

Dephosphorylation of 5'-ends of DNA using rSAP (M0371)

New England Biolabs

Abstract

Protocol for Dephosphorylation of 5´-ends of DNA using rSAP (M0371)

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Materials

Shrimp Alkaline Phosphatase (rSAP) - 500 units M0371S by New England Biolabs

Protocol

Step 1.

Prepare a 20 µl reaction as follows:



. M0371 no RE mixture

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P NOTES

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1 pmol of DNA ends is about 1 µg of a 3 kb plasmid

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Scale larger reaction volumes proportionally

Step 1.1.

DNA - 1 pmol of DNA ends

Step 1.2.

CutSmart Buffer (10X), 2 µl



2 μl Additional info:

Step 1.3.

rSAP, 1 unit

Step 1.4.

purified H2O to 20 μl

Step 2.

Incubate at 37°C for 30 minutes

O DURATION

00:30:00

Step 3.

Stop reaction by heat-inactivation at 65°C for 5 minutes

O DURATION

00:05:00