

Oct 14, 2019

Double Digestion and Dephosphorylation of Plasmid

DOI

dx.doi.org/10.17504/protocols.io.76shree

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DOI: dx.doi.org/10.17504/protocols.io.76shree

Protocol Citation: Igem Dusseldorf 2019. Double Digestion and Dephosphorylation of Plasmid. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.76shree>

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Protocol status: Working

We use this protocol and it's working

Created: October 14, 2019

Last Modified: October 14, 2019

Protocol Integer ID: 28594

Abstract

Mix the following components gently:

	Plasmid Purified 2016.07.15	ng/uL
	Plasmid DNA Tube X	
Component	Volume (μ L)	
Sterile MilliQ Water	#VALUE!	
10x FastDigest Buffer	3.0	
Plasmid DNA	#VALUE!	
FastDigest Enzyme 1	1.0	
FastDigest Enzyme 2	1.0	
FastAP Alkaline Phosphatase	1.0	
Total Volume	30.0	
Total ng of DNA	1000.0	

Incubate at enzyme suitable conditions

Inactivate the enzymes at suitable conditions

Load on a 1% agarose gel and run for 60 min (110 V).

Cut out digested band (~150 mg) and transfer to pre-weighed tube. Check weight.

Purify the band

