shows excellent vein autograft quality, and illustrates feasibility for elective and urgent isolated CABG and combined CABG scenarios.

INTRODUCTION:

Open atraumatic “low-touch” and "no-touch" techniques have been developed over the years for harvesting saphenous veins in coronary artery bypass graft (CABG) surgery or peripheral bypass grafting, producing grafts with excellent endothelial integrity and long-term patency. However, wound complications remain a major problem when using the open technique, especially in obese, diabetic, and chronic venous insufficiency patients[1-4]. The question arises of how physicians can harvest the saphenous vein with optimal graft quality and reduced risk for wound complications. Endoscopic vein harvesting (EVH) techniques have been proven to be cost-effective, and clinical outcome parameters are comparable with the open technique. However, strategies protecting endothelial integrity, histological structure, and physiological function of vein grafts during EVH are highly appreciated in order to preserve optimal graft quality[2]. Recent studies have presented superior graft patency after open harvesting compared to endoscopic techniques[5]. It has also been shown that bridging vein harvest techniques can directly improve vein quality[6]. Therefore, it is hypothesized that vein graft harvesting may be advanced through synergizing antegrade EVH with minimally invasive bridging vein harvesting, specific leg positioning, and vein isolation in a tensionless working channel.

To date, conventional EVH techniques for harvesting great saphenous veins have used antegrade approaches for the upper leg and retrograde approaches for the lower leg. However, we have experienced limitations of these techniques and hold concerns about graft quality. The great saphenous vein from the knee and upper leg frequently have revealed numerous side branches and occasionally shown dilated vessel diameter, leading to impaired vessel quality and mismatching of conduit and target vessels that can negatively affect long-term graft patency after CABG and re-revascularization rate[7-11]. In our experience, the retrograde EVH approach for the lower leg has repetitively resulted in prolonged blood stasis inside the vessel (with augmented intravenous blood pressure due to closed venous valves), increased mechanical stress on the tissue, bleeding, thrombus formations, graft damage, and impaired graft quality. Consequently, this standardized protocol was developed for safe antegrade EVH from the lower leg, combining the bridging vein harvest technique for minimally invasive access site with antegrade EVH in a tensionless working channel for adequate vein graft quality.

PROTOCOL:

The study conforms to the Declaration of Helsinki. The protocol follows the guidelines of an independent institutional ethics committee, and human biomaterials were obtained after informed written consent (ethics committee approval: A 2018-0037).

1. Positioning of the legs

NOTE: Patient inclusion criteria included a history of coronary artery disease with elective/urgent indication for CABG surgery and the need for harvesting of at least one venous bypass graft for complete revascularization. Patients with debilitating chronic disease, emergency operations, status post-deep vein thrombosis, and active wet gangrene were excluded. Pre- and post-operative procedures were comparable with previously described clinical studies[12,13]. 28 patients undergoing coronary artery bypass graft surgery were included for antegrade endoscopic vessel harvesting of 30 great saphenous veins from the lower leg after informed written consent. A cardiac surgeon certified and significant practical experience (>200 cases) executed the vein harvesting with antegrade endoscopic vessel harvesting of the great saphenous veins from the upper legs.

1.1. Organization of the surgical theatre

1.1.1. Before surgery, ensure supine position of the anesthetized patient on the surgical table following institutional standard procedures for CABG surgery.

1.1.2. Place the vein harvester on the right side of the patient. Place the surgical team and instrumental set-up for cardiac surgery on the left side of the patient. Place the instrumental set-up for EVH near the end of the table (**Figure 1**, **Figure 2**; see **Table of Materials**).

1.2. Specific positioning of the legs

1.2.1. Place two foam rollers (length: 60 cm, diameter: 12 cm) below the extended legs. Place one half-cylindrical foam roller just above the knee in order to avoid overstretched knees and common peroneal nerve lesions. Then, place another full cylindrical foam roller below the Achilles tendon for lifted and outward rotated foot position (**Figure 1A–D**).

2. Minimally invasive surgical access to the vein graft

2.1. Access site

2.1.1. Use institutional standard disinfection procedures with Octenidindihydrochlorid followed by standard sterile covering for aseptic surgical conditions.

2.1.2. Make one longitudinal skin incision (length: 1.5–2 cm) with a curved scalpel (size 10) on the lower leg. Start the incision with the distance of approximately one index finger above the imagined ankle joint and proceed upwards parallel to the medial margin of the tibia bone (**Figure 2C**).

2.2. Bridging vein harvesting technique

2.2.1. Obtain minimally invasive access to the great saphenous vein with surgical forceps, dissecting surgical scissors, and an electrosurgical pencil, if necessary. Starting from the skin incision, isolate the vessel 4 cm in each direction using a vessel loop, dissecting scissors, small soft tissue retractor, and Langenbeck hook, applying the standard bridging vein harvesting technique (**Figure 2C**).

2.2.2. Continuously check vein quality status and surrounding subcutaneous tissue in the working channel. Visualize in order to avoid injury to the saphenous nerve. Avoid harvesting of progressive varicose veins.

2.3. Practical hints

2.3.1. Make sure that one small finger can easily access the working channel (antegrade). Avoid surgical clipping of side branches at this time.

3. Antegrade EVH with the optical dissector

3.1. Insertion of optical dissector

3.1.1. Assemble the optical dissector by connecting an extended length endoscope (diameter: 7 mm, length: 48 cm) to an optical camera and dissection tip from the endoscopic vessel harvesting system, according to the manufacturer's instructions. Moisturize the optical dissector with saline containing heparin (i.e., NaCl + Hep: 5.000 per 200 mL).

3.1.2. Put the inflatable blocker balloon (also provided by the endoscopic vessel harvesting system and moisturized with NaCl + Hep) over the optical dissector. Gently insert the optical dissector (antegrade) and, after, the inflatable blocker balloon into the wound under permanent optical control of the vein (**Figure 2D–F**).

3.2. Dissection of the vein

3.2.1. Block the inflatable blocker balloon with room air (10 mL). Flood the working channel with CO_2 (flow: 5 L/min, pressure: 15 cm H_2O) and indicate to the anesthetic medical staff. Make sure that the working channel is extended by the gas pressure.

3.2.2. Move antegrade until reaching the imagined proximal medial end of tibial diaphysis using the optical dissector, following the manufacturer´s instructions. Gently dissect the main vessel from a majority of subcutaneous tissue until achieving clear identification of side branches.

3.2.3. For optimal results, dissect the main vessel through antegrade movement of the optical dissector 1) above of, then 2) below, the main vessel. Then, selectively dissect the side branches, with one side of the vein preserving the perivascular tissue as far as possible, followed by the other side (**Figure 2G–I**).

3.2.4. Continuously check vein quality status and mechanical stress in the working channel. Visualize in order to avoid injury to the saphenous nerve. Avoid harvesting of progressive varicose veins.

4. Antegrade EVH with the optical retractor

4.1. Insertion of optical retractor

4.1.1. Remove the optical dissector from the wound and disconnect the dissection tip.

4.1.2. Adapt the blocker balloon for the optical retractor and block the working channel with a 5 mL syringe. Assemble the optical retractor by connecting the extended length endoscope to the optical camera and retractor device from the endoscopic vessel harvesting system, which is provided with an internal bipolar electrocoagulation device (power output: level 3–4).

4.1.3. Use anti-fog fluid for the tip of the endoscope (**Figure 3A–C**). Again, moisturize the optical retractor with NaCl + Hep before antegrade insertion through the blocked balloon.

4.2. Isolation of the vein

4.2.1. Advance the optical retractor antegrade to the end of the working channel. Release the vein from the surrounding subcutaneous tissue with the retractor device and selectively interrupt side branches with the bipolar electrocoagulation device in a retrograde fashion (**Figure 3D–F**). Here, the bipolar electrocoagulation device must be positioned with the convex ending away from the main vessel.

4.2.2. Continuously check vein quality status and mechanical stress in the working channel. Visualize in order to avoid injury to the saphenous nerve.

5. Vein graft retrieval

5.1. Finishing of EVH

5.1.1. Execute a stab incision in the skin with a sharp scalpel (size 11) at the distal end of the dissected vein (regarding venous flow direction). Insert a smooth (anatomical) clamp through the stab incision and clamp the vein under optical control with the optical retractor.

5.1.2. Gently retrieve the clamped vein through the stab incision and cut it off proximally (regarding venous flow direction). Thereafter, gently remove the optical retractor through the blocked balloon simultaneously relieving the distal portion of the vein (**Figure 3G**). Deflate the blocker balloon and remove it from the wound.

5.1.3. Switch off CO_2 and indicate it to the anesthetic medical staff. At this time, use surgical clips and interrupt the remaining side branches before retrieval of the vein graft, if necessary.

5.2. Finishing of bridging vein harvesting

5.2.1. Execute a stab incision in the skin with a sharp scalpel (size 11) at the proximal end of the isolated vein approximately 3 cm above imagined ankle joint. Insert an anatomical clamp through the stab incision and retrieve the vein though the skin incision under digital and optical control. Visualize and avoid injury to the saphenous nerve.

5.2.2. Then, clamp the vein under direct vision and cut off distally (regarding venous flow direction). Thereafter, gently relieve the entire vein graft through the initial minimally-invasive surgical access site and cannulate the proximal end with a 3.0 mm flexible vessel cannula (**Figure 3H**).

6. Final preparation of the vein graft

6.1. Gently flush the released venous graft with NaCl + Hep (in a 10 mL syringe) alternating with double-clipping of all side branches (**Figure 3H**). Continuously check vein quality status and repair injuries, if necessary, with polypropylene sutures (7-0 or 8-0). Finally, a vein harvester and primary surgeon must evaluate graft quality of the endoscopic vein, applying the same criteria as is executed for veins harvested by the open technique.

6.2. If necessary, store the vein graft in a NaCl + Hep-moisturized compress at room temperature (RT) for short-term storage. However, avoid longer time periods of storage. Transfer the vein graft into heparinized blood as soon as arterial cannulation for cardiopulmonary bypass is accomplished.

7. Wound closure

7.1. Ligate the main vessel at both clamped vein ends, each with a 4-0 polyglactin 910 suture. Remove the clamps.

7.2. Insert a 10Fr Redon drain into the wound (**Figure 3I**). Fixate the Redon drain with 2-0 poly ethylene terephthalate suture at the skin.

7.3. Execute subcutaneous and intracutaneous wound closures at the minimally invasive access site with 2-0 and 4-0 polyglactin 910 sutures, respectively. Close the two small stab incisions at the proximal and distal ends, with one U-suture each, stitched intracutaneously (4-0 polyglactin 910). Drape the wounds with sterile plasters.

7.4. Wrap the leg, except in peripheral artery disease patients.

REPRESENTATIVE RESULTS:

A steep learning curve was demonstrated for an experienced cardiac surgeon performing antegrade EVH of the great saphenous vein from the lower leg (**Figure 4**). There were no conversions to surgical harvesting. However, there were four cases of vein injury in the beginning of the learning curve. In three of the four cases, major injuries occurred at the distal portion of the vein because of an inadequately narrow working channel when the surgeon isolated the vein above the tibial metaphysis. Disruption of a major side branch in two cases and dilaceration of the distal vein in one case were observed, leading to discarding of the distal portion of the vein. The remaining vein grafts were adequate for CABG in every case. In one of the four cases, minor injury was observed at two small side branches at the proximal portion of the vein, when surgical clipping of side branches was executed in the working channel before antegrade EVH. These injuries were repaired with polypropylene sutures. Final vein graft quality was macroscopically sufficient for CABG in every case regarding graft length, graft diameter, absence of injury, and vessel wall integrity.

In three randomly selected patients, antegrade EVH (as explained above) was extended by additional open vein harvesting from imagined tibial metaphysis upwards to the distal third of the upper leg. Vein samples (about 3 mm in diameter) used for these studies were taken from excess material near the ankle joint (bridging vein harvesting technique), imagined tibial metaphysis (open harvesting technique), and proximal medial end of the imagined tibial diaphysis (antegrade EVH technique). The samples were blinded to the participating pathologist, fixed in formalin, sectioned transversely, and embedded in paraffin by routine procedures. Hematoxylin and eosin-stained 5 µm sections were prepared for light microscopy, and CD31 immunostains were obtained to demonstrate further that endothelial cells and integrity remained intact.

These randomized blind microscopic analyses revealed an intact vascular morphology (**Figure 5A**,**B**) and completely preserved endothelial integrity (**Figure 5C**) in all analyzed vein samples after antegrade EVH as well as employed conventional alternatives. However, lack of experience and negligence for tissue preservation and vein quality can increase the risk for bleeding and graft injury. Therefore, a continuously optical vein quality control is highly recommended, as well as “tissue-gentle” vein isolation and preservation of especially perivascular tissue. In this regard, it should be noted that the lifted and outward-rotated foot position and a prolonged distance from the minimally-invasive access point to the ankle joint (one index finger) remarkably improved forward-downward movability of the endoscope. Consequently, synergizing antegrade EVH with bridging vein harvesting reduced mechanical stress and enhanced vein graft quality during antegrade EVH of the great saphenous vein from the lower leg.

Antegrade EVH from the lower leg was feasible, demonstrating straightforward graft dissection and adequate graft quality. No blood stasis, no thrombus formation, and low risk for bleeding and tissue damage were observed. Leg positioning and synergizing the antegrade EVH with bridging vein harvest technique were the two main factors leading to procedural success. Great saphenous vein grafts from the lower leg showed normal diameters (approximately 3–4 mm). After graft retrieval, veins usually demonstrated slight spasms, comparable to our institutional experiences for conventional vein harvesting techniques. Therefore, matching of vein grafts from the lower leg with cardiac target vessels was deemed appropriate for CABG. Thus, the introduced antegrade approach was applicable for both legs. In two cases, antegrade EVH from both lower legs was successfully executed. No wound complications were experienced in this small initial series. Patient acceptance of the method was high.

FIGURE LEGENDS:

Figure 1: Organization of surgical theatre and specific positioning of legs. (**A–C**) The instrumental set-up for EVH was prepared and placed near the end of the surgical table. (**D**) The instrumental set-up for cardiac surgery was placed on the left side of the anesthetized patient. Two foam rollers (dotted line, one half-cylindrical foam roller just above the knee, one full-cylindrical foam roller below the Achilles tendon) were placed below the extended legs. Foot position was lifted and outward-rotated for direct vision on imagined ankle joint (short line), expected minimally invasive access site (bold line), and medial margin of the tibial metaphysis (dashed line). a-z are described in the **Table of Materials**.

Figure 2: Minimally invasive surgical access and antegrade EVH allowed safe and unimpeded dissection of the great saphenous vein. (**A–C**) Following sterile draping of patients, the great saphenous vein from the lower leg was isolated and (**C**) looped through minimally invasive surgical access via the bridging vein harvesting technique. (**D–F**) The optical dissector was assembled under sterile conditions (**D**) and inserted antegrade through the inflatable blocker balloon (solid arrow) into the wound (**E**,**F**). (**G–I**) The protocol allowed for a simplified forward-downward movement (dotted arrow) of the optical dissector during EVH (**G,H**) without impeding the work of the primary surgeon and surgical nurse (**I**). aa-af are described in the **Table of Materials**.

Figure 3: Following antegrade EVH, all isolated vein grafts demonstrated adequate quality for CABG. (**A–E**) After removal of the optical dissector the working channel was blocked with a 5 mL syringe (**A,** arrow), the optical retractor was assembled, prepared with anti-fog fluid (**B**), and inserted antegrade through the inflatable blocker balloon into the wound (**C**). Again, the protocol allowed for a simplified and unimpeded forward-downward movement of the optical retractor during EVH (**D,E**). (**F–I**) An anatomical clamp was inserted into a stab incision (**F,** dotted arrow) and used to clamp the main vessel under endoscopic control before retrieval of the distal portion of the vein (**G**). Thereafter, another anatomical clamp was inserted into a stab incision at the proximal end of the isolated vein (**G**) followed by complete retrieval of the vein, proximal venous cannulation, clipping of side branches (**H,** dashed arrow), and wound closure (**I**). ag is described in the **Table of Materials**.

Figure 4: Learning curve for antegrade EVH from the lower leg. A total of 30 great saphenous

veins from the lower leg were isolated from 28 CABG patients using antegrade EVH. The graph illustrates an immediate dynamic reduction in time expenditure (min) starting from insertion of the optical dissector until termination of vein graft retrieval.

Figure 5: Antegrade EVH preserved integrity of endothelium and vascular wall. (**A–C**) Representative images from vein cross sections after antegrade EVH for hematoxylin/eosin staining (**A,B**), as well as for CD31 immunostaining (**C**), illustrated normal morphology of vascular wall and completely preserved endothelial integrity, respectively. No differences in histology were detected after comparing all harvesting techniques in the department (open vein harvesting, bridging vein harvesting, antegrade EVH).

DISCUSSION:

It should be stated that we prefer complete arterial coronary revascularization in our department. There is rising evidence that CABG using bilateral internal mammary artery (IMA) grafts can significantly improve long-term survival of patients[14-17]. However, there are valid reasons for a “single IMA plus vein grafts” strategy, especially in patients at advanced ages, patients with high risks for surgical site infection, patients in which the radial artery graft is not available, and in cases with chronically occluded coronary target vessels. In these scenarios, this protocol offers a standardized technique for safe antegrade EVH from the lower leg. Optimal vein grafts for CABG will reduce short-term wound complications and improve long-term outcomes with diminished revascularization and higher quality of life[1,2,8].

The protocol is based on three essential pillars: (1) specific positioning of the leg, (2) bridging vein harvesting technique for minimally invasive access site, and (3) antegrade EVH in a tensionless working channel. Sufficient leg positioning prevents interference of optical dissector and retractor with the food, guaranteed undisturbed EVH in the surgical theatre, allowed for easy graft preparation through forward-downward movement of the endoscope in the working channel, and reduced the risk of common peroneal nerve lesions. Sufficient bridging vein harvest technique simplified the insertion of endoscopic devices, reduced the tension in the working channel especially near the access site, and minimized the possibility of vein graft damage through improved movability of optical dissector and retractor. Moisturizing the EVH equipment with heparinized saline further simplified antegrade insertion of the EVH equipment as well as allowed for unimpeded endoscope movements. Instead of systemic heparin infiltration, heparin was administered locally in the working channel, which was sufficient here to prevent from both thrombus formation and bleeding. Most importantly, the tensionless working channel allowed for antegrade EVH with minimal risk for graft injury.

In this procedure, the surgeon’s attention must be concentrated on several aspects. The working channel must be extended by gas pressure before graft preparation. The optical dissector and retractor must be utilized to enlarge the working channel, and the vein harvester may need to enlarge the minimally invasive access site. To the greatest extent possible, the harvester should preserve perivascular tissue (especially near side branches of the vein graft) and be able to use the precise and tissue-protective bipolar electrocoagulation device for interruption of side branches through free movability of the endoscopic vessel harvesting system. In any case, it is recommended to avoid clips in the working channel before EVH. Harvesting beyond the imagined tibial metaphysis requires a higher level of experience, because the working channel became progressively narrow and the graft may be damaged.

Some specific tips for successful antegrade EVH include: Avoid bleeding. It is recommended to avoid a narrow working channel. If necessary, the surgeon should use fine surgical forceps to retract the skin incision during material insertion or enlarge the skin incision with the curved scalpel (and fixate the blocker balloon with an additional intracutaneous U-suture). It should be repetitively evaluated if the working channel is extended by the gas pressure and tension-free. The working channel should be enlarged with the optical dissector or retractor, if necessary. The side branches should be sufficiently dissected with the optical dissector. The integrity of the subcutaneous (especially the perivascular) tissue should be preserved as much as possible, addressing both graft injury prevention and maximal reduction of mechanical stress in the working channel. Surgeons should be aware of sufficient interruption of all side branches from the main vessel with the optical retractor. Additional skin incisions will be required if interruption of side branches was incomplete. Surgeons should be “tissue-gentle” through avoidance of extensive traction forces affecting the vein, especially in chronic venous insufficiency, diabetes, peripheral artery disease, obese, and elderly female patients. Antegrade EVH from the lower leg should be stopped, at the furthest, next to the imagined tibial metaphysis.

Following this protocol, the antegrade EVH approach enables “tissue-gentle” vein preparation at the lower leg, with adequate vein graft quality and low risk of wound complication, even in chronic venous insufficiency, diabetes, peripheral artery disease, obese, and elderly female patients[3,8-10]. Initial randomized histological analyses of vein grafts illustrated completely preserved endothelial and vessel wall integrity after all harvesting techniques in the department (open vein harvesting, bridging vein harvesting, antegrade EVH). Moreover, antegrade EVH from the lower leg illustrated vein grafts with low side-branching and adequate matching of conduit and target vessels during CABG. Therefore, it was hypothesized that this approach may be advantageous compared to the antegrade EVH approach from the upper leg in follow-up studies. Vein grafts harvested from the knee and upper leg usually show frequent side branching and occasional mismatching of dilated conduit vessels (diameters above 5 mm) with coronary target vessels, which can create more turbulent intraluminal flow conditions and lead to an increased risk for graft occlusion after CABG[11].

Further, the proposed technique may eliminate a majority of wound complications, especially after open vein isolation in the region of the knee where wound healing can be compromised due to tissue tension during post-operative mobilization of the patient. The manuscript lacks a systematic in-depth analysis of vein graft histology and its endothelial functionality. However, initial data point to possible non-inferiority of antegrade EVH compared to other harvesting techniques. The protocol emphasizes maximal protection of the surrounding subcutaneous (especially perivascular) tissue, addressing both preserved graft integrity and reduced mechanical stress in the working channel. Further studies are required on controlling the graft long-term patency. The small sample of patients may implicate that the learning curve is not finished. Therefore, the protocol for antegrade EVH from the lower leg was established and performed by surgeons experienced with the technique for the upper leg, which should guarantee sufficient vein graft quality[18,19]. Further investigation is warranted.

Antegrade EVH with the endoscopic vessel harvesting system produced additional peri-operative procedural costs during CABG and is not reusable. However, cost efficiency was proven, and the closed tunnel EVH system demonstrates multiple highly appreciated advantages over existing reusable or open EVH techniques[2,20]. In this set-up, a closed tunnel EVH system was chosen because it offers a long (maximum 35–40 cm), small lumen working channel and allows for antegrade EVH from the lower leg with only one minimally invasive skin incision. A single switch from optical dissector to optical retractor and the integration of directed, precise, and graft-protective electrocauterisation enabled a short time expenditure and optimized cost efficiency, quality of life, and vein graft histology[2]. Moreover, the antegrade approach can be applied for the full length of the leg if a second small incision (standard incision, recommended by the manufacturer) is added next to the imagined tibial metaphysis. As shown, antegrade EVH of the great saphenous vein can be repeated in the second lower leg. Apart from that, antegrade EVH of the small saphenous vein was enabled through specific positioning of the leg with lifted and inward-rotated foot position and lateral minimally invasive access site in two patients (unpublished data by A. Kaminski, 2018), comparable with the work of Rustenbach et al.[21]

The introduced antegrade EVH concept for the great saphenous vein from the lower leg illustrates feasibility for straightforward graft dissection and adequate graft quality, indicating a promising clinical perspective for routine use in CABG. The protocol has a steep learning curve in experienced cardiac surgeons. Besides the explained benefits, the antegrade EVH approach for the great saphenous vein from the lower leg displayed low risks for blood stasis, thrombus formation, bleeding, and graft injury, which was previously seen with the conventional retrograde approach. For these reasons, we had cancelled the retrograde approach earlier in patients before starting this study. However, there is a lack of data supporting a general assumption of superiority of the antegrade over the retrograde approach.

Absence of blood stasis (with augmented intravenous blood pressure because of closed venous valves) and limited mechanical stress on the vein may reduce the risk for graft injury and thrombosis as well as improve long-term patency of bypass grafts[18,22]. The data showed that all great saphenous veins isolated by antegrade EVH were adequate grafts for CABG surgery with respect to general macroscopic and randomly selected microscopic evaluations. However, we discarded distal vein graft parts with major injuries resulting from EVH preparation in an inadequately narrow working channel in the early phase of the learning curve. There were no graft injuries during the last 21 EVH procedures. The cardiac surgeon should not accept inadequate bypass graft material, because in these cases, graft occlusion is more frequent and could worsen clinical outcomes[23].

A study by Kodia et al. underlined that currently applied EVH protocols should be advanced in order to improve vein graft quality[5]. Nonetheless, a recent prospective randomized controlled trial clearly depicted that closed tunnel EVH demonstrated gains in quality of life, superior cost-effectiveness, and minor differences in graft integrity compared to open vein harvesting without affecting major adverse cardiac event (MACE) rate after CABG surgery[2]. Another prospective pilot study similarly showed improved post-operative physical recovery, better quality of life, and equal MACE rates after CABG with EVH compared to open vein harvesting[24]. Moreover, the question arises whether or not post-operative outcome after CABG with vein grafts isolated by conventional EVH (in clear majority from the upper leg) may be further enhanced with the described antegrade EVH approach for the lower leg. Follow-up studies are warranted. Experienced surgeons and a structural, protocol-based education of inexperienced colleagues may help maintenance of a higher vein graft quality standard isolated by antegrade EVH and broadening of the technique. Besides isolated and combined CABG scenarios, EVH was also shown to be feasible in elective and high-urgency peripheral bypass grafting scenarios[25,26]. However, caution should be taken. In high-urgency CABG scenarios, a higher level of experience in EVH is required in order to minimize the time exposure and guaranty adequate vein graft quality.

In conclusion, antegrade EVH from the lower leg is a safe method for the isolation of venous graft material for CABG. Macroscopic evaluation and initial histological analyses demonstrated excellent graft quality with preserved endothelial integrity, and this leads to promising clinical results underlining that the method may be a valid alternative to conventional antegrade EVH from the upper leg. Also illustrated is a steep learning curve in experienced cardiac surgeons and low risks for graft-associated complications. The protocol offers a specific institutional step-by-step procedure for antegrade EVH from the lower leg with practical hints, troubleshooting, and possible solutions. Three essential pillars for success were highlighted: (1) specific positioning of the leg, (2) synergizing with bridging vein harvesting technique for minimally invasive access site, and (3) antegrade EVH in a tensionless working channel under continuously optical control of vein graft quality. Therefore, it is proposed that this protocol may help both cardiac and vascular surgeons in developing optimal approaches for high-quality vein graft isolation.

ACKNOWLEDGMENTS:

We thank the entire surgical staff for excellent technical assistance.

DISCLOSURES:

Manuscript publication was funded by Getinge Group (Germany). Alexander Kaminski is a consultant to Getinge Group and receives speaker honoraria from Getinge Group. All authors declare study conduction and entire scientific analyses were executed independently from industrial partners. All authors declare responsibility for the integrity of the work as a whole and have given final approval to the version to be published. All authors declare that there are no conflicts of interest.

REFERENCES

1. Kopjar, T., Dashwood, M. R. Endoscopic Versus "No-Touch" Saphenous Vein Harvesting for Coronary Artery Bypass Grafting: A Trade-Off Between Wound Healing and Graft Patency. *Angiology*. **67** (2), 121-132 (2016).

2. Krishnamoorthy, B. et al. Study Comparing Vein Integrity and Clinical Outcomes in Open Vein Harvesting and 2 Types of Endoscopic Vein Harvesting for Coronary Artery Bypass Grafting: The VICO Randomized Clinical Trial (Vein Integrity and Clinical Outcomes). *Circulation*. **136** (18), 1688-1702 (2017).

3. Andreas, M. et al. Endoscopic vein harvesting is influenced by patient-related risk factors and may be of specific benefit in female patients. *Interactive CardioVascular and Thoracic Surgery*. **17** (4), 603-607 (2013).

4. Deppe, A. C. et al. Endoscopic vein harvesting for coronary artery bypass grafting: a systematic review with meta-analysis of 27,789 patients. *Journal of Surgical Research*. **180** (1), 114-124 (2013).

5. Kodia, K. et al. Graft patency after open versus endoscopic saphenous vein harvest in coronary artery bypass grafting surgery: a systematic review and meta-analysis. *The Annals of Thoracic Surgery*. **7** (5), 586-597 (2018).

6. Krishnamoorthy, B. et al. A randomized study comparing three groups of vein harvesting methods for coronary artery bypass grafting: endoscopic harvest versus standard bridging and open techniques. *Interactive CardioVascular and Thoracic Surgery*. **15** (2), 224-228 (2012).

7. Harskamp, R. E., Lopes, R. D., Baisden, C. E., de Winter, R. J., Alexander, J. H. Saphenous vein graft failure after coronary artery bypass surgery: pathophysiology, management, and future directions. *Annals of Surgery*. **257** (5), 824-833 (2013).

8. Paz, M. A., Lupon, J., Bosch, X., Pomar, J. L., Sanz, G. Predictors of early saphenous vein aortocoronary bypass graft occlusion. The GESIC Study Group. *The Annals of Thoracic Surgery*. **56** (5), 1101-1106 (1993).

9. Lopes, R. D. et al. Project of Ex Vivo Vein Graft Engineering via Transfection IV (PREVENT IV) Investigators. Relationship between vein graft failure and subsequent clinical outcomes after coronary artery bypass surgery. *Circulation*. **125** (6), 749-756 (2012).

10. Goldman, S. et al. Predictors of graft patency 3 years after coronary artery bypass graft surgery. Department of Veterans Affairs Cooperative Study Group No. 297. *Journal of the American College of Cardiology*. **29** (7), 1563-1568 (1997).

11. Sarzaeem, M. R. et al. Scoring system for predicting saphenous vein graft patency in coronary artery bypass grafting. *Texas Heart Institute Journal*. **37** (5), 525-530 (2010).

12. Yerebakan, C., Kaminski, A., Liebold, A., Steinhoff, G. Safety of intramyocardial stem cell therapy for the ischemic myocardium: results of the Rostock trial after 5-year follow-up. *Cell Transplantation*. **16** (9), 935-940 (2007).

13. Stamm, C. et al. Intramyocardial delivery of CD133+ bone marrow cells and coronary artery bypass grafting for chronic ischemic heart disease: safety and efficacy studies. *The Journal of Thoracic and Cardiovascular Surgery*. **133** (3), 717-725 (2007).

14. Kinoshita, T. et al. Bilateral versus single internal thoracic artery grafting in dialysis patients with multivessel disease. *The Heart Surgery Forum*. **13** (5), E280-E286 (2010).

15. Grau, J. B. et al. Propensity matched analysis of bilateral internal mammary artery versus single left internal mammary artery grafting at 17-year follow-up: validation of a contemporary surgical experience. *European Journal of Cardio Thoracic Surgery*. **41** (4), 770-776 (2012).

16. Lytle, B. W. et al. The effect of bilateral internal thoracic artery grafting on survival during 20 postoperative years. *The Annals of Thoracic Surgery*. **78** (6), 2005-2014 (2004).

17. Lytle, B. W. Prolonging patency--choosing coronary bypass grafts. *The New England Journal of Medicine*. **351** (22), 2262-2264 (2004).

18. Kiani, S. et al. Endoscopic venous harvesting by inexperienced operators compromises venous graft remodeling. *The Annals of Thoracic Surgery*. **93** (1), 11-18 (2012).

19. Nezafati, M. H., Nezafati, P., Amoueian, S., Attaranzadeh, A., Rahimi, H. R. Immunohistochemistry comparing endoscopic vein harvesting vs. open vein harvesting on saphenous vein endothelium. *Journal of Cardiothoracic Surgery*. **9**, 101 (2014).

20. García-Altés, A., Peiró, S. A systematic review of cost-effectiveness evidence of endoscopic saphenous vein harvesting: is it efficient? *European Journal Of Vascular And Endovascular Surgery*. **41** (6), 831-836 (2011).

21. Rustenbach, C. J., Wachter, K., Franke, U. F. W., Baumbach, H. Expanding Surgical Opportunities: Endoscopic Harvesting of the Vena Saphena Parva in Supine Position. *The Thoracic and Cardiovascular Surgeon.* **65** (4), 322-324 (2017).

22. Raja, S. G., Sarang, Z. Endoscopic vein harvesting: technique, outcomes, concerns & controversies. *Journal of Thoracic Disease*. **5 Suppl 6**, S630-S637 (2013).

23. Desai, P. et al. Impact of the learning curve for endoscopic vein harvest on conduit quality and early graft patency. *The Annals of Thoracic Surgery.* **91**, 1385-1392 (2011).

24. Luckraz, H., Cartwright, C., Nagarajan, K., Kaur, P., Nevill, A. Major adverse cardiac and cerebrovascular event and patients' quality of life after endoscopic vein harvesting as compared with open vein harvest (MAQEH): a pilot study. *Open Heart*. **5**, e000694 (2018).

25. Khan, S. Z. et al. Endoscopic vein harvest does not negatively affect patency of great saphenous vein lower extremity bypass. *Journal of Vascular Surgery*. **63** (6), 1546-1554 (2016).

26. Santo, V. J. et al. Open versus endoscopic great saphenous vein harvest for lower extremity revascularization of critical leg ischemia*. Journal of Vascular Surgery*. **59** (2), 427-434 (2014).

Figure 1

Click here to access/download;Figure;re-rev Figure 1-1.tif

Figure 2

Figure 3

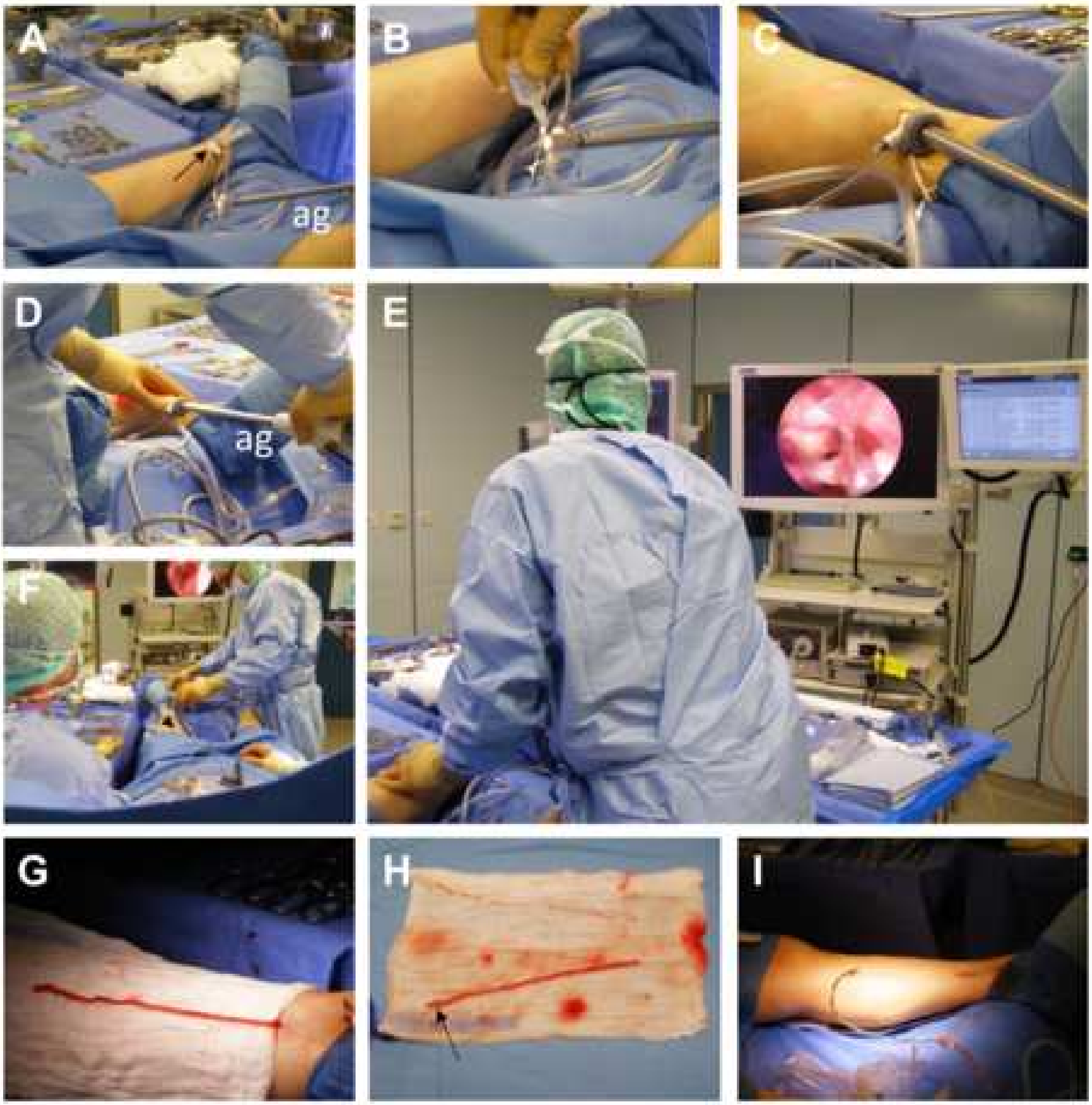
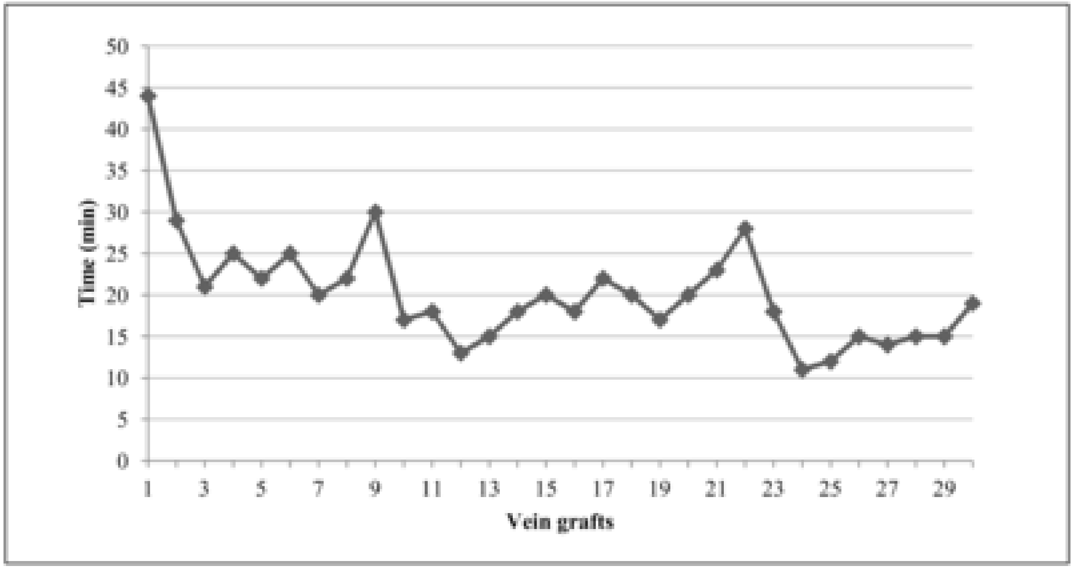
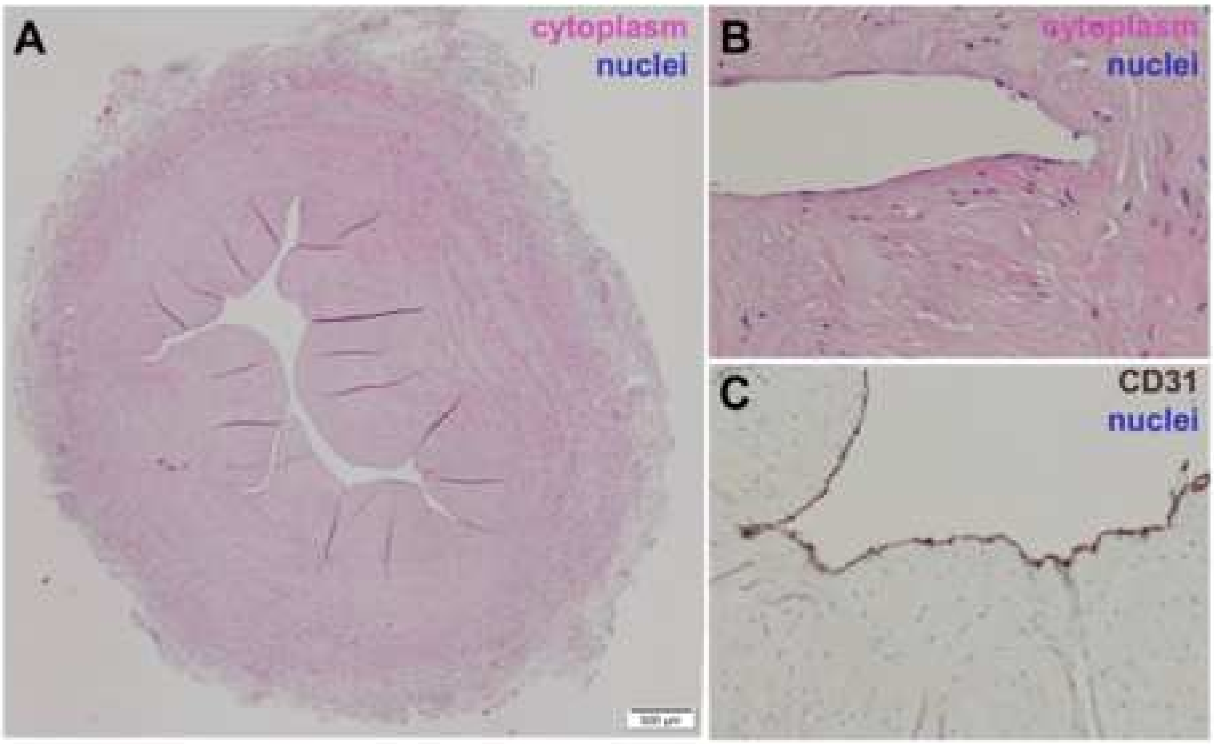


Figure 4



Time (min)
50
45
40
35
30
25
20
15
10
5
0
1
3
5
7
9
11
13
15
17
19
21
23
25
27
29
Vein grafts


A
cytoplasm
nuclei
B
cytoplasm
nuclei
C
CD31
nuclei

Name of Material/Equipment	Company	Catalog Number
disposable scalpel (size 11, Präzisa Plus)	Dahlhausen, Germany	
small curved smooth (anatomical) clamps	B. Braun Aesculap, Germany	
toothed (surgical) forceps	B. Braun Aesculap, Germany	
surgical scissors	B. Braun Aesculap, Germany	
holder for scalpel blade (size 10)	B. Braun Aesculap, Germany	
fine smoth (anatomical) forcep	B. Braun Aesculap, Germany	
sponge-holding clamp	B. Braun Aesculap, Germany	
clipping device	Fumedica, Switzerland	
18 Gauge cannula (Sterican)	B. Braun, Germany	
light handle	Simeon Medical, Germany	
needle holder	B. Braun Aesculap, Germany	
tissue retractor	B. Braun Aesculap, Germany	
Redon needle	B. Braun Aesculap, Germany	
adhesive hook and loop fastener	Mölnlycke, Germany	
extended length endoscope	Karl Storz, Germany	
optical cable	Karl Storz, Germany	
transparent drap camera cover	ECOLAB Healthcare, Germany	
connection cable for electrocauterisation	Maquet, Getinge Group, Germany	
gas insufflation set	Dahlhausen, Germany	
Fred Anti-Fog Solution	Medtronic, USA	
bipolar electrocoagulation device	Maquet, Getinge Group, Germany	
monitor (WideView)	Karl Storz, Germany	
light source (xenon 300)	Karl Storz, Germany	
gas insufflation controller (Endoflator)	Karl Storz, Germany	
half-cylindrical foam roller	Almatros, Gebr. Albrecht KG, Germany	
full-cylindrical foam roller	Almatros, Gebr. Albrecht KG, Germany	
bulldog clamp	B. Braun Aesculap, Germany	
flexible vessel cannula	Medtronic, USA	
vessel loop (Mediloops)	Dispomedica, Germany	
Heparin-Natrium (5000 U) in 200ml saline	B. Braun, Germany	
Langenbeck hooks	B. Braun Aesculap, Germany	

sutures (polygalctin 910, Vicryl 2-0, 4-0; poly ethylene terephthalate, Ethibond 2-0)	Ethicon, Johnson & Johnson, USA
Endoscopic vessel harvesting system, Vasoview Hemopro II	Maquet, Getinge Group, Germany
Octenidindihydrochloride, Octeniderm	Schuelke & Mayr GmbH , Germany

Comments
a
b
c
d
e
f
g
h
i
j
k
l
m
n
o
p
q
r
s
t
u
v
w
x
y
z
aa
ab
ac
ad
ae

af
ag

jove JOURNAL OF VISUALIZED EXPERIMENTS

1 Alewife Center #200
Cambridge, MA 02140
tel. 617.945.9051
www.jove.com

ARTICLE AND VIDEO LICENSE AGREEMENT

Title of Article: Synergizing antegrade endoscopic with bridging vein harvesting advanced great saphenous vein graft quality from the lower leg.

Author(s): Christian Klopsch, Alexander Kaminski, Pascal Dohmen, Friedrich Prall

Item 1 (check one box): The Author elects to have the Materials be made available (as described at http://www.jove.com/author) via: [X] Standard Access [ ] Open Access

Item 2 (check one box):

[X] The Author is NOT a United States government employee.

[ ] The Author is a United States government employee and the Materials were prepared in the course of his or her duties as a United States government employee.

[ ] The Author is a United States government employee but the Materials were NOT prepared in the course of his or her duties as a United States government employee.

ARTICLE AND VIDEO LICENSE AGREEMENT

1. Defined Terms. As used in this Article and Video License Agreement, the following terms shall have the following meanings: "**Agreement**" means this Article and Video License Agreement; "**Article**" means the article specified on the last page of this Agreement, including any associated materials such as texts, figures, tables, artwork, abstracts, or summaries contained therein; "**Author**" means the author who is a signatory to this Agreement; "**Collective Work**" means a work, such as a periodical issue, anthology or encyclopedia, in which the Materials in their entirety in unmodified form, along with a number of other contributions, constituting separate and independent works in themselves, are assembled into a collective whole; "**CRC License**" means the Creative Commons Attribution-Non Commercial-No Derivs 3.0 Unported Agreement, the terms and conditions of which can be found at: http://creativecommons.org/licenses/by-nc-nd/3.0/legalcode; "**Derivative Work**" means a work based upon the Materials or upon the Materials and other pre-existing works, such as a translation, musical arrangement, dramatization, fictionalization, motion picture version, sound recording, art reproduction, abridgment, condensation, or any other form in which the Materials may be recast, transformed, or adapted; "**Institution**" means the institution, listed on the last page of this Agreement, by which the Author was employed at the time of the creation of the Materials; "**JoVE**" means MyJove Corporation, a Massachusetts corporation and the publisher of *The Journal of Visualized Experiments*; "**Materials**" means the Article and / or the Video; "**Parties**" means the Author and JoVE; "**Video**" means any video(s) made by the Author, alone or in conjunction with any other parties, or by JoVE or its affiliates or agents, individually or in collaboration with the Author or any other parties, incorporating all or any portion of the Article, and in which the Author may or may not appear.

2. Background. The Author, who is the author of the Article, in order to ensure the dissemination and protection of the Article, desires to have the JoVE publish the Article and create and transmit videos based on the Article. In furtherance of such goals, the Parties desire to memorialize in this Agreement the respective rights of each Party in and to the Article and the Video.

3. Grant of Rights in Article. In consideration of JoVE agreeing to publish the Article, the Author hereby grants to JoVE, subject to **Sections 4** and **7** below, the exclusive, royalty-free, perpetual (for the full term of copyright in the Article, including any extensions thereto) license (a) to publish, reproduce, distribute, display and store the Article in all forms, formats and media whether now known or hereafter developed (including without limitation in print, digital and electronic form) throughout the world, (b) to translate the Article into other languages, create adaptations, summaries or extracts of the Article or other Derivative Works (including, without limitation, the Video) or Collective Works based on all or any portion of the Article and exercise all of the rights set forth in (a) above in such translations, adaptations, summaries, extracts, Derivative Works or Collective Works and (c) to license others to do any or all of the above. The foregoing rights may be exercised in all media and formats, whether now known or hereafter devised, and include the right to make such modifications as are technically necessary to exercise the rights in other media and formats. If the "Open Access" box has been checked in **Item 1** above, JoVE and the Author hereby grant to the public all such rights in the Article as provided in, but subject to all limitations and requirements set forth in, the CRC License.

4. Retention of Rights in Article. Notwithstanding the exclusive license granted to JoVE in **Section 3** above, the Author shall, with respect to the Article, retain the non-exclusive right to use all or part of the Article for the non-commercial purpose of giving lectures, presentations or teaching classes, and to post a copy of the Article on the Institution's website or the Author's personal website, in each case provided that a link to the Article on the JoVE website is provided and notice of JoVE's copyright in the Article is included. All non-copyright intellectual property rights in and to the Article, such as patent rights, shall remain with the Author.

5. Grant of Rights in Video – Standard Access. This **Section 5** applies if the "Standard Access" box has been checked in **Item 1** above or if no box has been checked in **Item 1** above. In consideration of JoVE agreeing to produce, display or otherwise assist with the Video, the Author hereby acknowledges and agrees that, Subject to **Section 7** below, JoVE is and shall be the sole and exclusive owner of all rights of any nature, including, without limitation, all copyrights, in and to the Video. To the extent that, by law, the Author is deemed, now or at any time in the future, to have any rights of any nature in or to the Video, the Author hereby disclaims all such rights and transfers all such rights to JoVE.

6. Grant of Rights in Video – Open Access. This **Section 6** applies only if the "Open Access" box has been checked in **Item 1** above. In consideration of JoVE agreeing to produce, display or otherwise assist with the Video, the Author hereby grants to JoVE, subject to **Section 7** below, the exclusive, royalty-free, perpetual (for the full term of copyright in the Article, including any extensions thereto) license (a) to publish, reproduce, distribute, display and store the Video in all forms, formats and media whether now known or hereafter developed (including without limitation in print, digital and electronic form) throughout the world, (b) to translate the Video into other languages, create adaptations, summaries or extracts of the Video or other Derivative Works or Collective Works based on all or any portion of the Video and exercise all of the rights set forth in (a) above in such translations, adaptations, summaries, extracts, Derivative Works or Collective Works and (c) to license others to do any or all of the above. The foregoing rights may be exercised in all media and formats, whether now known or hereafter devised, and include the right to make such modifications as are technically necessary to exercise the rights in other media and formats. For any Video to which this Section 6 is applicable, JoVE and the Author hereby grant to the public all such rights in the Video as provided in, but subject to all limitations and requirements set forth in, the CRC License.

7. Government Employees. If the Author is a United States government employee and the Article was prepared in the course of his or her duties as a United States government employee, as indicated in **Item 2** above, and any of the licenses or grants granted by the Author hereunder exceed the scope of the 17 U.S.C. 403, then the rights granted hereunder shall be limited to the maximum rights permitted under such statute. In such case, all provisions contained herein that are not in conflict with such statute shall remain in full force and effect, and all provisions contained herein that do so conflict shall be deemed to be amended so as to provide to JoVE the maximum rights permissible within such statute.

8. Likeness, Privacy, Personality. The Author hereby grants JoVE the right to use the Author's name, voice, likeness, picture, photograph, image, biography and performance in any way, commercial or otherwise, in connection with the Materials and the sale, promotion and distribution thereof. The Author hereby waives any and all rights he or she may have, relating to his or her appearance in the Video or otherwise relating to the Materials, under all applicable privacy, likeness, personality or similar laws.

9. Author Warranties. The Author represents and warrants that the Article is original, that it has not been published, that the copyright interest is owned by the Author (or, if more than one author is listed at the beginning of this Agreement, by such authors collectively) and has not been assigned, licensed, or otherwise transferred to any other party. The Author represents and warrants that the author(s) listed at the top of this Agreement are the only authors of the Materials. If more than one author is listed at the top of this Agreement and if any such author has not entered into a separate Article and Video License Agreement with JoVE relating to the Materials, the Author represents and warrants that the Author has been authorized by each of the other such authors to execute this Agreement on his or her behalf and to bind him or her with respect to the terms of this Agreement as if each of them had been a party hereto as an Author. The Author warrants that the use, reproduction, distribution, public or private performance or display, and/or modification of all or any portion of the Materials does not and will not violate, infringe and/or misappropriate the patent, trademark, intellectual property or other rights of any third party. The Author represents and warrants that it has and will continue to comply with all government, institutional and other regulations, including, without limitation all institutional, laboratory, hospital, ethical, human and animal treatment, privacy, and all other rules, regulations, laws, procedures or guidelines, applicable to the Materials, and that all research involving human and animal subjects has been approved by the Author's relevant institutional review board.

10. JoVE Discretion. If the Author requests the assistance of JoVE in producing the Video in the Author's facility, the Author shall ensure that the presence of JoVE employees, agents or independent contractors is in accordance with the relevant regulations of the Author's institution. If more than one author is listed at the beginning of this Agreement, JoVE may, in its sole discretion, elect not take any action with respect to the Article until such time as it has received complete, executed Article and Video License Agreements from each such author. JoVE reserves the right, in its absolute and sole discretion and without giving any reason therefore, to accept or decline any work submitted to JoVE. JoVE and its employees, agents and independent contractors shall have

full, unfettered access to the facilities of the Author or of the Author's institution as necessary to make the Video, whether actually published or not. JoVE has sole discretion as to the method of making and publishing the Materials, including, without limitation, to all decisions regarding editing, lighting, filming, timing of publication, if any, length, quality, content and the like.

11. Indemnification. The Author agrees to indemnify JoVE and/or its successors and assigns from and against any and all claims, costs, and expenses, including attorney's fees, arising out of any breach of any warranty or other representations contained herein. The Author further agrees to indemnify and hold harmless JoVE from and against any and all claims, costs, and expenses, including attorney's fees, resulting from the breach by the Author of any representation or warranty contained herein or from allegations or instances of violation of intellectual property rights, damage to the Author's or the Author's institution's facilities, fraud, libel, defamation, research, equipment, experiments, property damage, personal injury, violations of institutional, laboratory, hospital, ethical, human and animal treatment, privacy or other rules, regulations, laws, procedures or guidelines, liabilities and other losses or damages related in any way to the submission of work to JoVE, making of videos by JoVE, or publication in *JoVE* or elsewhere by JoVE. The Author shall be responsible for, and shall hold JoVE harmless from, damages caused by lack of sterilization, lack of cleanliness or by contamination due to the making of a video by JoVE its employees, agents or independent contractors. All sterilization, cleanliness or decontamination procedures shall be solely the responsibility of the Author and shall be undertaken at the Author's expense. All indemnifications provided herein shall include JoVE's attorney's fees and costs related to said losses or damages. Such indemnification and holding harmless shall include such losses or damages incurred by, or in connection with, acts or omissions of JoVE, its employees, agents or independent contractors.

12. Fees. To cover the cost incurred for publication, JoVE must receive payment before production and publication the Materials. Payment is due in 21 days of invoice. Should the Materials not be published due to an editorial or production decision, these funds will be returned to the Author. Withdrawal by the Author of any submitted Materials after final peer review approval will result in a US$1,200 fee to cover pre-production expenses incurred by JoVE. If payment is not received by the completion of filming, production and publication of the Materials will be suspended until payment is received.

13. Transfer, Governing Law. This Agreement may be assigned by JoVE and shall inure to the benefits of any of JoVE's successors and assignees. This Agreement shall be governed and construed by the internal laws of the Commonwealth of Massachusetts without giving effect to any conflict of law provision thereunder. This Agreement may be executed in counterparts, each of which shall be deemed an original, but all of which together shall be deemed to me one and the same agreement. A signed copy of this Agreement delivered by facsimile, e-mail or other means of electronic transmission shall be deemed to have the same legal effect as delivery of an original signed copy of this Agreement.

A signed copy of this document must be sent with all new submissions. Only one Agreement required per submission.

CORRESPONDING AUTHOR:

Name:	Christian Klopsch, M.D.
Department:	Department of Cardiac Surgery
Institution:	Rostock University Medical Center
Article Title:	Synergizing antegrade endoscopic with bridging vein harvesting advanced great saphenous vein graft quality from the lower leg.
Signature:	[signature] Date: 28.8.2018
after revision	[signature] 27.11.2018

Please submit a signed and dated copy of this license by one of the following three methods:

1) Upload a scanned copy of the document as a pfd on the JoVE submission site;
2) Fax the document to +1.866.381.2236;
3) Mail the document to JoVE / Attn: JoVE Editorial / 1 Alewife Center #200 / Cambridge, MA 02139

For questions, please email submissions@jove.com or call +1.617.945.9051

Editorial comments:

1. Please take this opportunity to proofread your manuscript.
Thank you for the advice. The manuscript was proofread thoroughly.

2. Please remove 'Figure 1.A-1.D', etc., from the Figures themselves.
Thank you for the advice. We removed the chronological order in the figures.

Reviewer comments

Reviewer #2:

Reviewer #2:
Major Concerns:
1-The title states an improved vein graft quality that is not shown or analyzed in your manuscript. I think you should be more cautious in your statements and revise to "may advance great saphenous vein graft quality".
We appreciate the reviewer´s comments. We revised the title as suggested by the reviewer. We think the reader is now able to understand that the herein described methodology for antegrade EVH definitely improved vein graft quality in our hands compared with our historic experiences and may also advance EVH procedural success in other centers.

2-One of the most important aspect of vein grafts is it's long term patency. A recent study showed that open harvesting has a much better graft patency than the endoscopic harvesting. I think this should be studied in the grafts harvested by your technique. (Kodia K, Patel S, Weber MP, Luc JGY, Choi JH, Maynes EJ, et al. Graft patency after open versus endoscopic saphenous vein harvest in coronary artery bypass grafting surgery: a systematic review and meta-analysis. Annals of Cardiothoracic Surgery 2018).
We thank the reviewer again for this criticism. The reference has been already included in the manuscript during the first revision step (see reference no. 5). Our manuscript focussed on the methodological description of an antegrade EVH approach for the great saphenous vein from the lower leg for CABG. There is obviously a lack of long term patency data, which will be focussed in follow-up studies. However, our initial histological analyses of vein grafts illustrated preserved endothelial and vessel wall integrity after all harvesting techniques in our department: open vein harvesting, bridging vein harvesting and antegrade EVH. These initial data point at a possible non-inferiority of antegrade EVH compared with the other harvesting techniques. The study of Kodia et al. underlined the cause for concern when vein graft integrity and long term patency of bypass grafts were addressed after EVH. However, a recent prospective randomized controlled clinical trial clearly depicted that closed tunnel EVH demonstrated gains in quality of life, superior cost-effectiveness and minor differences in graft integrity compared with open vein harvesting without affecting major adverse cardiac event (MACE) rate after CABG surgery (see reference no. 2). Another prospective pilot study similarly addressed improved postoperative physical recovery, better quality of life and equal MACE rate after CABG with EVH compared with open vein harvesting [Luckraz, H., Cartwright, C., Nagarajan, K., Kaur, P., Nevill, A. Major adverse cardiac and cerebrovascular event and patients' quality of life after endoscopic vein harvesting as compared with open vein harvest (MAQEH): a pilot study. Open Heart. 5, e000694 (2018)]. Moreover, it is questionable if post-operative results from vein grafts isolated by conventional EVH (in clear majority from the upper leg) can be compared with the herein described new antegrade EVH approach for the lower leg. Follow-up studies are warranted. The manuscript was revised.

3-Unclear to me the advantages that this procedure provides. I recognize that you state that the technique prevents blood stasis in the vein and concomitant complications, that conclusion is yet to be shown.

This issue has already been discussed during the first revision step. The retrograde EVH approach for the lower leg demonstrated repetitively severe blood stasis during the EVH procedure leading to concomitant complications. Therefore, it was early cancelled in our department before we started our antegrade EVH study for the lower leg. Applying the antegrade EVH approach we have not seen any issues with blood stasis and concomitant complications. Our study did not directly compare the antegrade and retrograde EVH approaches. Therefore, there is obviously a lack of data supporting the general assumption of superiority of the antegrade approach from the lower leg over the conventional EVH alternatives. However, the collected data set was compared with our former institutional experiences for the antegrade approach from the upper leg and retrograde EVH approach from the lower leg.

4-4/19 (21%) of the vein grafts were damaged by this procedure, not entirely unsubstantial. Did this occur in the beginning of the learning curve?
This issue has already been discussed during the first revision step. We had 4 cases of vein injury in the beginning of the learning curve. We had included 11 additional antegrade EVH procedures in the mean time (in total 30 EVH procedures). Moreover, during the last 21 EVH procedures we did not experience any graft injury. The manuscript was revised underlining the safety of the procedure in the late phase of the learning curve.

Reviewer #3:
Manuscript Summary:
The manuscript describes the method for antegrade harvesting of the saphenous vein in patients undergoing bypass surgery and demonstrates the feasibility of this approach and, moreover, the high quality of the vein grafts after harvesting. This method may be of major importance to thoracic and vascular surgeon that perform by pass surgery

Major Concerns:
well written manuscript, excellent description of the methodology. My only concern relates to the use NaCl+Hep for flushing and storing the grafts after harvesting (point 7.1). Using NaCl + Hep is a very poor perservation fluid, often resulting in endothelial activation, with frequent endothelial dysfunction or late endothelial loss, why don't the authors use at least heperanized blood or any other state-of-the-art preservation fluids for the harvested vein graft?
We thank the reviewer for the evaluations and this highly important concern. There are different opportunities available for graft storage during CABG surgery that definitely can affect graft quality. In our department the veins were transferred into heparinized blood for storage as soon as arterial cannulation for cardiopulmonary bypass was accomplished. The manuscript (especially the protocol) was revised.

Reviewer #4:
Manuscript Summary:
The authors presented their protocol for antegrade endoscopic vein harvesting from the lower leg which can be used in routine coronary artery bypass grafting. 28 patients were enrolled to the study. Endoscopic vein harvesting was combined with bridging vein harvesting as a novel method. Vascular histology was analyzed in a randomized selection of vein samples. Although comparative studies and long-term results are needed, early results from this paper say us this new technique may provide surgeons high-quality vein graft isolation.

Major Concerns:
There is no major concern.

Minor Concerns:
Introduction is enough.
Surgical technique is well-described.
Discussion is satisfactory.

<u>Minor correction for line 395: "...nein graft…"</u>
We thank the reviewer for the evaluations and the minor concern. The manuscript was revised.

We hope that the paper will now find your approval for publication in the Journal of Visualized Experiments.

Yours sincerely,

Dr. Christian Klopsch