

illustra Shrimp Alkaline Phosphatase

Product Specification Sheet

Introduction

Product codes

E70092X

E70092Y

E70092Z

Important

Read these instructions carefully before using the products.

Intended use

The products are intended for research use only, and shall not be used in any clinical or *in vitro* procedures for diagnostic purposes.

Safety

For use and handling of the products in a safe way, refer to the Safety Data Sheets.

Description

Shrimp Alkaline Phosphatase is a high specific activity, heat-labile alkaline phosphatase that can be completely and irreversibly inactivated in Tris buffers at pH 8.0-8.5 by heating for 15 minutes at 65°C. No further treatment is necessary.

Protocols

Dephosphorylation of 5' phosphorylated ends of DNA

Step	Action										
1	Prepare the following reaction mix:										
	<table><thead><tr><th>Component</th><th>Volume</th></tr></thead><tbody><tr><td>DNA (1 mol of 5' ends)</td><td>1 μL</td></tr><tr><td>Shrimp Alkaline Phosphatase (1 U μL⁻¹)</td><td>1 μL</td></tr><tr><td>10\times Reaction Buffer</td><td>2 μL</td></tr><tr><td>Water</td><td>16 μL</td></tr></tbody></table>	Component	Volume	DNA (1 mol of 5' ends)	1 μ L	Shrimp Alkaline Phosphatase (1 U μ L ⁻¹)	1 μ L	10 \times Reaction Buffer	2 μ L	Water	16 μ L
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DNA (1 mol of 5' ends)	1 μ L										
Shrimp Alkaline Phosphatase (1 U μ L ⁻¹)	1 μ L										
10 \times Reaction Buffer	2 μ L										
Water	16 μ L										
2	Incubate at 37°C for 60 minutes.										
3	Inactivate the Shrimp Alkaline Phosphatase by heating to 80°C for 15 minutes.										

Vector dephosphorylation

Step	Action
1	Add Shrimp Alkaline Phosphatase to your restriction digest reaction in the ratio 0.2 Units SAP per Unit of restriction enzyme.
2	Incubate as required for the restriction digest to run to completion, then heat inactivate as recommended for the restriction enzyme.

Rapid dephosphorylation after restriction digestion

Step	Action
1	Add 5 Units Shrimp Alkaline Phosphatase to your completed restriction digest reaction.
2	Incubate for a further 10 minutes at 37°C, then heat inactivate for 10 minutes at 65°C, or as needed for inactivation of the restriction enzyme.

PCR Clean-up using illustra™ Exonuclease I and illustra Shrimp Alkaline Phosphatase.

Step	Action
1	Remove the illustra Shrimp Alkaline Phosphatase and Exonuclease I from the freezer and keep on ice whilst preparing the reaction.
2	Take a 5 µL aliquot of the completed PCR reaction mix.
3	Add 1 µL of illustra Shrimp Alkaline Phosphatase to the reaction mix.
4	Add 1 µL of illustra Exonuclease I to the reaction mix.
5	Incubate at 37°C for 15 minutes.
6	Incubate at 80°C for 15 minutes to inactivate the enzymes.

This reaction is scalable for larger quantities of PCR reaction mix. Simply add Alkaline Phosphatase and Exonuclease I in proportion to the volume of PCR reaction mix required.



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