ICG Lymphography in a 4-week Postmortem Cadaver: Implications for a Supermicrosurgery Training Model

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Summary: Surgical models are invaluable resources for training and for research and innovation. In the field of supermicrosurgery (SM), options for surgical models remain limited and imperfect. We report the use of a fresh, previously frozen 4-week postmortem cadaveric specimen for successful distal to proximal indocyanine green (ICG) lymphography of the upper extremity. Our technique was confirmed with handheld SPY fluorescence imaging, which visualized a clearly defined, linear lymphatic system. By outlining a straightforward, reproducible method of lymphatic mapping in cadaveric specimens, our group aims to expand the frontiers of surgical models for SM. (Plast Reconstr Surg Glob Open 2021;9:e3468; doi: 10.1097/GOX.0000000000003468; Published online 26 March 2021.)

INTRODUCTION

Since the advent of supermicrosurgery (SM), by Professor Koshima in 1997, significant effort has been directed toward developing high-fidelity SM models.1 Supermicrosurgery calls for high-level surgical skill that is often unattainable until fellowship years.2 Well-designed models are opportune for both surgical skill development and innovation.2–4 Unfortunately, options for SM models remain limited.

Pafitanis et al2 assessed 15 SM models, ranging from non-biological to living animal models. Only ex vivo pig hind limb and in vivo rat models demonstrated validity for lymphovenous anastomosis (LVA) training. The in vivo rat model was proposed as the superior LVA training model, given the similarity of rat to human lymphatics and the relative ease in obtaining this animal model.2,5 The in vivo rat model also has significant limitations, which impede comprehensive skill development and translation to intraoperative proficiency.

Suami previously demonstrated the use of indocyanine green (ICG) lymphography in fresh, never frozen cadavers for anatomical studies.5,6,7 However, the feasibility of their approach for SM training remained unclear, given the cadavers were no more than 72 hours postmortem.6 Herein, we describe distal to proximal ICG lymphography of the upper extremity (UE) in a 4-week postmortem fresh human cadaver and discuss implications for SM training.

MATERIALS AND METHODS

Bilateral UE specimens from a fresh 4-week postmortem human cadaver were utilized. After being refrigerated at 40°F (4°C) for 2 weeks, the specimen was frozen and maintained below 20°F (–7°C) then defrosted over 3 days. The skin and subcutaneous tissue was relatively thin without notable scars. ICG (25 mg diluted with 10 mL saline) was injected distally in UEs by depositing approximately 0.3-mL subcutaneous wheals in the second, third, and fourth dorsal web spaces. Immediately following injection, ICG was “milked” proximally with gentle manual pressure and visualized using handheld SPY fluorescence imaging (SPY-PHI, Stryker Corp., Kalamazoo, Mich.). Proximal lymphatics were identified in the forearm as clear, linear structures on SPY fluorescence. (See Video [online], which displays ICG lymphatic mapping.) Lymphatics were visualized throughout the entire UE with continued manual pressure. Injection of methylene blue facilitated dissection of lymphatic vessels. Repeat SPY imaging confirmed identification and ICG mapping of the lymphatic system (Fig. 1).

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DISCUSSION

This report adds to the limited literature supporting the use of ICG lymphography in surgical models and its feasibility in fresh human cadavers. Several prior SM animal models incorporated ICG for vessel identification. Banda et al. recently described lymphography via subdermal injection of ICG in a pig foot. Use of ICG has also been described in a chicken wing SM model. However, ICG lymphography in fresh human cadavers has only been described for anatomic research.

Scaglioni and Suami used ICG lymphography in the lower extremity to more thoroughly analyze the relationship between lymphatic subgroups and node drainage. Suami also described successful anatomic mapping of lymphatics in cadaver torsos using ICG and hydrogen peroxide (3%) or acrylic blue dye for enhanced visualization. Most recently, Shinaoka et al. performed whole-body lymphatic mapping by injecting ICG along the lymphatic watershed lines and at the distal extremities of fresh cadavers no more than 72 hours postmortem. These studies laid the groundwork for application of ICG lymphatic mapping to surgical training and innovation. In particular, the findings of Scaglioni and Suami helped refine surgical techniques such as vascularized lymph node transfer. Clearly, novel, high-fidelity surgical models have profound implications for patients undergoing lymphatic and microsurgical surgery. Better anatomic mapping and surgical techniques could decrease the incidence of costly and devastating surgical sequelae, such as lymphedema.

As previously mentioned, few SM models exist with the in vivo rat model currently purported to be one of the best options. However, it is not comprehensive of all skills needed to perform LVA, as dissection of lymphatic vessels is notably easier in rats due to their thin subcutaneous tissue. Dissection of lymphatic vessels can present as many challenges as performing a submillimeter anastomosis. Thus, mastery of a rat model may not translate to success intraoperatively. Furthermore, rat models provide only a limited number of lymphatic vessels (1–3 per thigh) and are typically smaller than human lymphatics. Living animal models present ethical and financial considerations, which could be circumvented by human cadaveric use.

As with all nonliving models, there remain inherent limitations with human cadaveric models due to the absence of bleeding, thrombosis, and venous pressure. Cost and ease of access are also commonly cited barriers to use of cadavers. We have demonstrated ICG lymphography is successful in a 4-week postmortem cadaver. This significantly decreases the barriers to obtaining suitable cadaveric specimens because previous studies exclusively utilized fresh, never frozen cadavers no more than 72 hours postmortem. Additionally, although it is possible to preserve fresh cadavers with refrigeration alone for up to 4 weeks, previously frozen cadavers are easier to obtain. Fresh cadavers utilized for SM training may then be embalmed for anatomic dissection, which may defray expenses. Future studies are warranted to determine the impact of refrigeration and freezing temperatures, timing of freezing, and overall postmortem interval on suitability of fresh cadavers for lymphography and validate their utility as SM models.

Despite limitations, cadaveric models offer unique benefits. Using human specimens most accurately reflect the dimensions and skillset as experienced intraoperatively. Identification of numerous lymphatic vessels allows for the necessary repetition for consolidation of LVA technique and is the logical next step for LVA SM curriculum previously proposed by Pafitanis et al. The healthy lymphatic vessels as we identified in our cadaver are more fragile than sclerotic, diseased vessels; thus, mastery of this
model will ensure that trainees are well prepared for LVA lymphedema cases. The ability to train on healthy human lymphatic vessels also has important implications for the growing field of prophylactic LVA.

CONCLUSIONS

A fresh human cadaver model is ripe for advanced lymphovenous anastomosis supermicrosurgery training. Along with standardization of technical competencies and dedicated training curricula, use of human cadavers for supermicrosurgery training represents a significant opportunity to help develop the next generation of supermicrosurgeons and increase access to these valuable interventions.

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REFERENCES

1. Masia J, Olivares L, Koshima I, et al. Barcelona consensus on supermicrosurgery. J Reconstr Microsurg. 2014;30:53–58.
2. Pafitanis G, Narushima M, Yamamoto T, et al. Evolution of an evidence-based supermicrosurgery simulation training curriculum: A systematic review. J Plast Reconstr Aesthet Surg. 2018;71:976–988.
3. Scaglioni MF, Suami H. Lymphatic anatomy of the inguinal region in aid of vascularized lymph node flap harvesting. J Plast Reconstr Aesthet Surg. 2015;68:419–427.
4. Suami H. Lymphosome concept: Anatomical study of the lymphatic system. J Surg Oncol. 2017;115:13–17.
5. Yamamoto T, Yamamoto N, Yamashita M, et al. Establishment of supermicrosurgical lymphaticovenular anastomosis model in rat. Microsurgery. 2017;37:57–60.
6. Shinaoka A, Koshimune S, Yamada K, et al. A fresh cadaver study on indocyanine green fluorescence lymphography: A new whole-body imaging technique for investigating the superficial lymphatics. Plast Reconstr Surg. 2018;141:1161–1164.
7. Suami H, Shinaoka A. The methodology of lymphatic anatomy studies in a cadaver model: An overview. Plast Aesthet Res. 2019;6:33.
8. Banda CH, Mitsui K, Ishiura R, et al. A supermicrosurgery pig foot training model for practice of lymphaticovenular anastomosis. Microsurgery. 2020;40:91–92.
9. Ganry L, Fuse Y, Sakai H, et al. Refinement of the chicken wing supermicrosurgical training model: Pre-operative indocyanide green injection highlighting vessels’ visualization under 0.4 mm of diameter. Microsurgery. 2019;39:280–281.
10. Kovacs G, Levitan R, Sandeski R. Clinical cadavers as a simulation resource for procedural learning. AEM Educ Train. 2018;2:239–247.