

## Product Information

### RESTRICTION ENDONUCLEASE Stu I

Product No. **R 8013**

Store at 0 to -20 °C

#### Product Summary

Recognition Sequence: 5'-AGG/CCT-3'

Activity: 10,000 units/ml

Cutting: 100%

Ligation: >90%

Recutting: >95%

No degradation detected with >20 units for 16 hrs.

Fold over digestion: 320 (20 units x 16 hrs.)

Package Size: 1,000 units

#### Unit Definition

One unit is the enzyme activity that completely cleaves 1 µg  $\lambda$  DNA in 1 hr. at 37 °C in a total volume of 25 µl of SB restriction enzyme buffer.

#### Specificity

Stu I recognizes the sequence AGG/CCT and generates fragments with blunt ends.<sup>1</sup> Stu I is inhibited at AGG/<sup>m</sup>C<sup>m</sup>CT by the presence of 5'-methylcytosine.

#### Comments:

Digestion Buffer SB is supplied as a 10x concentrate.

Information is not available for heat inactivation of Stu I.

#### Stu I Storage and Dilution Buffer Composition

20 mM Tris-HCl

100 mM NaCl

0.1 mM EDTA

10 mM 2-mercaptoethanol

0.01% Triton<sup>®</sup>X-100

50% (v/v) glycerol

pH 7.5

#### 1x Digestion Buffer SB (B 8781) Composition for

#### Stu I: 100% Digestion at 37 °C.

10 mM Tris-HCl

100 mM NaCl

5 mM MgCl<sub>2</sub>

1 mM 2-mercaptoethanol

pH 8.0

#### Quality Control Testing

#### Absence of non-specific endonuclease activities:

1 µg  $\lambda$  DNA is incubated for 16 hours in 50 µl of buffer SB with excess >20 units of Stu I.

**Ligation and recutting assay:** Stu I fragments, obtained by complete digestion of 1 µg  $\lambda$  DNA, adjusted to pH 7.5 at 20 °C. The Stu I fragments are then ligated with 2.0 units T4-DNA ligase at room temperature at pH 7.5. A 10 µl reaction mixture, incubated for 16 hours at pH 7.5 at room temperature, contained 2.0 units T4-DNA ligase, 66 mM Tris-HCl, 5 mM MgCl<sub>2</sub>, 1 mM dithioerythritol and 1 mM ATP.

The degree of ligation and subsequent recutting with Stu I to yield the typical pattern of  $\lambda$  x Stu I fragments is determined.

#### References

1. Shimolsu, H., et al., *Gene*, **11**, 219 (1980)