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# **ProductInformation**

#### **RESTRICTION ENDONUCLEASE MIu I**

Product Number R 8257

Store at 0 °C to -20 °C

#### **Product Summary**

Recognition Sequence: 5' A/CGCGT'3

Activity: 10,000 units/ml

Cutting: 100% Ligation: >95% Recutting: >95%

No degradation detected with >30 units for 16 hrs.

Fold over digestion: 480 (30 units x 16 hrs.)

Package Size: 500 units

# **Specificity**

Mlu I recognizes the sequence A/CGCGT and generates fragments with 5'-cohesive termini. Mlu I generates compatible ends to BssHII. Mlu I is inhibited at A<sup>m</sup>CGCGT by the presence of 5'-methylcytosine. Mlu I is not influenced by the presence of N<sup>6</sup>-methyladenine.

#### **Unit Definition:**

One unit is the enzyme activity that completely cleaves  $1\mu g \ \lambda$  DNA in 1 hr. At 37 °C in a total volume of 25  $\mu$ l of buffer SH for restriction endonucleases.

### Comments:

Digestion Buffer SH is supplied as 10× concentrate.

Information for heat inactivation of Mlu I is not available.

# Mlu I Storage and Dilution Buffer Composition

20 mM Tris-HCl 50 mM KCl 0.1 mM EDTA 10 mM 2-mercaptoethanol 50% (v/v) glycerol pH 8.0

# **Quality Control Testing**

 $1 \times$  Digestion Buffer SH (B 3657) Composition for MIu I: 100% Digestion at 37 °C.

50 mM Tris-HCl 100 mM NaCl 10 mM MgCl<sub>2</sub>

1 mM dithioerythritol (DTE)

pH 7.5

#### Absence of non-specific endonuclease activities

1  $\mu g~\lambda$  DNA is incubated for 16 hrs. in 50  $\mu l$  of buffer SH with excess units of Mlu I.

## Ligation and recutting assay

Mlu I fragments, obtained by complete digestion of 1  $\mu$ g  $\lambda$  DNA, are adjusted to pH 7.5 at 20 °C. The Mlu I fragments are then ligated with 0.5 units T4-DNA ligase at pH 7.5 at 20 °C. A 10  $\mu$ I reaction mixture, incubated for 16 hrs. at 20 °C contained 0.5 units T4-DNA ligase, 66 mM Tris-HCl, 5 mM MgCl<sub>2</sub>, 5 mM dithioerythritol, 1 mM ATP.

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

#### References

1. Sugisaki, H. and Kanazawa, S., Gene, 16, 73(1981).

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