As the role of cartilage-bone crosstalk becomes more evident in disease, development, and repair, in vitro models of the osteochondral complex are progressively recognized as essential tools to study joint pathologies and possible treatments. However, osteochondral models present the challenge to capture the specific microenvironments of cartilage and bone, including the contribution of other tissue types, as in bone vascularization. To progressively approach the complexity of the signaling network governing the osteochondral complex, we developed for the first time a novel triphasic model of the osteochondral interface to study the cross-talk among chondrocytes, osteoblasts, and endothelial cells. Wet-spun poly(e-caprolactone) (PCL) and PCL/hydroxyapatite (HA) scaffolds in combination with a methacrylated gelatin (gelMA) hydrogel were used as the polymeric backbone of the constructs. The scaffold components were engineered with human bone marrow derived mesenchymal stem cells (hMSCs) and human umbilical veins endothelial cells (HUVECs). We exploited a unique dual chamber bioreactor to create distinct microenvironments for cartilage and bone by the simultaneous, separate flow of different media composition for the compartment-specific differentiation towards a cartilaginous or osseous lineage. Within this engineered Microphysiological Vascularized Osteochondral system (µVOCs), hMSCs showed a good separation of chondrogenic and osteogenic markers in terms of histology and gene expression in the cartilage and bone compartment, respectively. HUVECs formed a stable capillary-like network within the engineered bone construct and supported osteogenesis of hMSCs. Remarkably, the presence of hydroxyapatite hindered the pro-osteogenic action of HUVECs, suggesting caution in the design of hydroxyapatite-based engineered bone implants.

Most notably, HUVECs from the osseous compartment enhanced chondrogenesis in the adjacent cartilage compartment, pointing to yet not fully explored role for endothelial cells during limb development and fracture repair.

**Figure 1.** Representative macroscopic images, histology & immunohistochemistry of the vascularized osteochondral constructs made of gelMA and (a) PCL or (b) PCL/HA. Only cartilaginous compartment showed GAG depositions by Alcian Blue stain, cells have a round-shape morphology (H&E). Only osseous compartment showed calcium deposits by Alizarin Red stain, cells have a fusiform morphology (H&E). CD31 immunostaining showed a network of capillary-like HUVECs in the osseous component, and no CD31 staining in the cartilaginous compartment.