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Formulation and characterisation of enzyme-based biomaterials for µFluidic experiments

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PURPOSE OF THE ABSTRACT

The fundamental principle of biological compartmentalisation of cellular life provides the basis for space-time resolved reaction processes. Based on this, intensive work is currently done on the use of interconnected, continuously flowing reaction chambers in order to improve the reaction control and efficiency of chemical syntheses, especially with integrated biocatalysts (so called flow biocatalysis). To this end we have developed formulations of enzyme-based biomaterials based on a genetically encoded coupling system.[1] The resulting enzyme fusions are being used as modular building blocks for the assembly of catalytically active materials, which can be formulated as self-assembling all-enzyme hydrogels (AEHs) or thin films, and characterized in terms of their immobilization and biocatalytic activity in miniaturized flow reactors.

The concept of self-assembling AEHs is compatible with a wide range of enzymes.[2-7] For example, our initial work established a material made of the two homotetrameric enzymes, a (R)-selective alcohol dehydrogenase (ADH) and the cofactor regenerating glucose 1-dehydrogenase (GDH), which are genetically fused with either the SpyCatcher (SC) or the SpyTag (ST).[1] This site-specific mediated enzyme conjugation allows the formulation of a very stable and highly active biomaterial which consists almost exclusively of enzymes, therefore making optimal use of the reaction space compared to other carrier-based immobilization techniques. The novel material was initially characterised via dynamic light scattering (DLS) and scanning electron microscopy (SEM) in terms of their physicochemical behaviour. Moreover, the gels showed excellent stereoselectivity, stable conversion rates, high space-time yields (STY) and cofactor retention for more than seven days in continuous flow experiments. Therefore, AEH is a highly promising material whose high application potential for sustainable and environmentally friendly industrial biocatalysis will be further enhanced by innovative formulations.

FIGURES

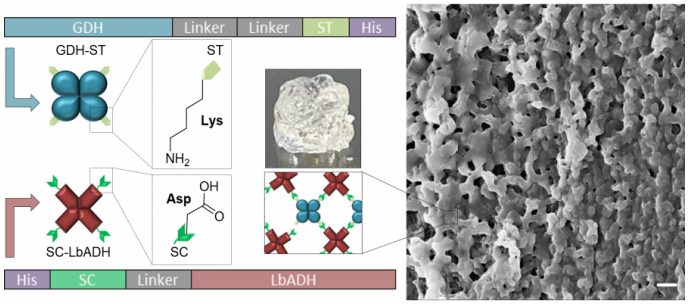

FIGURE 1

Design, formation and morphological characterization schematic illustration of the enzymatic builing blocks, that can self-assemble to a hydrogel upon mixing via SC/ST-mediated conjugation and representative SEM image.

FIGURE 2

KEYWORDS

hydrogels | immobilization | flowbiocatalysis | microreactors

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