

GFast Gene **Restriction Enzyme** Nco I

Cat.#	Size	Conc.
FG-Ncol	1,000 units	10 units/µl

Store at -20°C

Supplied with: 10X FastGene® Buffer III (FG-REB3) 10X FastGene® FastCut Buffer (FG-REBHF) 6X DNA Loading Buffer Sterile water

Recognition site

For Research Use Only. Not for use in diagnostic procedures. ISO9001

Source: Nocardia corallina

III (37°) 65°

Reaction conditions 1X FastGene® Buffer III, 37°C 1X FastGene® FastCut Buffer, 37°C

FastGene® FastCut Buffer

FastGene® restriction enzyme can cut substrate DNA in 5-15 min with FastGene® FastCut Buffer.

1X FastGene® Buffer III

50 mM Tris-HCl (pH 7.9 at 25°C) 100 mM NaCl 10 mM MaCl₂ 100 µg/ml BSA

Unit definition

One unit is defined as the amount of enzyme required for complete digestion of 1 μg bacteriophage λ at 37°C for 1 hr in 50 µl reaction mixtures.

Quality control

- Unit definition assay
- Overdigestion assay - Endonuclease assay
- Extreme pure assay

Dilution buffer FastGene® Diluent A

Heat Inactivation Nco I can be inactivated at 65°C for 20 min.

Methylation sensitivity

dam methylation: Not sensitive dcm methylation: Not sensitive CpG methylation: Not Sensitive

Prolonged incubation

A minimum amount of enzyme required to digest 1 µg substrate DNA for 16 hr: 0.25 U.

Relative activity in FastGene® Buffers

FastGene® Buffer I: 50% FastGene® Buffer II: 100% FastGene® Buffer III: 100% FastGene® Buffer IV: 75% FastGene® FastCut Buffer: 100%

Note

It is not affected by *dam. dcm* or mammalian CpG methylation. Its recognition sequence includes ATG, and therefore it is possible to express a target protein without additional amino acids after cloning an Nco I-cleaved fragment to the initiation site of an expression vector.

Standard reaction condition

Normal protocol

Component	Final Conc.	Volume
Substrate DNA	1 µg	Xμl
10X FastGene [®] Buffer III	1 X	5 µl
Nco I	10 unit	1 µl
Sterile water		up to 50 µl
→ Incubate at 37°C for 1 hr		

Fast protocol

Component	Final Conc.	Volume
Substrate DNA	1 µg	Xμl
10X FastGene® FastCut Buffer	1 X	5 µl
Nco I	10 unit	1 µl
Sterile water		up to 50 µl
→ Incubate at 37°C for 15 mir	า	

X We recommend 5-10 units of enzyme per μg DNA and 10-20 units for genomic DNA in a 1 h digest

enetics NIPPON Genetics EUROPE GmbH

www.nippongenetics.eu www.n-genetics.com

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1X FastGene® FastCut Buffer, 37°C

FastGene® FastCut Buffer

FastGene® restriction enzyme can cut substrate DNA in 5-15 min with FastGene® FastCut Buffer.

1X FastGene® Buffer III

50 mM Tris-HCI (pH 7.9 at 25°C) 100 mM NaCl 10 mM MqCl₂ 100 µg/ml BSA

Unit definition

One unit is defined as the amount of enzyme required for complete digestion of 1 μ g bacteriophage λ at 37°C for 1 hr in 50 µl reaction mixtures.

Quality control

- Unit definition assay
- Overdigestion assay

Dilution buffer FastGene® Diluent A

Heat Inactivation Nco I can be inactivated at 65°C for 20 min.

dcm methylation: Not sensitive

Prolonged incubation

A minimum amount of enzyme required to digest 1 µg substrate DNA for 16 hr; 0.25 U.

Relative activity in FastGene[®] Buffers

FastGene [®] Buffer I:	50%
FastGene [®] Buffer II:	100%
FastGene [®] Buffer III:	100%
FastGene [®] Buffer IV:	75%
FastGene [®] FastCut Buffer:	100%

Note

It is not affected by dam, dcm or mammalian CpG methylation. Its recognition sequence includes ATG, and therefore it is possible to express a target protein without additional amino acids after cloning an Nco I-cleaved fragment to the initiation site of an expression vector.

Standard reaction condition

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Component	Final Conc.	Volume
Substrate DNA	1 µg	Xμl
10X FastGene [®] Buffer III	1 X	5 µl
Nco I	10 unit	1 µl
Sterile water		up to 50 µl
\rightarrow Incubate at 37°C for 1 hr		

- Fast protocol

	Volume
1 µg	X µl
1 X	5 µl
10 unit	1 µl
	up to 50 µl
	1 X

→ Incubate at 37°C for 15 min

※ We recommend 5-10 units of enzyme per μg DNA and 10-20 units for genomic DNA in a 1 h digest

Methylation sensitivity

CpG methylation: Not Sensitive

dam methylation: Not sensitive

- Endonuclease assav

- Extreme pure assay