Jun 20, 2020

Protocol for T7 Exonuclease (NEB #M0263)

New England Biolabs[1]

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1 Works for me This protocol is published without a DOI.

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ABSTRACT

T7 Exonuclease efficiently degrades nicked and linear dsDNA (with blunt or 3' overhangs) from 5' to 3' direction, leaving supercoiled dsDNA inctact.*

***Note:** For more precise results or partial digestions, we recommend titration of the enzyme to the intended substrate.

EXTERNAL LINK

https://neb.com/protocols/2019/07/24/protocol-for-t7-exonuclease-m0263

PROTOCOL CITATION

New England Biolabs 2020. Protocol for T7 Exonuclease (NEB #M0263). **protocols.io** https://protocols.io/view/protocol-for-t7-exonuclease-neb-m0263-7r7hm9n

EXTERNAL LINK

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CREATED

Sep 27, 2019

LAST MODIFIED

Jun 20, 2020

OWNERSHIP HISTORY

Sep 27, 2019 Anita Broellochs protocols.io

Jun 18, 2020 New England Biolabs Tech Support New England Biolabs

PROTOCOL INTEGER ID

28191

MATERIALS

NAME	CATALOG #	VENDOR
EDTA	17892	Thermo Fisher
T7 Exonuclease	M0263	New England Biolabs

SAFETY WARNINGS

Please see SDS (Safety Data Sheet) for hazards and safety warnings.

1 

Set-up the reaction as follows:

Components	50 µl REACTION
DNA	up to 1 µg
NEBuffer 4 (10x)	5 µl (1X)
T7 Exonuclease	1 µl (10 units)
Nuclease-free H2O	up to 50 µl

2 

Incubate at **25 °C** for **00:30:00** .

3 

Stop reaction by adding *EDTA* to at least **11 Milimolar (mM)** .

4 To clean up treated samples, we recommend using **one of the following steps**:

a. Column clean up (we recommend the Monarch® PCR & DNA Cleanup Kit, NEB #T1030) or

b. Running the reaction on an agarose gel, and then extracting the DNA (we recommend the Monarch Gel Extraction Kit, NEB #T1020), or

c. Performing a phenol/chloroform extraction followed by ethanol precipitation.