



CERTIFICATE OF ANALYSIS

Thermolabile
Alkaline
Phosphatase



S

E365

200 units
5,000 u/ml

Lot: 25

Exp: 07/19

Store at -20C

Source:

An *E.coli* strain that carries the cloned Alkaline Phosphatase gene from *Alteromonas undina* P2.

Description:

Thermolabile Alkaline Phosphatase (TAP) catalyzes the removal of 5'-phosphate groups from DNA and RNA.

Storage Conditions:

20 mM Tris-HCl (pH 7.6), 0.1 mM ZnCl₂, 50% glycerol.
Store at - 20°C

Reaction Conditions:

1x SE-Buffer W. Incubate at 25° C.

1X SE-Buffer W (pH 8.5@ 25° C):

10 mM Tris-HCl, 100 mM NaCl, 10 mM MgCl₂, 1 mM DTT.

Applications:

- Removing 5' phosphoryl groups from nucleic acids
- Preparing templates for 5' end labeling
- Preventing fragments from self ligating

Unit Definition:

One unit is defined as the amount of enzyme that will dephosphorylate 1µg of pUC19 DNA (linearized with Hind III) in 30 minutes at 25° C. Dephosphorylation is defined as > 95 % inhibition of recirculation in a self-ligation reaction that is measured by transformation into *E.coli*.

Quality Control Assays:

Nonspecific endonuclease assay:

No appearance of nicked DNA was detected after incubation of 1µg of supercoiled form pUC19 DNA with 5 units of enzyme for 4 hours at 25° C.

Nonspecific endo- and exonuclease assay:

No alteration of the pattern of DNA bands was

detected after incubation of 1µg λ/HindIII DNA fragments with 5 units of enzyme in 50 µl of reaction mixture for 4 hours at 25° C.

Heat Inactivation:

Enzyme is inactivated by incubation at 65° C for 20 minutes.

Reagents Supplied with Enzyme: 10X SE- Buffer W.

Notes: Thermolabile Alkaline Phosphatase is also active in SE-Buffers B,G,O and Y.

Vector Dephosphorylation Protocol:

1. Add 1/10 volume of 10X SE-Buffer W to 0,5-1 µg of DNA cut with any restriction endonuclease in any buffer.
2. Add 1µl of TAP (5 units) and mix.
3. Incubate for 15 or 30 min at 25° C.
4. Heat inactivate for 20 minutes at 65° C.
5. Proceed with ligation.

For more details
scan the code



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