



Plasmid transduction using competent cell V.1

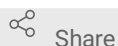
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Version 1 ▼


Oct 06, 2022

1 Works for me



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ABSTRACT

Plasmid can be transduced into bacteria at competent state using heat shock. This protocol helps transduce plasmid into competent cells.

DOI

dx.doi.org/10.17504/protocols.io.j8nlkw4e5l5r/v1

PROTOCOL CITATION

An.Huang 2022. Plasmid transduction using competent cell. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.j8nlkw4e5l5r/v1>



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CREATED

Oct 06, 2022

LAST MODIFIED

Oct 06, 2022










PROTOCOL INTEGER ID

70897

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- 1 Take competent cell out from -80°C fridge and thaw on ice.
- 2 When the cells are completely thawed, pipette  **2 µL** plasmid DNA solution into  **100 µL**^{30m} competent cell.
Put the cell in ice for  **00:30:00**
- 3 Conduct heat shock on the competent cell by placing the cell in  **42 °C** water bath for ^{3m 30s}
 **00:01:30** .
Put the cells back into ice for  **00:02:00**
- 4 Add  **900 µL** LB broth medium into competent cell mixture. Shake at  **180 rpm, 37°C**^{45m} for
 **00:45:00**

5 Centrifuge at  **6000 rpm, Room temperature, 00:05:00** .


5m

Centrifuge radius = 6 cm.

6 Discard  **900 µL** supernatant and resuspend the pellet in the rest  **100 µL** supernatant.

7 Spread the cells onto LB agar plates.

LB agar plates may contain antibiotics, which is determined by the transduced plasmid.

8 Place the plate with lid on upside for  **01:00:00** .

1h

9 Invert the plate and culture at  **37 °C** in a biomedical incubator overnight.

If the bacteria turn out to be too concentrated, dilute the cell before spreading on the plate next time.