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## Transform pCas9

## Qiaowa Gong<sup>1</sup>

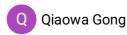
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1 Works for me



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## **ABSTRACT**

this protocol is aimed to provide instructions of transforming pCas9, which is a key step of knock-out gene by Crispr/Cas9

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PROTOCOL CITATION

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**KEYWORDS** 

transformation, Crispr/Cas9, pCas9



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MATERIALS TEXT

competent MG1655, pCas9 plasmids, LB agar mediums (contain chloramphenicol), LB broth

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- 1 Mix **□50 μL** of competent MG1655 with **□10 μL** of pCas9
- 2 Keep the reaction mix on ice for © 00:30:00 minutes

30m

Then, transfer the mix to the water bath set at 42 °C for © 00:00:45

45s

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- 4 Next, keep the mix on the ice for **© 00:02:00**
- 5 Add  $\blacksquare$ 940  $\mu$ L LB broth and incubate the cells at § 37 °C with vigorous shaking (200 rpm) for  $\bigcirc$  01:00:00

- 6 Plate the cells on LB-agar plates containing chloramphenicol
- 7 Incubate at § 37 °C © Overnight