

**21/06/2016 13:17**

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1h RT ligation, let's see the results, if negative, second batch of overnight 15 C ligation

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is being prepared

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(Control and sample are probably mixed up during gel purification)

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- 1** Aliquot of 100 uL of DH5α Ca<sup>2+</sup> competent cells was mixed with 20 uL of ligation 210616 and incubated on ice for 30 min.

1) 250\_gp

2) 500\_gp

3) 250\_pt

4) 500\_pt

0) dh2o

make pre-warmed 37  
C medium

- 2** Uptake of DNA was induced by heat shock **1 min at 42 C**  
maybe put some water for better heat transfer

- 3** After the heat shock the cells were diluted in 500 uL of prewarmed LB medium (37C) and incubated for 1 h by shaking at 250 rpm

Make new plates

- 4** 400 and 200 uL aliquotes of cell suspensions were plated onto LB agar with Amp 100

:1a, 1b, 2a, 2b, 3a, 3b, 4a, 4b, 0a, 0b

- 5** Plates were incubated overnight at 37 C