A Vascularized 3D Neuroblastoma Model for In Vitro Drug Screening and Tumor Microenvironment Studies

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In pediatric oncology, neuroblastoma (NB) remains one of the most aggressive and deadly solid tumors, with limited treatment success in high-risk cases[1]. Traditional 2D in vitro models fall short in replicating the in vivo complex cellular and structural interactions, as well as the role of vascularization in tumor progression and therapy response[2]. To address this limitation, we developed a vascularized 3D tumor model replicating key features of the NB tumor microenvironment (TME), with potential applications in drug screening and personalized medicine. The platform consists of a polydimethylsiloxane (PDMS)-based microfluidic device incorporating multiple 3D Bioprinted hydrogel compartments: an endothelialized vascular channel, a stromal region, and a tumor area embedded in gelatin methacrylate (GelMA). A clamp unit ensures hydraulic seal during perfusion. Fluid dynamics simulations using COMSOL Multiphysics® evaluate shear stress distributions and nutrient concentration across the device. Permeability tests highlight the selective diffusion properties of the vascular channel, with reduced values measured in the presence of endothelial cells. Live/Dead assays confirm cell viability in long-term culture. Immunofluorescence staining show physiological cellular structures, with Phalloidin and DAPI confirming cytoskeletal organization and nuclei integrity. CD31 and Vimentin staining indicate the successful formation of an endothelial barrier and stromal cell integration. The tumor component is engineered by generating multicellular tumor spheroids (MCTs) composed of NB (SK-N-AS) and fibroblast (BJ) cells, cultured for seven days in ultra-low attachment (ULA) plates. MCTs are characterized using AnaSP and Cell Trackers to verify cell distribution and then integrated into the central compartment. The integrated system is perfused for 7 days, maintaining high spheroid viability and structural integrity, as confirmed by fluorescence imaging. This study demonstrates the feasibility of combining advanced biomaterials, 3D bioprinting, and microfluidics to create physiologically relevant tumor models. The platform offers a promising tool for preclinical drug testing and contributes to the growing field of tumor-on-chip technologies in chemical and biomedical engineering.

Keywords: 3D bioprinting, multicellular tumor spheroids, vascularization, tumor microenvironment, drug screening

References:

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