



Oct 14, 2019

E. coli transformation

[iGEM Dusseldorf¹](#)¹Heinrich-Heine Universität Düsseldorf

2

Works for me

[dx.doi.org/10.17504/protocols.io.76jhrcn](https://doi.org/10.17504/protocols.io.76jhrcn)iGEM Dusseldorf 

ABSTRACT

- Take tubes with 50 µL of TOP10F' competent cells from -80°C freezer and place on ice.
- Add 5 µL of each ligation reaction directly to competent cells and mix by tapping gently. **Do not mix cells by pipetting up and down.** Store any remaining ligation at -20°C.
- Incubate cells on ice for 30 min. [while waiting, turn on 42°C water bath]
- Incubate for exactly 30 sec in 42°C water bath. Do not mix or shake.
- Remove vials and place quickly on ice. Store for 2 min on ice.
- Add 500 µL of LB medium (pH. 7.5) to each tube.
- Incubate tubes (taped horizontally to platform) for 60 min at 37°C and 225 rpm.
- Plate 550 µL (variable) of each transformation on LB + AB plates.
- Incubate overnight at 37°C, in the dark.



This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited