# pBacPAK-Nterm 6xHN-GUS Vector Information



TAATTAATTA ATTGATCCGG GTTATTAGTA CATTTATTAA GCGCTAGATT CTGTGCGTTG TTGATTTACA GACAATTGTT GTACGTATTT TAATAATTCA

Figure 1. Restriction Map and Multiple Cloning Site (MCS) of pBacPAK-Nterm 6xHN-GUS Vector.\* pBacPAK-Nterm 6xHN-GUS contains coding sequences for beta-glucuronidase cloned into the *Sal* I and *Hind* III sites of pBacPAK-Nterm 6xHN Vector. The unshaded and filled arrows on the upstream and downstream side of the GUS insert, respectively, represent the positions of the recommended forward (Bac1) and reverse (Bac2) primers.

# Description

Available as part of the In-Fusion<sup>™</sup> Ready BacPAK Vector Set (Cat. No. 631410), pBacPAK-Nterm 6xHN-GUS Vector is a positive control transfer vector designed for high-level expression of 6xHN-tagged beta-glucuronidase driven by the strong AcMNPV polyhedrin promoter. Flanking AcMNPV sequences allow recombination with viral DNA to transfer the expression cassette to the polyhedrin locus of the viral DNA.

This vector can be used to monitor transfection efficiency and virus production. Expression of GUS can be easily detected by addition of X-Gluc to the medium.

pBacPAK-Nterm 6xHN Vector has a pUC origin of replication, an M13 origin for single-stranded DNA production, and an ampicillin resistance gene in *E. coli*.

<u>Vector Information</u>



Clon**tech** 

United States/Canada 800.662.2566 Asia Pacific +1.650.919.7300 Europe +33.(0)1.3904.6880 Japan +81.(0)77.543.6116

Clontech Laboratories, Inc. ATakara Bio Company 1290 Terra Bella Ave. Mountain View, CA 94043 Technical Support (US) E-mail: tech@clontech.com www.clontech.com

# Location of features

- AmpR: 5426-6286
- AcMNPV sequences: 1–1146
- 6xHN tag: 1155-1190
- Enterokinase site: 1200-1214
- Beta glucuronidase (GUS): 1227-3032
- AcMNPV sequences: 3083-5334
- M13 single-strand DNA origin: 4566–5039
- ORF1629 C-terminus: 63-1406 (complementary)
- Polyhedrin mRNA polyadenylation signal: 3409-3414
- Transcription start point: 1098–1098
- Polyhedrin promoter: 1076-1145
- pUC plasmid replication origin: 6434–7077

# **Primer Locations**

Bac1 Primer: 1077-1096	aaccatctcgcaaataaata
Bac2 Primer: 3291–3272	acgcacagaatctagcgctt

# Propagation in *E. coli*

- Suitable host strains: DH5 $\alpha$  and other general-purpose strains. Single-stranded DNA production requires a host containing an F plasmid such as JM101 or XL1-Blue.
- Selectable marker: plasmid confers resistance to ampicillin (100 µg/ml) in E. coli hosts.
- *E. coli* replication origin: pUC
- Copy number: high

## References

See the In Fusion Ready BacPAK Vector Set User Manual (PT3908-1) for In-Fusion cloning techniques. See the BacPAK Baculovirus Expression System User Manual (PT1260-1) for generation of baculovirus and related protocols. See the BacPAK Baculovirus Rapid Titer Kit User Manual (PT3153-1) for titration of recombinant baculoviruses.

\*Note: The attached sequence file has been compiled from information in the sequence databases, published literature, and other sources, together with partial sequences obtained by Clontech. This vector has not been completely sequenced.

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