

Peptide-Inspired Modular Self-Assembly of Carbonic Anhydrase on Cellulose Interfaces for Direct Air Capture of CO₂

Haneef Ur Rehman and Jenny Molloy

¹ Biomanufacturing group, International Center for Genetic Engineering and Biotechnology (ICGEB), Trieste, Italy

The continuous rise in atmospheric CO₂ levels is a key contributor to global climate change, driving the need for advanced functional materials capable of efficient carbon capture. Peptide self-assembly principles offer a framework for constructing hierarchical, functional biomaterials through modular, non-covalent organization. Therefore, we worked on a hybrid self-assembling biocatalytic system for direct air capture (DAC) of CO₂ using engineered enzyme–cellulose interfaces. Carbonic anhydrase (CA), a highly efficient zinc metalloenzyme, catalyzes rapid CO₂ hydration, however, its practical deployment depends on stable immobilization within structured and recyclable material platforms. To achieve this, we designed a recombinant fusion protein composed of the cellulose-binding module CBM3a (from *Clostridium thermocellum* CipA), green fluorescent protein (GFP) as a structural tracer and thermostable carbonic anhydrase from *Sulfurihydrogenibium azorense* (SazCA). This modular architecture enables spontaneous organization on cellulose substrates, mimicking peptide-driven supramolecular assembly through affinity-guided molecular recognition.

The fusion protein was expressed recombinantly and confirmed by SDS-PAGE (~75 kDa). Upon incubation with cellulose cheesecloth, the construct self-assembled into a stable functional layer via CBM-mediated binding. GFP fluorescence confirmed uniform surface distribution and successful nanoscale organization of the fusion proteins on the cellulose matrix.

The immobilized enzyme retained high catalytic efficiency, maintaining >80% activity after 10 days, while free enzyme lost over 50% activity. Reusability tests showed ~50% residual activity after 10 catalytic cycles, demonstrating strong operational stability.

This study highlighted how peptide-inspired modular assembly concepts can be extended to enzyme–biomaterial engineering, enabling the formation of self-organized, functional cellulose-based catalytic interfaces for sustainable CO₂ capture applications.