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 µl
- Lambda 0.3 mg/ml : 3 µl (≈1 µg DNA)
- 10X HiBuffer H5 : 5 µl
- Nuclease free water : upto 50 µl

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°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.

MBRE015_O/0517 MBRE015-07

Consult instructions for use

Do not use if package is damaged

HiMedia Laboratories Pvt. Limited,
Reg. Off: 23 Vadhani Industrial Estate,
LBS Marg, Mumbai - 400086,
India Works: B/4-6, M.I.D.C., Dindori,
Nashik, India (or respective plant address)
Customer Care No: 022-6116 9797
www.himedialabs.com