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LASER PHOTOFABRICATION OF CELL-CONTAINING HYDROGEL CONSTRUCTS

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Two-photon polymerization (2PP) is based on localized cross-linking of photopolymers, induced by two-photon absorption of femtosecond laser pulses [1]. Compared to other additive manufacturing technologies, 2PP offers higher spatial resolution and a possibility to produce 3D structures within the volume of the sample, without the necessity to deposit the material layer-by-layer. In the recent years 2PP attracted much attention as a tool for the fabrication of tissue engineering scaffolds. Incorporating living cells in the fabrication process is advantages with regard to initial cell density and distribution in the scaffold. Light-induced cell encapsulation can be executed at mild pH and temperature conditions. Furthermore it provides advantages of temporal and spatial control of the polymerization process. Cells and tissue are transparent in the near-IR wavelength range commonly used for 2PP, besides the employed laser radiation parameters are known to be harmless to cells. In order to incorporate living cells during 2PP, suitable materials supporting the viability of cells throughout the fabrication process have to be developed.

In this contribution our first results on 2PP fabrication of cell-containing hydrogel constructs are presented. Gelatine-based material formulations with up to 80% cell culture medium have been processed successfully. The cells trapped within the 2PP-produced structures stayed viable and continued to proliferate. The live/dead staining after 3 weeks revealed viable cells occupying most of the space within the 3D hydrogel constructs. The presented results indicate the general practicability of 2PP for 3D processing of cell-containing materials. Potential applications of this highly versatile approach span from precise engineering of 3D tissue models to the fabrication of cellular microarrays.

[1] A. Ovsianikov, V. Mironov, J. Stampf, and R. Liska, Engineering 3D cell-culture matrices: multiphoton processing technologies for biological and tissue engineering applications, *Expert Rev. Med. Devices* 9(6), 613–633 (2012).