

# 3D photofabrication by femtosecond laser pulses and its applications in biomedicine

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Multiphoton absorption of femtosecond laser pulses can be used trigger chemical reactions inducing highly localised modifications within the sample. Nonlinear nature of such light-material interaction allows true 3D processing and realisation of structures with submicrometer spatial resolution. By moving the laser focus in 3D the material is modified along the trace. This way direct laser recording of complex 3D patterns into the volume of photoreactive material is possible (**Figure 1a**).

Two-photon polymerization (2PP) is a method based on localised cross-linking/polymerization of the material induced by femtosecond pulses. Ability of the 2PP to produce true 3D structures with high precision and reproducibility is particularly appealing for the fabrication of tissue engineering scaffolds. The primary function of the scaffold structure is to provide a micro- and nano-structured 3D environment for the cells. In order to facilitate tissue formation the geometry/porosity, chemical composition and surface properties of the scaffold have to resemble natural cell environment. Once the cells of the engineered tissue have built their own connective matrix, the scaffold becomes redundant. Hence, for 2PP fabrication of scaffolds photopolymerizable materials that are biodegradable are required. In addition, femtosecond laser can be used for selective biofunctionalisation of the scaffold by photografting. There are indications that an appropriate combination of biomolecules would ultimately provide control over adherence, migration or even differentiation of cells. Multiphoton grafting allows to tailor local chemical properties of the scaffold surface or a 3D matrix, by space-resolved docking of molecules. **Figure 1b** shows an image of a fluorescent pattern obtained by photografting of aromatic azide compound onto a poly(ethylene glycol) hydrogel matrix using femtosecond laser pulses focused by a conventional 20x microscope objective. Laser scanning microscopy shows that true 3D patterns with a lateral resolution of around 4µm are produced. Most cells are few times larger, therefore by varying the pattern spacing 3D photografting allows to adjust the density of the docked molecules on a biologically relevant scale.

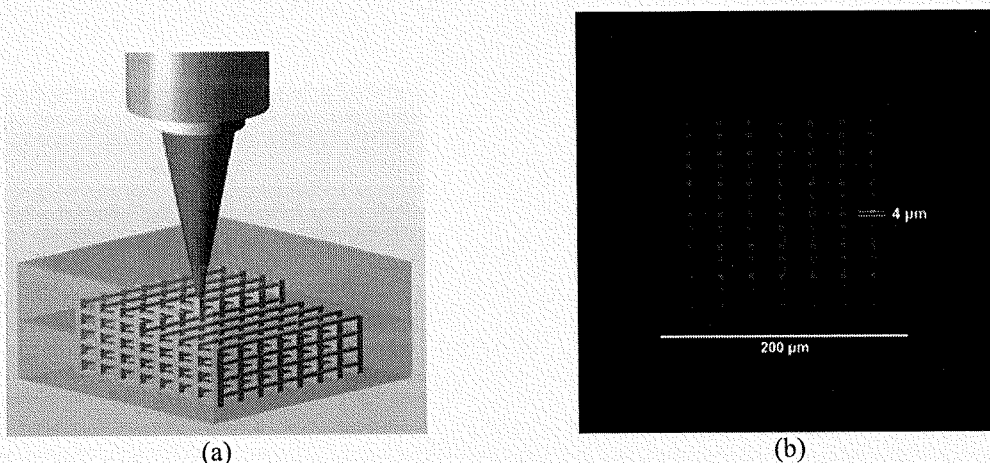


Figure 1 3D photofabrication by femtosecond laser: (a) schematic representation of the process. (b) Laser scanning microscope image of a fluorescent pattern obtained by multiphoton-induced photografting within a hydrogel matrix.

In this contribution, our recent advances on development and microstructuring of novel biophotopolymers and 3D photografting with femtosecond laser are presented. Such important issues as biocompatibility and structurability of applied materials and compounds will be discussed. The presentation is supported by numerous examples.