

6th International Conference on Nanotechnologies and Biomedical Engineering Proceedings of ICNBME-2023, September 20–23, 2023, Chisinau, Moldova - Volume 1: Nanotechnologies and Nano-biomaterials for Applications in Medicine

## Effectiveness of Tissue Engineering in Obtaining the Extracellular Composite Vascularized Bone Matrix

Alina Stoian, Elena Pavlovschi, Nicolae Caproş, Grigore Verega, Viorel Nacu

## https://doi.org/10.1007/978-3-031-42775-6\_39

## Abstract

Massive bone defects are considered to be one of the basic causes of functional disability. The gold standard, which nowadays is autologous grafting, is a perfect combination of mineralized extracellular matrix, bone marrow, and osteogenic cells. However, the available amount of such biological material is limited and the bone large defects remain a challenge. The lack of oxygen and nutrient transport actually remains the basic technical challenge in tissue engineering that limits the achievement of an effective bone allograft in the treatment of massive bone defects. The purpose of the paper is to present the results collected from the experimental study in obtaining the biocompatible extracellular composite vascularized bone matrix (vECCBM). We present a universal approach to a decellularization protocol based on the consecutive use of an isotonic solution, a chelating agent, anionic and ionic detergent as well as an enzyme solution. The effectiveness of decellularization was tested by histological examination (H&E and DAPI staining) and DNA quantification. The biocompatibility test was performed using the cultivation of the STEM cells from the bone marrow. Results: we were able to obtain a protocol for decellularization of the composite grafts, bone + vessel (soft and hard tissue) with the preservation of the vascular pedicle integrity and its connection with the bone compartment having in this way the possibility of applying anastomoses between the decellularized matrix and host.

Keywords: bone defects, extracellular composite bone matrix, bone allograft, tissue engineering



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## References

1. Pereira, H.F., Cengiz, I.F., Silva, F.S., Reis, R.L., Oliveira, J.M.: Scaffolds and coatings for bone regeneration. J. Mater. Sci. Mater. Med. **31**, 27 (2020)

2. Gerli, M.F.M., Guyette, J.P., Evangelista-Leite, D., Ghoshhajra, B.B., Ott, H.C.: Perfusion decellularization of a human limb: a novel platform for composite tissue engineering and reconstructive surgery. PLoS ONE **13**(1), e0191497 (2018). https://doi.org/10.1371/journal.pone.0191497

3. Atala, A.: Methods and compositions for organ decellularization (2002).

https://patentimages.storage.googleapis.com/0b/ab/49/aea2b448149315/US6376244B1.pdf

4. Peneda Pacheco, D., Suárez Vargas, N., Visentin, S., Petrini, P.: From tissue engineering to engineering tissues: the role and application of in vitro models. Biomater Sci. **9**, 70–83 (2021)

5. Nacu, V., Cos,ciug, S., Cobzac, V., Tîmb´alari, T.: Medicina regenerativ`a în restabilirea țesuturilor scheletice. Arta Medica **63**, 30–33 (2017)

6. Sarvazyan, N.: Tissue Engineering: Principles, Protocols, and Practical Exercises. Springer Nature (2020)

7. Diaz-Siso, J.R., Bueno, E.M., Sisk, G.C., Marty, F.M., Pomahac, B., Tullius, S.G.: Vascularized composite tissue allotransplantation–state of the art. Clin. Transplant. **27**, 330–337(2013)

8. Zor, F.: Facial vascularized composite tissue allotransplantation (2013).

https://doi.org/10.5455/gulhane.39844

9. Messner, F., Guo, Y., Etra, J.W., Brandacher, G.: Emerging technologies in organ preservation, tissue engineering and regenerative medicine: a blessing or curse for transplantation? Transpl. Int. **32**, 673–685 (2019)

10. Arenas-Herrera, J.E., Ko, I.K., Atala, A., Yoo, J.J.: Decellularization for whole organ bioengineering. Biomed. Mater **8**, 014106 (2013)

11. Baino, F.: Scaffolds in Tissue EngineeringMaterials, Technologies and Clinical Applications. BoD – Books on Demand (2017)

12. Banfi, A., Holnthoner, W., Martino, M.M., Ylä-Herttuala, S.: Editorial: vascularization for regenerative medicine. Front. Bioeng. Biotechnol. **6**, 175 (2018)

13. Basu, J., Ludlow, J.W.: Overview of tissue engineering/regenerative medicine (2012). https://doi.org/10.1533/9781908818119.1

14. Song, J.J., Ott, H.C.: Organ engineering based on decellularized matrix scaffolds. Trends Mol. Med. **17**, 424–432 (2011)



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Smith, C.A., Richardson, S.M., Eagle, M.J., Rooney, P., Board, T., Hoyland, J.A.: The use of a novel bone allograft wash process to generate a biocompatible, mechanically stable and osteoinductive biological scaffold for use in bone tissue engineering. J. Tissue Eng. Regen. Med. 9, 595–604 (2015)
Chiara, G., et al.: Nanostructured biomaterials for tissue engineered bone tissue reconstruction. Int. J.

Mol. Sci. 13, 737–757 (2012)

17. Yang, P., et al.: Individual tissue-engineered bone in repairing bone defects: a 10-year followup study. Tissue Eng. Part A **26**, 896–904 (2020)

18. Xing, F., Xiang, Z., Rommens, P.M., Ritz, U.: 3D bioprinting for vascularized tissueengineered bone fabrication (2020). https://doi.org/10.3390/ma13102278

19. Petite, H., et al.: Tissue-engineered bone regeneration. Nat. Biotechnol. 18, 959–963 (2000)

20. Borzunov, D.Y., Kolchin, S.N., Malkova, T.A.: Role of the Ilizarov non-free bone plasty in the management of long bone defects and nonunion: problems solved and unsolved. World J. Orthop. **11**, 304–318 (2020)

21. Han, W., Shen, J., Wu, H., Yu, S., Fu, J., Xie, Z.: Induced membrane technique: advances in the management of bone defects. Int. J. Surg. **42**, 110–116 (2017)

22. Iordăchescu, R., Stoian, A., Gornea, T., Ivanov, V., Verega, G.: Corticoperiosteal-skin flap in the treatment of septic pseudarthrosis of the calf. Clinical case. (2020)

23. Verega, G.: Lambourile insulare ale membrului pelvin, (2008)

24. Houben, R.H., Kotsougiani, D., Friedrich, P.F.: Outcomes of vascularized bone allotransplantation with surgically induced autogenous angiogenesis in a large animal model: bone healing, remodeling, and material. J Reconstr Microsurg. **36**(2), 82–92 (2020)

**25.** PROCESS FOR BONE TISSUE DECELLULARIZATION - European Patent Office - EP3095469 A1. https://dx.doi.org/EP-3095469-A1-20161123