

## Protocol for the production of Mu transpososomes using Domus v.3 MuA Transposase (in vivo Integrator)

### DESCRIPTION

Domus v.3 MuA Transposase (in vivo Integrator) is a hyperactive MuA transposase variant (1). It has a superior performance especially in genomic integration assays, both with bacteria and eukaryotic cells. When v.3 MuA Transposase is incubated *in vitro* with Mu transposon in the absence of Mg<sup>2+</sup>, stable transpososomes are formed. These complexes can be electroporated into living cells, where they encounter Mg<sup>2+</sup> and become activated for transposition. Subsequently, transpososomes are able to integrate the delivered transposon DNA into the host chromosome. Transposons can contain any DNA (e.g. selectable markers or other genetic elements), between MuA binding sites at both ends.

### PROTOCOL

Reagent	Volume
1 x MuA Assembly buffer	18 $\mu$ l
Transposon DNA (1.1 pmol/ $\mu$ l)	1 $\mu$ l
v.3 MuA Transposase (400 ng/ $\mu$ l)	1 $\mu$ l
	$\Sigma$ 20 $\mu$ l (See <b>Note 1</b> )

1. Assemble the reactions on ice without v.3 MuA Transposase.
2. Add v.3 MuA Transposase to start the reaction and incubate at 30°C, typically for 2 h (see **Note 2**).
3. Dilute the Mu transpososome preparation 1:4 or 1:8 with H<sub>2</sub>O to reduce the salt concentration onto a level suitable for electroporation.
4. Use 1  $\mu$ l of the diluted Mu transpososome preparation for electroporation into a recipient strain and plate on appropriate selection plates.

## NOTES

**Note 1.** A standard 20 µl reaction protocol is described here, but the reaction can be scaled up to reaction volume of 80 µl.

**Note 2.** Extended complex assembly time may be needed with long transposons, or if the transposon contains modified R-ends. The transpososome preparation can be flash-frozen under liquid nitrogen and stored at -80 °C for later use.

## REFERENCES

1. Rasila,T.S., Pulkkinen,E., Kiljunen,S., Haapa-Paananen,S., Pajunen,M.I., Salminen,A., Paulin,L., Vihinen,M., Rice,P.A. and Savilahti,H. (2018) Mu transpososome activity-profiling yields hyperactive MuA variants for highly efficient genetic and genome engineering. *Nucleic Acids Res.*, 46, 4649–4661.
2. Haapa-Paananen S, Savilahti H. Applications of the Bacteriophage Mu In Vitro Transposition Reaction and Genome Manipulation via Electroporation of DNA Transposition Complexes. *Methods Mol Biol.* 2018;1681:279-286.

## Citation

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## Precautions

This product is for research use only. Not for use in diagnostic or therapeutic procedures.

## Patent

v.3 MuA Transposase (in vivo Integrator) is covered by International Patent No. WO 2014/013127 AI

Please contact us for more information

[www.domusbiotechnologies.com](http://www.domusbiotechnologies.com)

email: [domus@domusbiotechnologies.com](mailto:domus@domusbiotechnologies.com)