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Protocol for removing ssDNA from dsDNA or RNA Samples (NEB #M0568)

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1 Works for me dx.doi.org/10.17504/protocols.io.7ryhm7w

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ABSTRACT

The protocol described below will enable degradation of up to 20 pmol of a 25 nt ssDNA (~ 200 ng). In order to degrade larger amounts of ssDNA or ssDNAs longer than 25 nt, we recommend adding more enzyme instead of extending the reaction time. Users should note that ssDNAs longer than 25 nt may form secondary structures that hinder Thermolabile Exonuclease I activity.

EXTERNAL LINK

<https://neb.com/protocols/2018/08/06/protocol-for-removing-ssdna-from-dsdna-or-rna-samples-m0568>

MATERIALS

NAME	CATALOG #	VENDOR
NEBuffer 3.1 - 5.0 ml	B7203S	New England Biolabs
Thermolabile Exonuclease I	M0568	New England Biolabs
Nuclease-free Water	B1500	New England Biolabs

SAFETY WARNINGS

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



- 1 Prepare a **20 µl** reaction as follows:

Sample containing ssDNA (up to 20 pmol of a 25-mer)	x µl
NEBuffer 3.1 (NEB #B7203)*	2 µl**
Thermolabile Exonuclease I	1 µl
Nuclease-free water	to 20 µl***

*Most PCR buffers are compatible.

**No reaction buffer is necessary if enzyme is added to a PCR reaction.

***Scale larger reaction volumes proportionally.



- 2 Incubate at **37 °C** for **00:04:00**.

- 3 Stop reaction by heat-inactivation at **80 °C** for **00:01:00**.