

## pTXB1 Vector



1-800-632-7799  
info@neb.com  
www.neb.com



N6707S 006150518051

# N6707S

**10 µg**      **Lot: 0061505**      **Exp: 5/18**

**0.20 mg/ml**      **Store at -20°C**

**Description:** pTXB1 is an *E. coli* expression vector in the IMPACT™ Kit (1,2). It is designed for the in-frame insertion of a target gene into the polylinker upstream of the Mxe intein/chitin binding domain (27 kDa)(2,3). The fusion protein is bound to chitin beads and the thiol-induced cleavage activity of the intein releases the target protein. pTXB vectors are recommended for use in intein-mediated protein ligation and C-terminal labeling (2). This double stranded vector is 6,706 base pairs in length.

**Source:** pTXB1 contains the intein (198 amino acids) from the *Mycobacterium xenopi* GyrA gene (2,4).

Supplied in: 10 mM Tris-HCl (pH 8.0), 1 mM EDTA.

### Features of this vector:

- The NdeI site in the polylinker contains an ATG sequence for translation initiation.

### Polylinker Region:

pTXB1

T7 Universal Primer →

5'...CGG GGA TCT CGA TCC CGC GAA ATT AAT ACG ACT CAC TAT AGG GGA ATT GTG AGC  
T7 Promoter lac operator

GGA TAA CAA TTC CCC TCT AGA AAT AAT TTT GTT TAA CTT TAA GAA GGA GAT ATA  
XbaI ShineDalgarno

Met Ala Ser Ser Arg Val Asp Gly Gly Arg Glu Phe Leu Glu Gly Ser Ser Cys1  
CAT ATG GCT AGC TCG CGA GTC GAC GGC GGC CGC GAA TTC CTC GAG GGC TCT TCC TGC

NdeI NheI NruI SalI NotI EcoRI XhoI SapI

▼ Intein →

ATC ACG GGA GAT GCA CTA GTT GCC CTA CCC GAG GGC GAG TCG GTA  
SpeI

CGC ATC GCC GAC ATC GTG CCG ...3'

- Origin of DNA replication from bacteriophage M13, which allows for the production of single-stranded DNA by helper phage superinfection of cells bearing the plasmid (M13K07 Helper Phage, NEB #N0315).
- Ampicillin resistance
- Other IMPACT vectors are available which allow for fusion of a target gene to N- or C-terminus of an intein. The cleavage reaction may be induced by thiol reagent or temperature/pH shift.
- Companion vector pTXB3 (NEB #N6708) contains an NcoI site in place of NdeI.
- A wide range of *E. coli* host strains: T7 Express Competent *E. coli* (High Efficiency) (NEB #C2566) or BL21(DE3) Competent *E. coli* (NEB #C2527) and derivatives.

(see other side)

CERTIFICATE OF ANALYSIS

## pTXB1 Vector



1-800-632-7799  
info@neb.com  
www.neb.com



N6707S 006150518051

# N6707S

**10 µg**      **Lot: 0061505**      **Exp: 5/18**

**0.20 mg/ml**      **Store at -20°C**

**Description:** pTXB1 is an *E. coli* expression vector in the IMPACT™ Kit (1,2). It is designed for the in-frame insertion of a target gene into the polylinker upstream of the Mxe intein/chitin binding domain (27 kDa)(2,3). The fusion protein is bound to chitin beads and the thiol-induced cleavage activity of the intein releases the target protein. pTXB vectors are recommended for use in intein-mediated protein ligation and C-terminal labeling (2). This double stranded vector is 6,706 base pairs in length.

**Source:** pTXB1 contains the intein (198 amino acids) from the *Mycobacterium xenopi* GyrA gene (2,4).

Supplied in: 10 mM Tris-HCl (pH 8.0), 1 mM EDTA.

### Features of this vector:

- The NdeI site in the polylinker contains an ATG sequence for translation initiation.

### Polylinker Region:

pTXB1

T7 Universal Primer →

5'...CGG GGA TCT CGA TCC CGC GAA ATT AAT ACG ACT CAC TAT AGG GGA ATT GTG AGC  
T7 Promoter lac operator

GGA TAA CAA TTC CCC TCT AGA AAT AAT TTT GTT TAA CTT TAA GAA GGA GAT ATA  
XbaI ShineDalgarno

Met Ala Ser Ser Arg Val Asp Gly Gly Arg Glu Phe Leu Glu Gly Ser Ser Cys1  
CAT ATG GCT AGC TCG CGA GTC GAC GGC GGC CGC GAA TTC CTC GAG GGC TCT TCC TGC

NdeI NheI NruI SalI NotI EcoRI XhoI SapI

▼ Intein →

ATC ACG GGA GAT GCA CTA GTT GCC CTA CCC GAG GGC GAG TCG GTA  
SpeI

CGC ATC GCC GAC ATC GTG CCG ...3'

- Origin of DNA replication from bacteriophage M13, which allows for the production of single-stranded DNA by helper phage superinfection of cells bearing the plasmid (M13K07 Helper Phage, NEB #N0315).
- Ampicillin resistance
- Other IMPACT vectors are available which allow for fusion of a target gene to N- or C-terminus of an intein. The cleavage reaction may be induced by thiol reagent or temperature/pH shift.
- Companion vector pTXB3 (NEB #N6708) contains an NcoI site in place of NdeI.
- A wide range of *E. coli* host strains: T7 Express Competent *E. coli* (High Efficiency) (NEB #C2566) or BL21(DE3) Competent *E. coli* (NEB #C2527) and derivatives.

(see other side)

CERTIFICATE OF ANALYSIS

## References:

1. Chong, S., Mersha, F.B., Comb, D.G., Scott, M. E., Landry, D., Vence, L.M., Perler, F.B., Benner, J., Kucera, R.B., Hirvonen, C.A., Pelletier, J.J., Paulus, H. and Xu, M.-Q. (1997). Single-column purification of free recombinant proteins using a self-cleavable affinity tag derived from a protein splicing element. *Gene* 192, 271–281.
2. Evans, T.C., Benner, J. and Xu, M.-Q. (1998). Semisynthesis of cytotoxic proteins using a modified protein splicing element. *Protein. Sci.* 7, 2256–2264.
3. Watanabe, T., Ito, Y., Yamada, T., Hasmimoto, M., Sekine, S. and Tanaka, H. (1994). The role of the C-terminal domain and type III domains of chitinase A1 from *Bacillus circulans* WL-12 in chitin degradation. *J. Bacteriol.* 176, 4465–4472.
4. Telenti, A., Southworth, M., Alcaide, F., Daugelat, S., Jacobs, W.R. Jr. and Perler, F.B. (1997). The *Mycobacterium xenopi* GyrA protein splicing element: Characterization of a minimal intein. *J. Bacteriol.* 179, 6378–6382.
5. Dubendorff, J.W. and Studier, F.W. (1991). Controlling basal expression in an inducible T7 expression system by blocking the target T7 promoter with *lac* repressor. *J. Mol. Biol.* 219, 45–59.

Additional information such as vector sequences and frequently asked questions, are available at [www.neb.com](http://www.neb.com).



NEW ENGLAND BIOLABS® is a registered trademark of New England Biolabs, Inc.

U.S. Patent Nos. 5,496,714, 5,834,247, 6,569,669

Notice to Buyer/User: The buyer/user has a non-exclusive license for Research Purposes Only. See RESEARCH USE ASSURANCE STATEMENT on [neb.com](http://neb.com).