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Double Digestion and Dephosphorylation of Plasmid

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ABSTRACT

Mix the following components gently:

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Protocol status: Working
 We use this protocol and it's working

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Plasmid Purified 2016.07.15	ng/uL
Plasmid DNA Tube X	
Component	Volume (μL)
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