

**MBRE015****Stu I****Components**

Reagents provided	MBRE015			
	250 Units	500 Units	1000 Units	5000 Units
Stu I	12.5 µl	25 µl	50 µl	250 µl
10X HiBuffer H5	200 µl	400 µl	800 µl	4 ml
10X HiBuffer DB	200 µl	400 µl	800 µl	4 ml
Diluent E Buffer	200 µl	400 µl	800 µl	4 ml

**NOTE: BSA included in all Reaction Buffer**

**Source:** An *E.coli* strain that carries the Stu I gene from *Streptomyces tubercidicus*.

**Concentration:** 20 U/µl

**Unit Definition:**

1 U is defined as the amount of enzyme that is required to digest 1µg of DNA in 1 hour at 50°C in 50 µl of assay buffer.

Enzyme	Optimum reaction temperature (°C)	Thermal Inactivation (°C)	% activity of Buffers				
			H1	H2	H3	H4	H5
Stu I	50	80	75	75	75	100	100

**Reaction Buffer:****10X HiBuffer H5:**

30mM Tris-acetate (pH 7.9 at 30°C), 10mM Mg-acetate, 60mM K-acetate and 100 µg/ml BSA.

**NOTE:** Incubate at 50°C.

**Storage Buffer:**

10mM Tris-HCl (pH 7.5), 100mM NaCl, 0.1mM EDTA, 7mM 2-mercaptoethanol, 100µg/ml BSA and 50% glycerol. Store at -20°C.

**NOTE: 10X HiBuffer DB is provided for double digestion.**

**Quality Control Assays:****Ligation / Recutting Assay:**

After 10-fold over digestion with Stu I, 70% of the DNA fragments can be ligated and recut.

**Over digestion Assay:**

An unaltered banding pattern was observed after 1µg of DNA was digested with 5U of Stu I for 16 hours at 50°C.

### Example of Digestion conditions:

- Enzyme : 1  $\mu$ l
- Lambda 0.3 mg/ml : 3  $\mu$ l ( $\approx$ 1  $\mu$ g DNA)
- 10X HiBuffer H5 : 5  $\mu$ l
- Nuclease free water : upto 50  $\mu$ l



Lambda DNA, 0.7% agarose  
**Lane 1:** Digestion after 1 hour  
**Lane 2:** Digestion after 16 hours

### Note:

- Total reaction volume is dependent on the experiment.
- The amount of enzyme to be used is dependent on the DNA template.
- For plasmid DNA, 5-10X more enzyme is required.
- High enzyme concentration may result in **Star activity**.

**Storage conditions:** Stu I should be stored at  $-20^{\circ}\text{C}$ . When stored under the recommended conditions, the product is stable for 2 years.

### Technical Assistance

At HiMedia, we pride ourselves on the quality and availability of our technical support. For any kind of technical assistance, mail at [mb@himedialabs.com](mailto:mb@himedialabs.com).

MBRE015\_O/0517

MBRE015-07



Consult instructions for use



Do not use if package is damaged



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