VERSION 2

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Electrotransformation of Clostridium species V.2

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ABSTRACT

A brief protocol for electrotransformation of Clostridium species

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Protocol status: In development

We are still developing and optimizing this protocol

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22162

- 1 Inoculate 10 μ L BHI-supplemented broth with 100 μ L stock culture overnight.
- 2 Use the overnight culture to inoculate 100 mL BHI-supplemented broth to a starting density of OD 0.02.
- 3 Harvest early-exponential phase culture (OD 0.2 to 0.25) by centrifugation by 12,000g x 00:15:00 at 25 °C .
- 4 Wash once in 10 mL SML electroporation buffer .
- 5 Resuspend in 10 mL SMP electroporation buffer .
- 6 Take 400 μ L of aliquots and and mix with 500 ng of DNA, transfer to prechilled cuvettes with 0.2cm gap.
- 7 Incubate on ice for 00:10:00 .
- 8 Electroporate at the following parameters: 25 μ F, resistance 200 Ω , voltage 1.8kV.

9 Immediately transfer the cells into 10 mL of BHI broth and incubate for 03:00:00 .

10 Plate cells in dilutions on solid selective and non-selective BHI agar.