



Transformation of the Gram-positive platform organism *Corynebacterium glutamicum* in a fully automated laboratory environment

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Introduction

Background:

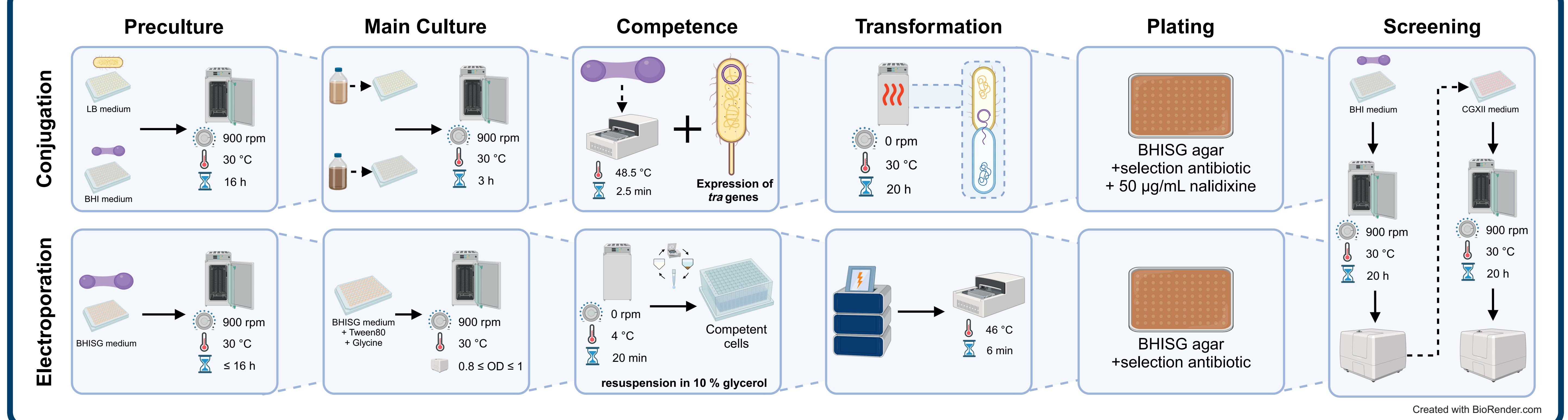
- Rising demand for novel producer strains to satisfy bioeconomic needs
 - *Corynebacterium glutamicum*: a platform organism with diverse application opportunities
- Insufficient genetic engineering capacities available to construct optimized strains for a broad range of products

Approach:

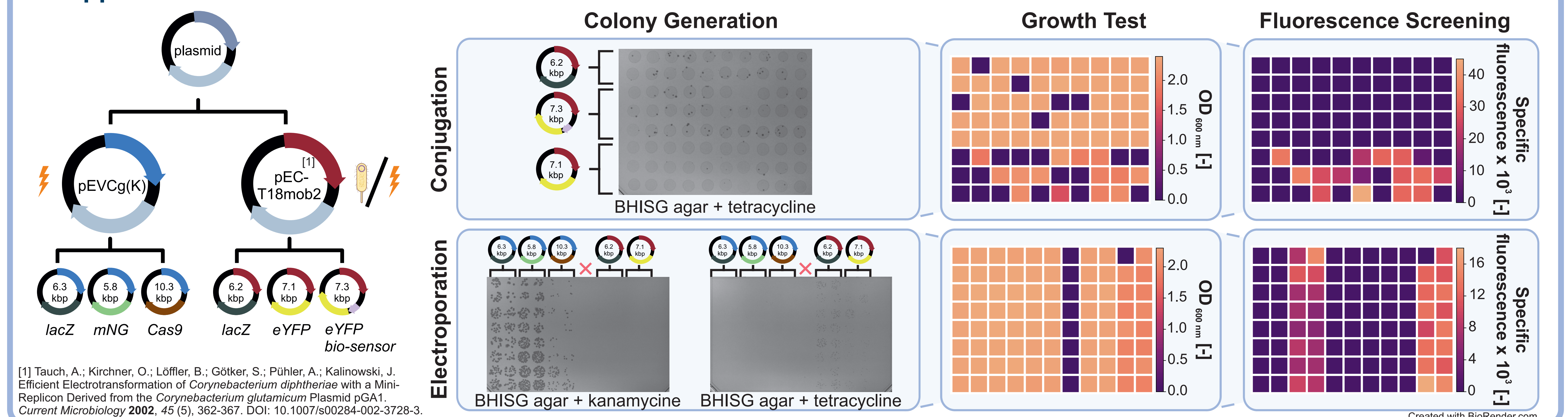
- Fully automated transformation of *C. glutamicum* in 96-well format
 - Conjugation and electroporation
- Transformation of plasmids into the final recipient directly after assembly
 - Assembly purification to enable μ L scale electroporation



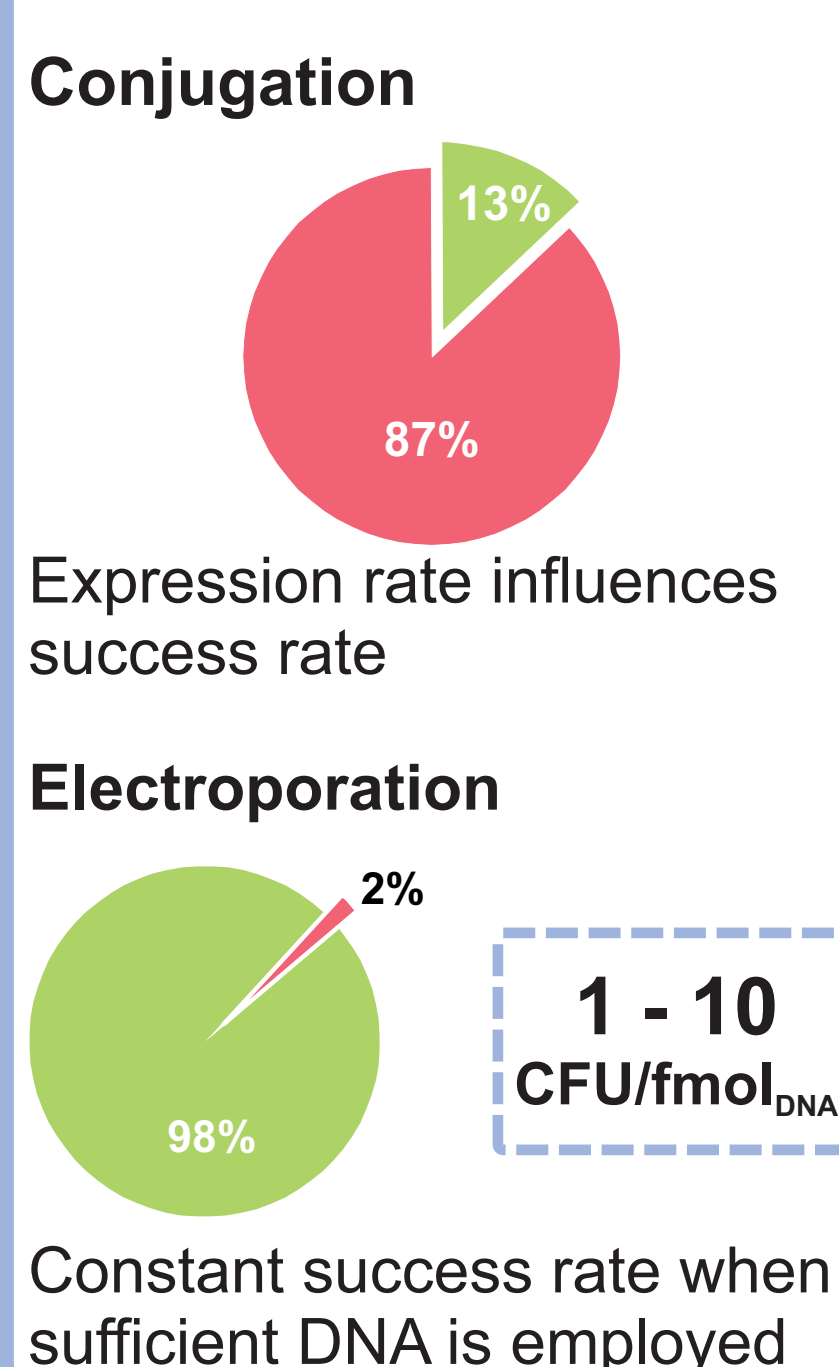
Automated Transformation Workflows



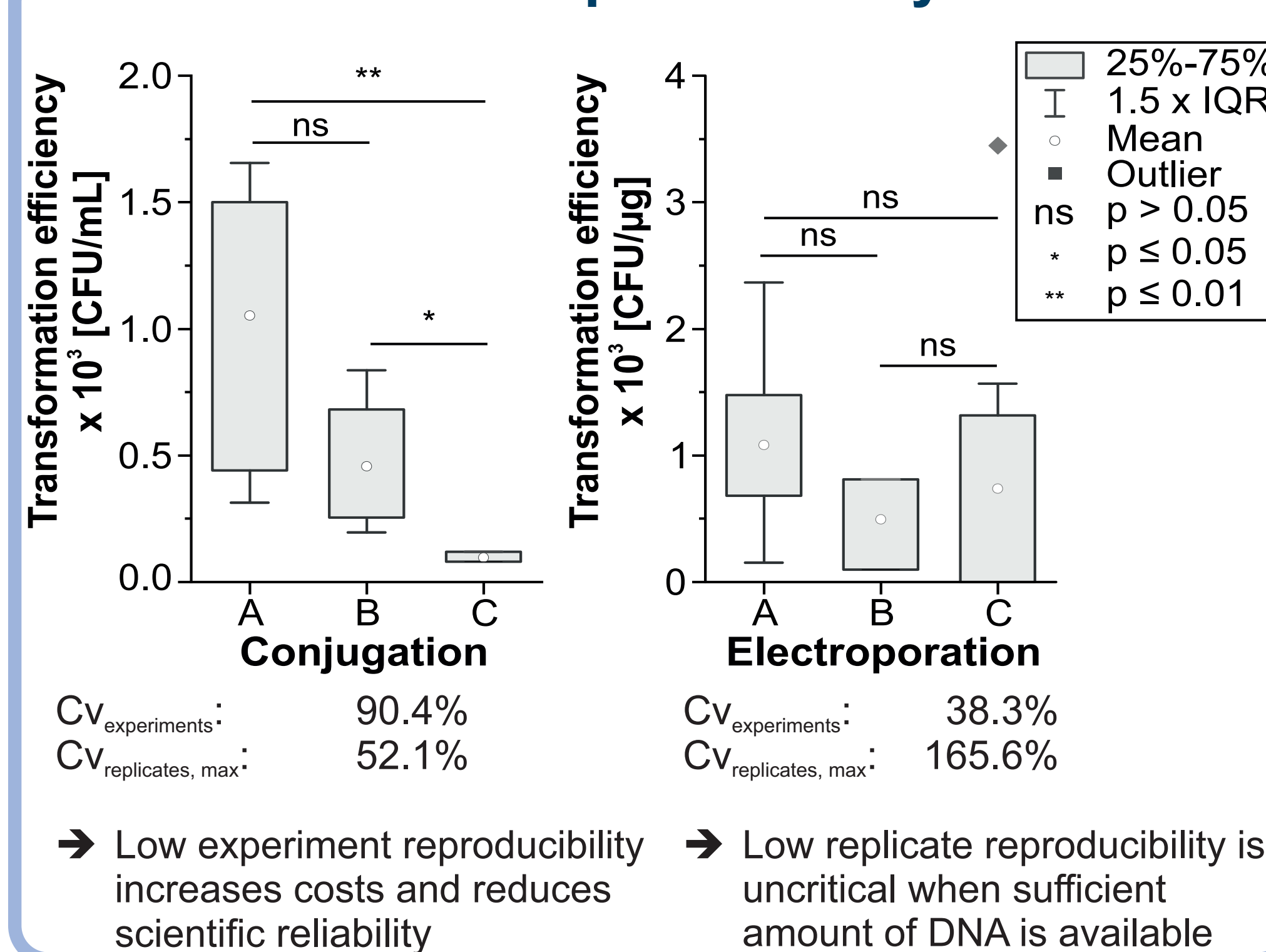
Application of Automated Transformation



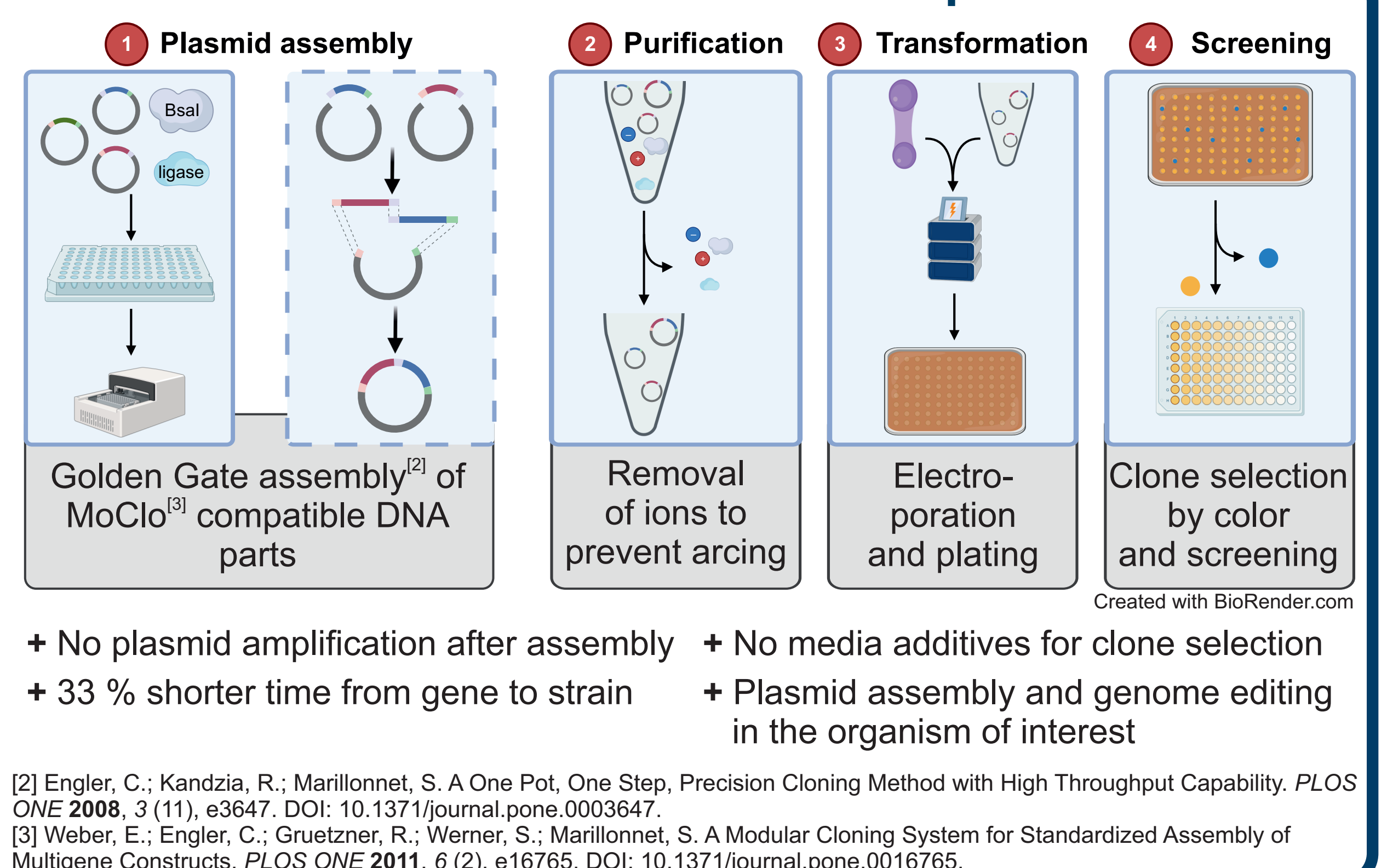
Transformation success



Transformation reproducibility



Direct transformation of assembled plasmids



Conclusion and Outlook

- **Conjugation:** limited plasmids and unreliable results
- **Electroporation:** enables robust automated transformations
- Complex rational strain design will become viable and integratable into automated DBTL cycles

- **Purification of plasmid assembly mixtures streamlines automated molecular biology workflows for Gram-positive bacteria**
- Pitfalls accompanying DNA manipulation with intermediate hosts can be circumvented

