A SCALABLE BIOREACTOR FOR THE EXPANSION OF ANCHORAGE-DEPENDENT STEM CELLS

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Currently, there is a lack of suitable bioreactors for the expansion of stem cells and other anchorage-dependent cells. Existing bioreactors are either limited in scalability, have a significantly different culture environment from the traditional 2D culture, or difficult to harvest single-cells suspensions. Cell manufacturing using existing bioreactors is an expensive, labor-intensive cell culture process, and typically requires a high-cost ISO 5 cleanroom environment. Southwest Research Institute® (SwRI®) has developed a novel cell expansion bioreactor to propagate cells using a 3D printed, single-use, scalable device, and a closed-loop system. SwRI's patented bioreactor (Figure 1) features tightly packed interconnected spherical voids providing a large surface-to-volume ratio for cell proliferation under perfusion flow. The concave spherical surfaces reduce the hydrodynamic shear to less than 3×10^{-4} Pa, suitable for shear-sensitive anchorage-dependent cell proliferation [2]. This average shear stress in the bioreactor is much lower than the average shear stress in hollow fiber and microcarrier-based bioreactors [3, 4]. The 3D printed bioreactor is easy to scale up while maintaining the same structure.

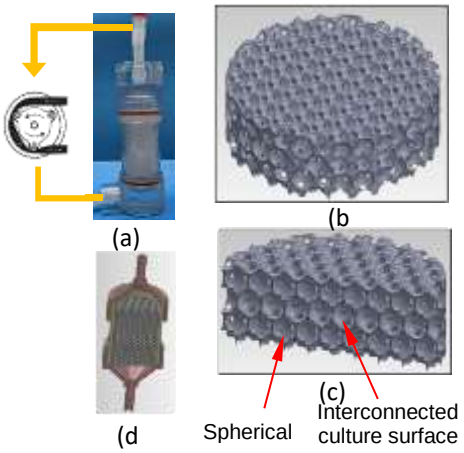

Figure 1 - (a) Pump driven continuous perfusion 3D bioreactor. (b) The 3D bioreactor matrix. (c) Matrix cross-section (d) Assembled bioreactor.

In the expansion of bone marrow-derived mesenchymal stem cells within the bioreactor, the viable cell yields per cm2 were equivalent in comparison to 2D culture (bioreactor = $3.59\text{x}10^4$ ±1.6 vs 2D = $4.36\text{x}10^4$ ±7.6, p=ns). Cell viability was also equivalent between the cells harvested from the bioreactor compared to 2D (bioreactor = 94% ±1.7 vs 2D = 97% ±1.5, p=ns); however, the cell diameter was significantly smaller on the cells harvested from the bioreactor compared to 2D (bioreactor = 17±0.3 vs 2D = 18±0.4, p≤0.05). To indirectly monitor cell confluency from the closed-loop system, metabolites such as lactate and ammonium from a media sample can be measured (Figure 2a); however, to minimize the risk of contamination, a pressure sensor can also be used to indirectly monitor cell confluency during the cell growth phase (Figure 2b). These results suggest that the single-use bioreactor is a convenient tool for expansion of bone marrow mesenchymal stem cells and suitable alternative to 2D cultures.

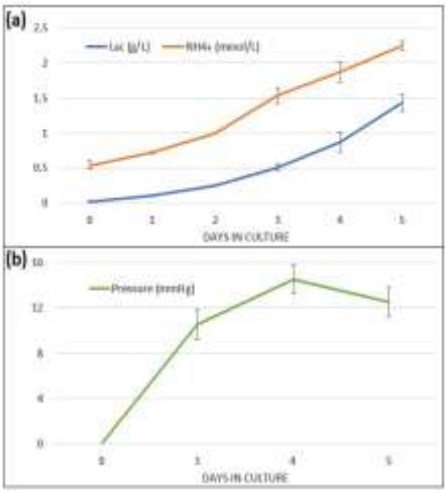

Figure 2 - Pressure, lactate, and ammonium measured based on cell expansion at each day of culture.

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