and envisioning a radical change in the management of traumatic amputations and upper extremity transplantation.

Vascularized tongue extracellular matrix production in a porcine model

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Background: Tongue reconstruction is a true challenge due to its very specific and complex functions, motor and sensory. Despite the most advanced autologous surgical techniques, functional outcomes remain poor. To overcome the current limitations, vascularized composite tissue engineering is thus a promising solution.

Methods: Seven porcine tongues were harvested with their vascular lingual arteries and nervous pedicles. Perfusion decellularization was achieved with a detergent-based protocol. Cell removal was established by conventional stainings and DNA quantification. The extracellular matrix structural preservation was appreciated with Masson's trichrome and laminin stainings, three-dimensional histology, and scanning electron microscopy. Vascular bed preservation and access was assessed by angioscopy and Indian ink gel injection. To study cell compatibility, tongue extracellular matrix biopsies were cultured with C2C12 muscle progenitor cells. In vivo biocompatibility was examined by scaffold implantation in rat quadriceps for 4 weeks.

Results: Complete decellularization of porcine tongues could be obtained within 8 days. Cell clearance was confirmed by absence of nuclei on hemotoxylin and eosin sections, and a significant DNA content reduction. Masson's trichrome staining, three-dimensional histology, and scanning electron microscopy showed a well-preserved microscopic structure. Laminin, an important factor for muscle regeneration, was highly preserved. The vascular bed was patent, for both large and small vessels. Seeded cells were viable and implanted scaffolds demonstrated a satisfying integration in the recipient muscle, at macro- and microscopic levels.

Conclusion: We could produce complete porcine tongue graft extracellular matrix, with a patent and accessible vascular tree, which was cyto- and bio-compatible. These results enable future research in whole-tongue vascularized composite tissue engineering.

Vascularized human finger decellularization: a subunit approach to hand tissue engineering

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Background: In hand surgery, conventional autologous techniques are limited for the reconstruction of lost digits, with a high impact on hand function. Finger subunit allotransplantation has been described, but is restricted by limited applications due to immunosuppressive treatment. From previous report in acellular porcine ear graft matrix production, we applied the perfusion-decellularization technique to human finger subunit grafts obtained from cadaveric source, then extended to whole hands.

Methods: Nine long-digit grafts were procured postmortem, along with their collateral ulnar and radial pedicles, from four fresh human donors at the Department of Anatomy. The specimens were decellularized by sequential arterial perfusions of heparinized saline, 1% sodium dodecyl sulfate, isopropanol, phosphate-buffered saline, and type-I DNAse. Cellular clearance was assessed by DNA quantification and hematoxylin and eosin staining. Extracellular matrix preservation was assessed by Masson's trichrome for soft tissues and bone mineral density for phalangeal bones. Acellular samples from skin, bone, and tendon were cultured with NIH-3 T3 fibroblastic cell line for 4 days, and examined with live/dead staining. Finally, a scaffold was re-anastomosed in a porcine recipient, for a short reperfusion study. Finally, decellularization was applied to three whole human hands, and vascular tree assessed by angio-computed tomographic scan.

Results: Digital grafts were successfully decellularized, with quick epidermolysis, nail loss, and complete bleaching. Cell clearance was demonstrated by absence of nuclei on hematoxylin and eosin sections, and an overall 95.3 % DNA content reduction, compared to native tissues ($p < 0.0001$). Masson's trichrome staining showed microscopic structural preservation. Decellularized phalanx mean bone mineral density was 510 mg hydroxyapatite/cm^3, similar to control. Seeded NIH-3 T3 cells were viable and homogeneously distributed on all scaffolds. In vivo, blood quickly reperfused the whole scaffold; after 3 h, the vascular tree was still patent, as demonstrated by fluoroscopy, fingertip pulse, and oxygen saturation at 93%. The extension to a whole-hand graft decellularization was achieved successfully, with an excellent preservation of the superficial and deep vascular tree.

Conclusion: Following our subunit approach strategy, we could produce finger and hand extracellular matrix scaffolds