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Title: Femoral Vascular Graft Implantation in a Swine Model to Test Small-Diameter Vascular Grafts

Authors and Affiliations:

Georgina Iraola-Picornell^{1,2*}, Esther Jorge^{1,3*}, Albert Teis^{1,4,5}, Oriol Rodríguez-Leor^{1,4,5}, Daina Martínez-Falguera^{1,5}, Maria Kalil^{1,2}, Borja Montejo¹, Antoni Bayes-Genis^{1,2,4,5}, Elisabet Berastegui^{1,4}, Christian Muñoz-Guijosa^{1,4,6}, Carolina Gálvez-Montón^{1,4,5}

Corresponding Authors:

Carolina Gálvez-Montón cgalvez@igtp.cat

Email Addresses for All Authors:

Georgina Iraola-Picornell <u>giraola@igtp.cat</u>
Esther Jorge <u>ejorge@igtp.cat</u>

Albert Teis ateis.germanstrias@gencat.cat
Oriol Rodríguez-Leor oriolrodriguez@gmail.com
Daina Martínez-Falguera mdmartinez@igtp.cat

Maria Kalilmkalil@igtp.catBorja Montejobmontejo@igtp.catAntoni Bayes-Genisabayesgenis@gmail.com

Elisabet Berastegui <u>elisabetberasteguigarcia@gmail.com</u>

Christian Muñoz-Guijosa <u>chrmunozg@gmail.com</u>

Carolina Gálvez-Montón <u>cgalvez@igtp.cat</u>

¹ICREC Research Program, Germans Trias i Pujol Health Research Institute (IGTP)

²Department of Medicine, Autonomous University of Barcelona

³Department of Basic Sciences, Faculty of Health Sciences at Manresa, University of Vic - Central University of Catalonia (UVic-UCC)

⁴Heart Institute (iCOR), Germans Trias i Pujol University Hospital

⁵CIBER Cardiovascular, Instituto de Salud Carlos III

⁶Cardiac Surgery Department, 12 de Octubre University Hospital

^{*}These authors contributed equally



Author Questionnaire

- **1. Microscopy**: Does your protocol require the use of a dissecting or stereomicroscope for performing a complex dissection, microinjection technique, or something similar? **No**
- **2. Software:** Does the part of your protocol being filmed include step-by-step descriptions of software usage? **No**
- 3. Filming location: Will the filming need to take place in multiple locations? No

Current Protocol Length

Number of Steps: 22 Number of Shots: 35



Introduction

Videographer: Obtain headshots for all authors available at the filming location.

- 1.1. <u>Georgina Iraola-Picornell:</u> We present a standardized surgical protocol to evaluate tissue-engineered vascular grafts in a porcine model of an end-to-end femoral artery transplantation, bridging preclinical testing and clinical application, especially for vascular grafts less than 6 mm of diameter [1].
 - **1.1.1.** INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 2.3.1*

What technologies are currently used to advance research in your field?

- 1.2. <u>Christian Muñoz-Guijosa:</u> In our field, several advanced technologies play a key role in driving research forward. Reproducibly implanting small-caliber vascular grafts in the femoral position of pigs requires refined microsurgical techniques, precise perioperative management protocols and also, high-resolution ultrasound and angiography [1].
 - **1.2.1.** INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 3.3.1*

What research gap are you addressing with your protocol?

- 1.3. <u>Georgina Iraola-Picornell:</u> Our protocol aims to standardize large-animal models for testing small-diameter grafts in arterial positions like the femoral artery to evaluate new tissue-engineering vascular grafts [1].
 - **1.3.1.** INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 3.8.1*

What research questions will your laboratory focus on in the future?

- 1.4. <u>Christian Muñoz-Guijosa:</u> We aim to evaluate short-term patency and host response to a bioengineered vascular graft, named VasCraft, in a preclinical model. This graft comprises in a decellularized human saphenous vein and reendothelialized with umbilical cord blood-derived endothelial cells [1].
 - **1.4.1.** INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 4.2.1*

Videographer: Obtain headshots for all authors available at the filming location.



Ethics Title Card

This research has been approved by the Animal Experimentation Unit Ethical Committee at the Germans Trias i Pujol Health Research Institute (IGTP) and Government Authorities of the Generalitat de Catalunya



Protocol

Videographer's NOTE: Some shots without a clapboard were B-roll and are not necessary.

2. Hemodynamic Monitoring and Preparation of the Surgical Area

Demonstrator: Georgina Iraola-Picornell

- 2.1. To begin, place the anesthetized animal in a ventral recumbent position on the operating table [1-TXT]. Using tape or bandages, secure each limb to the table to prevent movement during the procedure [2-TXT].
 - 2.1.1. WIDE: Talent placing the animal on the operating table in a ventral recumbent position. TXT: Anesthesia (Induction): Dexmedetomidine (0.03 mg/kg); midazolam (0.3 mg/kg); Ketamine (3 mg/kg)
 - 2.1.2. Talent taping or bandaging the limbs securely to the table. **TXT: Maintain** anesthesia with sevoflurane (1 3%)
- 2.2. Place surface electrodes on the animal's limbs to monitor electrocardiogram changes and heart rate throughout the surgical procedure [1].
 - 2.2.1. Talent attaching electrodes to each limb of the animal.
- 2.3. Next, position a pulse oximeter on the tongue or the corner of the lip to monitor oxygen saturation continuously [1].
 - 2.3.1. Talent positioning the pulse oximeter sensor gently on the animal's tongue or lip.
- 2.4. Wrap a non-invasive blood pressure cuff around the animal's forelimb [1] and insert a temperature probe into the esophagus to measure body temperature [2].
 - 2.4.1. Talent fitting a blood pressure cuff snugly around the forelimb of the animal.
 - 2.4.2. Talent inserting the probe carefully into the esophagus of the animal.
- 2.5. Now, using surgical clippers, shave both the right and left femoral areas [1]. Wash the shaved areas with surgical soap to remove any debris [2] and disinfect the cleaned areas with three alternating rounds of 0.7 percent iodine and 70 percent ethanol under sterile conditions [3].
 - 2.5.1. Talent shaving the right and left femoral regions using surgical clippers.



- 2.5.2. Talent scrubbing the shaved areas with surgical soap.
- 2.5.3. Talent disinfecting the areas by wiping the area with an iodine-soaked cotton.
- 2.6. Perform a surgical handwash thoroughly [1], then wear a sterile gown and gloves [2].
 - 2.6.1. Talent scrubbing hands and forearms following surgical protocol.
 - 2.6.2. Talent donning a sterile gown and gloves.
- 2.7. Drape the animal with a sterile surgical sheet to maintain aseptic conditions [1] and monitor the depth of anesthesia every 10 minutes throughout the surgery [2].
 - 2.7.1. Talent covering the animal completely with a sterile drape.
 - 2.7.2. Talent performing toe pinch to check anesthesia.

3. Vascular Graft Implantation

Demonstrator: Christian Muñoz-García

- 3.1. For intraoperative analgesia, administer a fentanyl bolus if the animal shows signs of pain such as an increase in heart rate [1]. Author's NOTE: Step removed
 - 3.1.1. Talent injecting fentanyl into the animal's catheter upon observing elevated heart rate.
- 3.2. Use an ultrasound probe to locate the right femoral artery and assess the blood flow [1-TXT].
 - 3.2.1. Shot of the Ultrasound screen showing the right femoral artery and its blood flow. TXT: For intraoperative analgesia, administer a fentanyl bolus NOTE: The deleted information from step 3.1 is placed as a text overlay here
- 3.3. Using an electric scalpel, make a 7 to 10-centimeter vertical incision perpendicular to the linea inguinalis [1].
 - 3.3.1. Talent making a vertical incision with an electric scalpel over the marked surgical site. Videographer's NOTE: 3.3.1, 3.4.1, and 3.4.2 are combined.
- 3.4. Spear the subcutaneous tissues to access the underlying muscle layers [1] and electrocoagulate the small surrounding blood vessels to achieve hemostasis [2].
 - 3.4.1. Talent spreading apart the subcutaneous tissues.



- 3.4.2. Talent applying electrocoagulation tool to small vessels until bleeding stops.
- 3.5. Use two Weitlaner retractors to gently separate the Sartorius, Rectus Femoris, Gracilis, and Pectineus muscles for better exposure to the superficial femoral artery [1].
 - 3.5.1. Talent inserting and adjusting two Weitlaner retractors to separate the Sartorius, Rectus Femoris, Gracilis, and Pectineus muscles.
- 3.6. Administer 300 international units per kilogram of heparin intravenously to prevent intraoperative thrombosis prior to femoral artery clamping [1]. Author's NOTE: Step removed
 - 3.6.1. Talent injecting heparin intravenously using a syringe.
- 3.7. Now, apply vascular clamps proximally and distally on the right superficial femoral artery to achieve complete occlusion [1-TXT], ensuring the distance between clamps is approximately 1 centimeter longer than the segment to be resected [2].
 - 3.7.1. Talent placing vascular clamp above the intended resection site. TXT:

 Administer 300 IU/kg heparin IV to prevent thrombosis before femoral artery clamping NOTE: The deleted information from step 3.6 is placed as a text overlay here
 - Videographer's NOTE: 3.7.1 and 3.7.2 are combined
 - 3.7.2. Shot showing both the clamps in place.
- 3.8. Confirm that the replacement graft is anatomically compatible in both length and diameter with the arterial segment to be resected [1-TXT].
 - 3.8.1. Talent holding the graft alongside the clamped arterial segment. **TXT: Ensure** clamps are correctly positioned to control blood flow near the resection site NOTE: The deleted information from step 3.9 is placed as a text overlay here
- 3.9. Ensure the clamps are properly positioned to control blood flow around the area of resection [1]. Author's NOTE: Step removed
 - 3.9.1. Talent pointing to the clamped area.
- 3.10. Next, excise a 5-centimeter segment of the right superficial femoral artery in a slightly oblique manner to facilitate posterior termino-terminal anastomosis near the clamps [1].
 - 3.10.1. Shot of cutting the artery at an oblique angle using surgical scissors or a scalpel.



- 3.11. Then, cut the proximal and distal ends of the graft to match the resected segment [1].

 Author's NOTE: Step removed
 - 3.11.1. Talent trimming the end of the graft using micro scissors.
- 3.12. Using a continuous 7-0 (7-oh) Prolene suture in a running stitch pattern, suture the proximal and distal anastomoses of the experimental graft to the clamped artery [1] from posterior to anterior and medial to lateral [2].
 - 3.12.1. Talent performing termino-terminal anastomosis with running suture.
 - 3.12.2. Shot of fully stitched segment.
- 3.13. Now, unclamp the proximal and distal clamps to restore vascular flow [1] and verify the absence of blood leakage from the anastomoses [2].
 - 3.13.1. Talent carefully releasing clamps.
 - 3.13.2. Talent pointing to the are showing no blood leakage.
- 3.14. Then, close the muscle and subcutaneous tissue using a resorbable size 1 continuous suture [1], and close the skin with a resorbable size 0 intradermal continuous suture [2].
 - 3.14.1. Talent suturing muscle and subcutaneous tissue using a resorbable size 1 continuous suture.
 - 3.14.2. Talent closing the skin with a resorbable size 0 intradermal continuous suture.
- 3.15. Finally, repeat the same surgical procedure on the left superficial femoral artery using the autograft as a control of the graft technique, if needed, and apply a sterile transparent dressing over the surgical site [1]. NOTE: VO is modified to get covered with the filmed shot 3.15.1.
 - 3.15.1. Talent placing and pressing down a transparent dressing to cover the incision.
 - 3.15.2. Talent positioning tools and graft materials.



Results

4. Results

- 4.1. The successful incorporation of the autograft into the femoral artery and its patency were verified non-invasively using Doppler ultrasound. On day 7, the ultrasound confirmed 100% graft patency with no signs of acute rejection or infection [1].
 - 1.1.1. LAB MEDIA: Figure 2A. *Video editor: Highlight the area labeled as "proximal suture"*.
- 4.2. On day 30, Doppler ultrasound confirmed persistent graft patency and consistent diameter, with no visible defects [1].
 - 4.2.1. LAB MEDIA: Figure 2C.
- 4.3. Angiography on day 30 further validated patency with no structural abnormalities in the arterial graft [1].
 - 4.3.1. LAB MEDIA: Figure 2D.
- 4.4. Histological staining of the extracted graft at day 30 showed intact vascular wall structure, including clearly defined tunica adventitia [1], media [2], and intima layers [3].
 - 4.4.1. LAB MEDIA: Figure 3C. *Video editor: Highlight the part labeled "tunica adventitia"*.
 - 4.4.2. LAB MEDIA: Figure 3C. Video editor: Highlight the part "tunica media".
 - 4.4.3. LAB MEDIA: Figure 3C. Video editor: Highlight the part "tunica intima".
- 4.5. Immunohistofluorescence staining revealed proper localization of endothelial and smooth muscle markers across all vascular layers of the autograft, supporting functional tissue integration [1].
 - 4.5.1. LAB MEDIA: Figure 3D



Pronunciation Guide:

dexmedetomidine

Pronunciation link: https://www.howtopronounce.com/dexmedetomidine YouTubeCollins

<u>Dictionary+10How To Pronounce+10Definitions+10</u>

IPA (AmE): /dεksˌmεdəˈtoʊmɪˌdin/ Phonetic: deks-med-eh-TOH-mi-deen

midazolam

No confirmed link found (not available in Merriam-Webster)

IPA (AmE): /ˌmɪdəˈzoʊləm/ Phonetic: mid-uh-ZOH-luhm

sevoflurane

No confirmed link found IPA (AmE): /ˌsɛvoʊˈflʊərin/ Phonetic: sevo-FLOOR-een

linea inguinalis

linea link: https://www.merriam-webster.com/medical/linea How To PronounceMerriam-

Webster+6Merriam-Webster+6Merriam-Webster+6

IPA: /ˈlɪniə/

Phonetic: LEE-nee-uh

inquinal link: https://www.merriam-webster.com/dictionary/inguinal Merriam-

Webster+3Merriam-Webster+3Definitions+3

IPA: /ˈɪngwənəl/

Phonetic: ING-gwuh-nuhl

anastomosis

Link: https://www.merriam-webster.com/dictionary/anastomosis Merriam-

Webster+13Merriam-Webster+13Oxford English Dictionary+13

IPA: /əˌnæs.təˈmoʊ.sɪs/

Phonetic: uh-NAS-tuh-MOH-sis

esophagus

No confirmed link found (not looked up, but standard)

IPA: /ɪˈsɒf.əgəs/

Phonetic: ih-SOF-uh-guhs

Weitlaner

No confirmed link found (proper noun, surgical retractor)

IPA (estim.): /ˈvaɪtlənər/ Phonetic: VYTL-uh-nuhr



electrocoagulate

No confirmed link found IPA: /ɪˌlɛk.troʊˈkoʊ.əgjʊ.leɪt/

Phonetic: ih-LEK-troh-KOH-uh-gyu-layt

anastomoses (plural)

Same as anastomosis but plural

IPA: /əˌnæs.təˈmoʊˌsiːz/

Phonetic: uh-NAS-tuh-MOH-seez

fentanyl

No confirmed link found

IPA: /ˈfɛntənəl/

Phonetic: FEN-tuh-nul

Sartorius, Gracilis, Pectineus, Rectus Femoris (muscles)

No confirmed links found; classical Latin-derived terms

Sartorius: /sɑrˈtɔːriəs/ → sar-TAWR-ee-us

Gracilis: /ˈgreɪsɪlɪs/ → GRAY-sih-lis
Pectineus: /pɛkˈtɪniəs/ → pek-TIN-ee-us

Rectus: /ˈrɛktəs/ → REK-tus

Femoris: /fɪˈmɔːrɪs/ → fih-MOR-is

adventitia, intima

No confirmed links found

adventitia: /əˌdvɛnˈtɪʃə/ → uh-dven-TISH-uh

intima: /ˈɪntɪmə/ → IN-tih-muh