3D Bioprinting of Enzyme-Containing Hydrogels

Rakib Sheikh [a,b], Luca Altevogt [a,b], Thomas G. Molley,[d] Joel Young, [a] Kang Liang, [a] Patrick Spicer, [a] Kristopher A. Kilian,[c,d] Peter Wich [a,b,c]*

[a] School of Chemical Engineering, University of New South Wales, Sydney, NSW 2052, Australia

[b] Centre for Advanced Macromolecular Design, University of New South Wales, Sydney, NSW 2052, Australia

[c] Australian Centre for NanoMedicine, University of New South Wales, Sydney, NSW 2052, Australia

[d] School of Materials Science and Engineering, University of New South Wales, Sydney, NSW 2052, Australia

r.sheikh@unsw.edu.au

*p.wich@unsw.edu.au

Hydrogels have become a go-to material in tissue engineering and drug delivery, owing to their biocompatibility and ability to replicate the natural extracellular matrix. However, their functionality is often limited to the passive delivery of proteins, drugs, or cells. The use of hydrogels in tissue engineering has been revolutionized by the introduction of 3D printing technology. This technology allows the precise and accurate deposition of hydrogel layers, resulting in the creation of complex 3D structures that closely resemble native tissues. However, the potential of hydrogel-based 3D printing in tissue engineering can be limited without additional functionalities. In this context, enzymes present a unique opportunity to introduce active functionalities to hydrogel bioinks. An enzyme bioink can catalyze chemical reactions and scavenge reactive oxygen species, which can improve tissue regeneration outcomes.

Our study has addressed the challenge of enzyme delivery in hydrogel bioinks by developing a gelatin-based bioink containing redox enzymes. The enzymes are covalently attached to the hydrogel matrix, preventing enzyme leaching and ensuring its presence for an extended period. The enzyme's catalytic activity is not compromised by the attachment process, and our biocompatibility studies have shown that the enzyme hydrogels do not induce any cytotoxic effects on cells. Furthermore, these 3D-printed enzyme hydrogels have demonstrated beneficial activity *in vitro*, making them a promising candidate for wound dressing or wound patch applications. The use of hydrogels in tissue engineering, coupled with the active functionalities provided by enzymes, has the potential to significantly enhance tissue regeneration outcomes and lead to the development of novel therapies for a range of medical conditions.