

CspCI



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www.neb.com



R0645S 004120613061

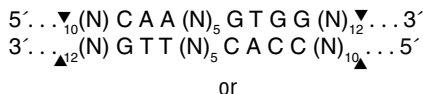
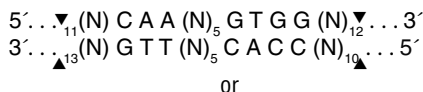
R0645S



500 units 5,000 U/ml Lot: 0041206

RECOMBINANT Store at -20°C Exp: 6/13

Recognition Site:



New Reaction Buffer

CspCI



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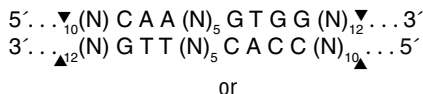
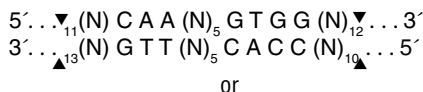
R0645S



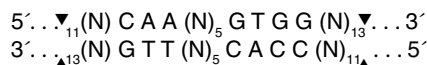
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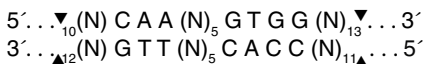
Recognition Site:



New Reaction Buffer



or

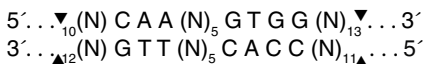
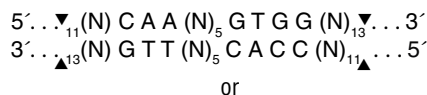


Source: An *E. coli* strain that carries the cloned CspCI gene from *Citrobacter* species 2144 (C. Nkenfou)

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 7.5), 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol.

Reagents Supplied with Enzyme: 10X NEBuffer 4, 1600X S-adenosylmethionine (SAM) (32 mM).

Reaction Conditions: 1X NEBuffer 4, supplemented with 20 µM S-adenosylmethionine (SAM supplied). Incubate at 37°C.



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1X NEBuffer 4:

50 mM potassium acetate
20 mM Tris-acetate
10 mM magnesium acetate
1 mM dithiothreitol
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to digest 1 µg of λ DNA in 1 hour at 37°C in a total reaction volume of 50 µl.

Diluent Compatibility: Diluent Buffer A, 50 mM KCl, 10 mM Tris-HCl, 0.1 mM EDTA, 1 mM dithiothreitol, 200 µg/ml BSA and 50% glycerol (pH 7.4 @ 25°C)

Quality Control Assays

16-Hour Incubation: A 50 µl reaction containing 1 µg of λ DNA and 10 units of enzyme incubated for 16 hours resulted in the same pattern of DNA bands as a reaction incubated for 1 hour with 1 unit of enzyme.

Exonuclease Activity: Incubation of 50 units of enzyme with 1 µg sonicated [³H] DNA (10⁶ cpm/µg) for 4 hours at 37°C in 50 µl reaction buffer released < 0.1% radioactivity.

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Enzyme Properties

Activity in NEBuffers:

NEBuffer 1	10%
NEBuffer 2	100%
NEBuffer 3	10%
NEBuffer 4	100%

When using a buffer other than the optimal (supplied) NEBuffer, it may be necessary to add more enzyme to achieve complete digestion.

Survival in a Reaction: A minimum of 0.5 unit is required to digest 1 µg of substrate DNA in 16 hours.

Heat Inactivation: 65°C for 20 minutes.

Notes: S-adenosylmethionine or SAM is supplied as a 32 mM solution in 0.005 M sulfuric acid and 10% ethanol. Under these conditions SAM is stable for up to 6 months when stored at -20°C.

CspCI cleaves DNA substrates twice to excise its recognition site generating a 35 base-pair fragment with 2-base 3' overhangs.

(see other side)

CERTIFICATE OF ANALYSIS

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NEBuffer 3	10%
NEBuffer 4	100%

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Survival in a Reaction: A minimum of 0.5 unit is required to digest 1 µg of substrate DNA in 16 hours.

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CERTIFICATE OF ANALYSIS

The cleavage point may shift one base pair depending on the DNA sequence context before and after the recognition site. For a given sequence, one site will predominate. For details, see www.neb.com.

Requires S-adenosylmethionine for optimal activity (supplied with enzyme).


 = Time-Saver™ Qualified (See www.neb.com for details).

U.S. Patent No. 7,247,464

Page 2 (R0645)

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Page 2 (R0645)