



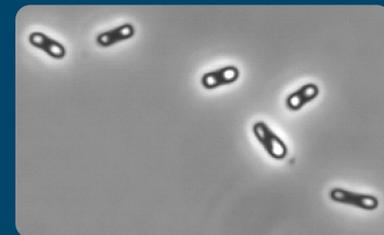
Accelerated production and screening of catalytically active inclusion body libraries via automated workflows

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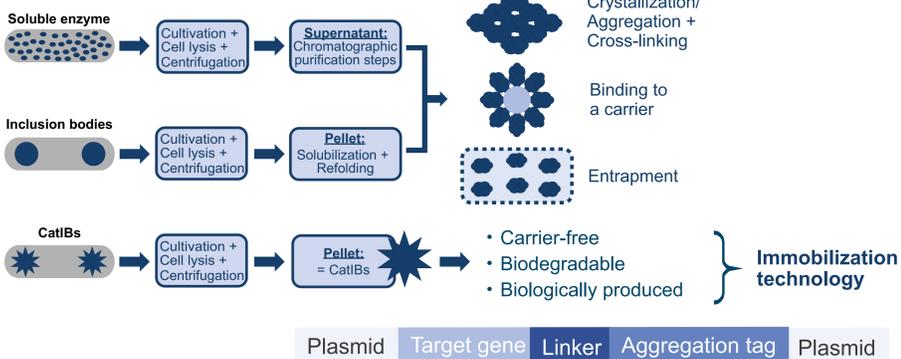
- The generation of successful CatIB variants is time-consuming due to repetitive manual steps
→ Only a few CatIB variants are currently published
- No a priori prediction is possible for successful CatIB formation
→ Automation enables testing of many variants to find the best combination for immobilization
- Aim: Development of automated workflows for high-throughput CatIB library construction, CatIB screening, purification and characterization



CatIBs (Catalytically active inclusion bodies)

= High stability, low cost production, fast & simple purification

Immobilization:



Jäger et al. (2020); Lamm (2019)

Automated microscopy

Cultivation conditions:

- FlowerPlate®
- 3 h 37 °C
- 69 h 15 °C
- M9 autoinduction medium

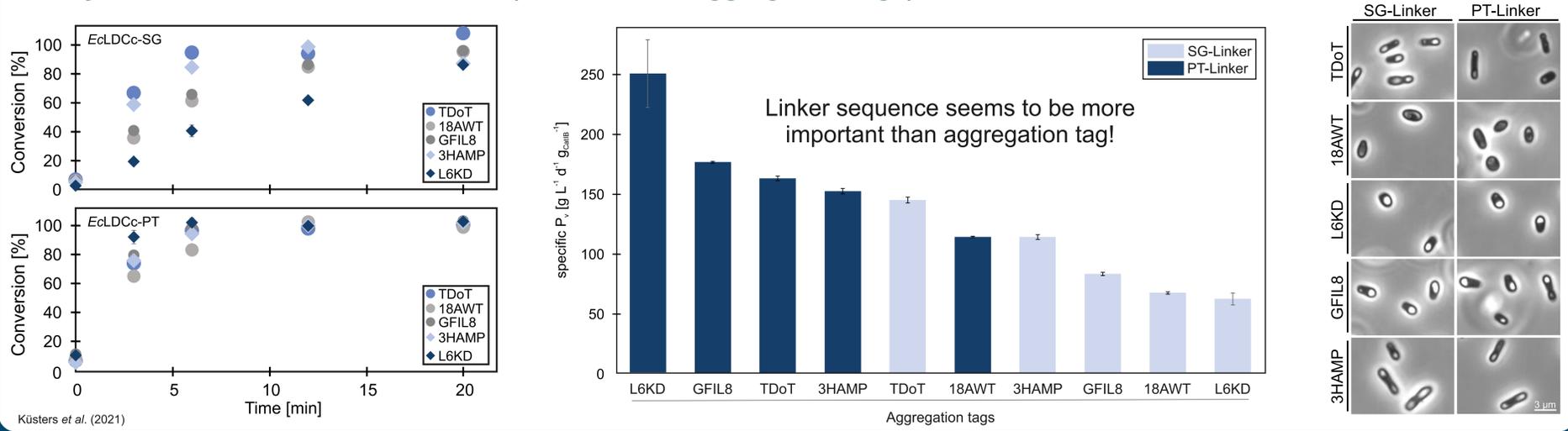
Lysine Decarboxylase (EclDCC):

L-lysine → Decarboxylation by EclDCC → 1,5-diaminopentane (Cadaverine)

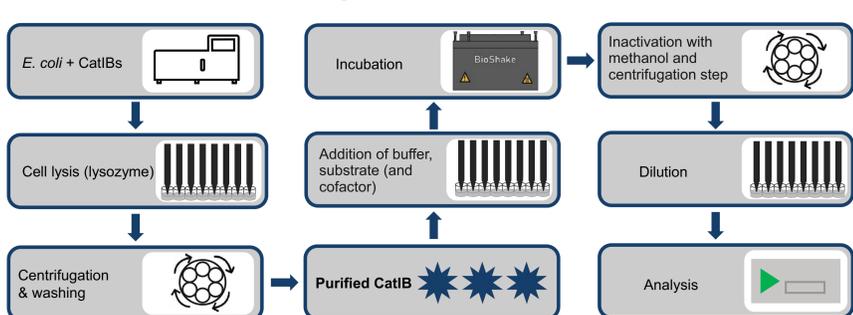
precursor for bio-based polyamides

Visible EclDCC-CatIBs were formed after 31.5 h

Analysis of 10 EclDCC CatIB variants (2 Linkers + 5 Aggregation tags)



Automated CatIB analysis workflow



48 strains: Manual work = 100 h vs. Automated work = 7.5 h

- Golden Gate cloning for high-throughput CatIB library generation
 - Microscopic analysis for phenotyping of inclusion body producing strains
 - CatIB purification and characterization for testing the activity of the generated inclusion bodies
- In combination with automation: Fast technology to implement a set of CatIB variants for diverse target proteins!

This project was funded by the European Regional Development Fund (ERDF)



Member of the Helmholtz Association