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Agro Transformation for *Mimulus in Planta* Transformation [↗](#)

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Working

[dx.doi.org/10.17504/protocols.io.3pagmie](https://doi.org/10.17504/protocols.io.3pagmie)

Mimulus



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ABSTRACT

This protocol is part of a [collection for *Mimulus in planta* transformation](#).

EXTERNAL LINK

http://mimubase.org/FTP/Protocols/Stable_Transformation/Mimulus%20in%20planta%20transformation.pdf



Mimulus in planta
transformation.pdf

SAFETY WARNINGS

For Safety Warnings and Hazard Information please refer to the SDS (Safety Data Sheet).

Agro Transformation

1

Please select from the following two options:

1. Agro Transformation - Using Electroporator
2. Agro Transformation - Using Freeze -Thaw Method

step case

Agro Transformation - Using Electroporator

Select this option for agro transformation steps using an electroporator.



2

Thaw agro competent cells on ice (stored at **-80 °C**)– Agrobacterium GV3101

3

Chill 2-mm electroporation cuvette on ice.

4

Aliquot **1 ml LB** into eppie tube.

- 5 Add  **1.5 µl plasmid DNA** to agro cells.
- 6 Transfer the agro + plasmid DNA to the cuvette.
- 7 Electroporate: (Bio-Rad Electroporator program Agr)

Capacitance: 25 µF

Voltage: 2.4 kV

Resistance: 200 Ohm




Pulse length: 5 msec
- 8 Immediately add  **1 ml LB** to the cuvette.
- 9 Transfer to an eppie tube and shake at  **28 °C**-  **30 °C** for 1-2 hrs  **01:00:00** .
- 10 Plate  **100 µl** on LB + Kan + Gent + Rif plate and incubate for  **48:00:00** at  **28 °C**-  **30 °C** .

step case

Agro Transformation - Using Freeze -Thaw Method

Select this option for agro trasformation steps using a Freeze -Thaw Method.



- 2 Thaw competent Agrobacterium on ice (use  **250 µl** per transformation reaction) and add  **10 µl standard E. coli miniprep plasmid**.
- 3 Incubate the mixture on ice for  **00:05:00** .



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