Slovak Society of Chemical Engineering
Institute of Chemical and Environmental Engineering
Slovak University of Technology in Bratislava

PROCEEDINGS

52nd International Conference of the Slovak Society of Chemical Engineering SSCHE 2026

Hotel SOREA TRIGAN
Štrbské Pleso, Slovakia
May 26 - 29, 2026

Editors: Assoc. prof. Mário Mihaľ

ISBN: 978-80-8208-177-3, EAN: 9788082081773

Published by the Faculty of Chemical and Food Technology, Slovak University of Technology in Bratislava in Slovak Chemistry Library for the Institute of Chemical and Environmental Engineering; Radlinského 9, 812 37 Bratislava, 2026

Zlatník, J., Mamedova, A., Heidenberger, J., Rothbauer, M., Tokárová, V., Řehoř, I.: Hydrogel Microactuators for Laser-Induced Mechanical Stimulation in 3D Cell Culture, Editors: Mihaľ, M., In *52nd International Conference of the Slovak Society of Chemical Engineering SSCHE 2026*, Štrbské Pleso, Slovakia, 2026.

Hydrogel Microactuators for Laser-Induced Mechanical Stimulation in 3D Cell Culture

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Key words: hydrogel microactuators; mechanical stimulation; 3D cell culture; stop-flow lithography; photocrosslinkable hydrogels

Cells in living tissues are continuously exposed to mechanical stimuli that regulate their morphology, migration, and biochemical signaling. However, delivering localized and well-controlled mechanical stimulation to cells embedded in soft three-dimensional (3D) matrices remains challenging for in vitro studies. Most current approaches rely on bulk deformation of hydrogels or macroscopic mechanical loading, limiting spatial precision and control of applied forces. To address these limitations, we developed a soft hydrogel-based microactuator capable of generating controllable mechanical stimulation directly within 3D cell culture environments.

The microactuator is fabricated using stop-flow lithography in microfluidic channels and consists of a composite hydrogel structure. A passive polyethylene glycol diacrylate (PEGDA) backbone is combined with a thermoresponsive poly(N-isopropylacrylamide) (PNIPAM) segment doped with gold nanoparticles. Upon green laser irradiation (532 nm), localized plasmonic heating induces reversible contraction of the active segment, generating deformation of a central hydrogel bridge containing encapsulated cells.

Several photocrosslinkable hydrogels (Dex-HEMA, HAMA, ColMA, and methacrylated human platelet lysate – hPLMA) were evaluated as matrices for cell encapsulation using anterior cruciate ligament fibroblasts. Most hydrogels maintained cell viability above 80 % after seven days of culture. Protein-based hydrogels supported favorable cell morphology, while Dex-HEMA constructs showed depth-dependent viability due to diffusion limitations. Based on biocompatibility and fabrication compatibility, hPLMA was selected as the most suitable hydrogel for integration into the actuator system.

The developed platform enables reversible and tunable mechanical stimulation with forces in the micronewton range and stable cyclic actuation. This system provides a versatile tool for studying mechanobiological responses of cells in soft 3D microenvironments.