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Miniprep Protocol (QIAGEN)

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1 Resuspend pelleted bacterial cells in 250 µl Buffer P1 and transfer to a microcentrifuge tube.

2 Add 250 µl Buffer P2 and mix thoroughly by inverting the tube 4–6 times.

3 Mix gently by inverting the tube. Do not vortex, as this will result in shearing of genomic DNA.

4 Add 350 µl Buffer N3 and mix immediately and thoroughly by inverting the tube 4–6 times.

5 Centrifuge for 10 min at 13,000 rpm (~17,900 x g) in a table-top microcentrifuge. A compact white pellet will form.
17900 x g

6 Apply 800 µl of the supernatant from step 4 to the QIAprep 2.0 spin column by pipetting.

7 Centrifuge for 30–60 s. Discard the flow-through.
00:01:00

8 Wash the QIAprep 2.0 spin column by adding 0.5 ml Buffer PB and centrifuging for 30–60 s. Discard the flow-through.
00:01:00

9 Wash QIAprep 2.0 spin column by adding 0.75 ml Buffer PE and centrifuging for 30–60 s.
00:01:00

10 Discard the flow-through, and centrifuge at full speed for an additional 1 min to remove residual wash buffer.
00:01:00

11 Place the QIAprep 2.0 column in a clean 1.5 ml microcentrifuge tube. To elute DNA, add 50 µl Buffer EB.