

INSTRUCTIONS FOR USE

Product Name: DNase I, GMP-Grade (2 U/ μ L)**Catalog # DE01-E59C**

Plasmid template digestion:

Step 1. In a RNase-free centrifuge tube, add the following components:

1 μ L	10 x DNase I Buffer*
1 μ L	DNase I
X	RNA
Up to 10 μ L	RNase-free ddH ₂ O

* 10 X DNase I Buffer: 10 mM Tris-HCl, 2.5 mM MgCl₂, 0.5 mM CaCl₂, pH7.6 at 25°C

Step 2. Incubate the reaction at 37°C for 15 – 30 min

Step 3. To inactivate the reaction add 2.5mM EDTA solution and mix well and incubate at 65°C for 10 min. The processed template is suitable for subsequent reactions, including the capping reaction.

DNase I inactivation or inhibition:

Alternatively, DNase I can be inactivated using a phenol and chloroform extraction. The following conditions significantly inhibit DNase I activity: metal ion chelating agents, millimolar concentrations of zinc ions, 0.1% SDS, DTT, mercaptoethanol, other reducing agents, and salt concentrations above 50–100 mM.