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## Certificate of Analysis

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## Sca I

Catalog No: 2378

**Lot No:** See Product Label

**Package Size:** See Product Label

**Concentration:** See Product Label

**Source:** *Streptomyces caespitosus*

**Storage Conditions:** Store at -20°C

### Recognition/Cut Site

5'-AGT↓ACT-3'  
3'-TCA↑TGA-5'

### Unit Definition

One unit is the amount of enzyme required to completely digest 1 µg of lambda DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

### Reaction Temperature

37°C

### Heat Inactivation

10 minutes at 80°C

### Assay Conditions

50 mM Tris-HCl (pH 7.4)

6 mM MgCl<sub>2</sub>

50 mM NaCl

50 mM KCl

1 µg of lambda DNA

Reaction volume 50 µl

### Storage Buffer

50 mM Tris-HCl (pH 7.5)

50 mM KCl

0.1 mM EDTA

1 mM DTT

500 µg/ml Bovine Serum Albumin

50% (v/v) Glycerol

### Quality Control

**Endonuclease:** Incubation of 20, 35, and 50 units of Sca I with 1 µg of Lambda DNA at 37°C for 5 hours (a 250-fold over-digestion) resulted in the same sharp characteristic banding pattern as 4 U/µg in the standard 1 hour assay reaction, as determined by agarose gel electrophoresis. A second incubation using pBR322 DNA as the DNA substrate resulted in the same sharp characteristic banding pattern as 4 U/µg in the standard 1 hour assay reaction, as determined by agarose gel electrophoresis.

**Nickase:** Incubation of 20, 35, and 50 units of Sca I with 1 µg of φX-174 DNA at 37°C for 5 hours (a 250-fold over-digestion) resulted in ≤30% conversion from RF I to RF II and ≤5% conversion to RF III, as determined by agarose gel electrophoresis. Reaction volume of 50 µl.

**3'-Exonuclease:** Incubation of 5, 10, 15, 20, and 30 units of Sca I with 5 pmole of lambda/Taq I fragments (3'-labeled with Klenow exo<sup>-</sup> [<sup>3</sup>H]dCTP), incubated for 1 hour at 37°C resulted in a ≤0.2 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

**5'-Exonuclease/5'-Phosphatase:** Incubation of 5, 10, 15, 20, and 30 units of Sca I with 0.25 µg of [5'-<sup>32</sup>P]lambda/Hae III fragments for 1 hour at 37°C resulted in a ≤0.2 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

**Ligation:** Incubation of 4 units of Sca I with 1 µg of Lambda DNA at 37°C produces fragments that ligate with > 65% efficiency. The recut efficiency of these ligated fragments is 100%.

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