

The traditional top-down approach in tissue engineering fails to develop tissues with the intrinsic micro-architecture as seen *in vivo*. This limitation is addressed via the more recent bottom-up approach; however, it too has issues pertaining to mechanical strength, tissue assembly process and other technical problems associated with the most sophisticated 3D organ printing. The integrating approach wherein, both the top-down and bottom-up approaches are combined together is currently promising. Although there are some preliminary reports; however, the combined strategy is not well explored in the field of bone and vascular tissue engineering. This thesis utilizes the integrating approach to engineer bone and vascular tissues using microtissues, biocomposite matrix, and hydrogel.

Firstly, a biocomposite material (GCnHP) composed of gelatin (G), carboxymethyl chitosan (C), polyvinyl alcohol (P), and nano-hydroxyapatite (nH) was fabricated and characterized physicochemically and biologically. Results showed that the scaffold had a heterogeneous interconnected pore size and substantial Young's modulus and hardness, indicating its use in the construction of cancellous bone or as a filler for non-load bearing bone tissue engineering application. The scaffold also supported the growth of human bone marrow mesenchymal stem cells (hBM-MSCs) and human umbilical vein endothelial cells (HUVEC).

Secondly, hBM-MSC microtissues (MTs) were seeded on the fabricated GCnHP scaffold and its osteogenic potential was evaluated both *in vitro* and *in vivo*. *In vitro* results showed substantial mineralization within 7 days, ALP production and the most prevalent osteogenic gene expression by day 14, indicating differentiation of MSCs to osteoblast-like cells. *In vivo* results revealed the formation of ectopic bone-like tissue in immunocompromised mice within 4 weeks.

Finally, HUVEC MTs were embedded in goat tendon collagen (GTC)-human fibrin (HF) hydrogel and vasculogenesis was extensively studied in terms of MT size, age, media conditions and matrix strength and the most optimized condition was validated *in vivo*. Results revealed that, endothelial media (containing 10 % FBS, 50 ng/mL VEGF and 30 ng/mL bFGF), 24h old HUVEC MTs of 500 cells, seeded at 200 MTs/cm³ in GTC-HF gel of ~100 Pa elastic modulus, resulted in most optimal vasculogenesis with intact lumen that were stable up to a week, without any supporting cells. *In vivo* too, the model was stable for 7 days. In conclusion, it may be said that top-down approach in combination with bottom-up approach may be a versatile tool for the development of more complex functional tissues of clinical relevance.

Keywords: Top-down approach, bottom-up approach, integrating approach, human bone marrow mesenchymal stem cell microtissues, human umbilical vein endothelial cell microtissues, biocomposite material, hydrogel, ectopic bone, goat tendon collagen, vasculogenesis