

Technical Data Sheet

pAcGHLT-A,B,C Baculovirus Transfer Vector Set

Product Information

Material Number:	554792
Component:	51-21460P
Description:	pAcGHLT-A
Size:	20 µg (1 ea)
Concentration:	1.0 mg/ml
Component:	51-21461P
Description:	pAcGHLT-B
Size:	20 µg (1 ea)
Concentration:	1.0 mg/ml
Component:	51-21462P
Description:	pAcGHLT-C
Size:	20 µg (1 ea)
Concentration:	1.0 mg/ml

Description

The pAcGHLT-A,B,C baculovirus transfer vectors are derivatives of the pAcG1 vector. They contain a 6xHis[†] tag and a glutathione S-transferase (GST) tag upstream of the MCS and will express the desired protein as a 6xHis-containing GST fusion protein. 6xHis fusion proteins bind with high affinity to Ni-NTA Agarose[†] and GST fusion proteins have a high affinity for reduced glutathione. Therefore, a highly efficient single step affinity purification can be done on GST-6xHis-tagged proteins using either metal chelate matrices (Ni-NTA Agarose[†]) or glutathione agarose beads. A protein kinase A site follows the 6xHis sequence in the plasmids. This allows the purified proteins to be phosphorylated with protein kinase A. This phosphorylation should not alter the binding affinity of the recombinant protein to any of its ligands. After purification, the GST and 6xHis tags can be removed by incubating the fusion protein in the presence of the proteinase cleavage enzyme thrombin. All foreign inserts must be in frame with the GST open reading frame (ORF). These vectors are derived from the polyhedrin locus and are recommended for use with BD BaculoGold™ baculovirus DNA (Cat. No. 554739).

Preparation and Storage

Store undiluted at -20° C.

The plasmid DNA has been purified using a silica bead matrix and dissolved in TE buffer (10 mM Tris-HCl pH 7.5; 1 mM EDTA).

Application Notes

Recommended Assay Procedure:

For expression of a fusion protein under the polyhedrin promoter, insert your gene of interest into a suitable restriction site that is in frame with the GST ORF in the vector. Transform and amplify the plasmid DNA in E. coli strains (DH5α, HB101 or other suitable strain) under ampicillin selection. For generation of recombinant virus, perform a co-transfection of the pAcGHLT vector and linearized baculovirus DNA (BD BaculoGold™ viral DNA Cat. No. 554739) into a susceptible cell line (Sf9 or Sf21). Monoclonal antibodies to GST (clone G172-1138, Cat. No. 554805 or clone B19-2, Cat. No. 554824) may be used to detect recombinant GST-fusion proteins by western blot analysis.

*Please visit us at www.bdbiosciences.com to view or download a Baculovirus Expression Vector System Manual.

[†]The 6xHis Vectors included in the kit were developed by BD Biosciences Pharmingen and produced in collaboration with QIAGEN. The Ni-NTA Agarose is produced by QIAGEN.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554739	Linearized Baculovirus DNA	5 transfections	(none)
560129	Transfection Kit	5 transfections	(none)

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www.bdbiosciences.com

United States 877.232.8995 Canada 888.259.0187 Europe 32.53.720.550 Japan 0120.8555.90 Asia Pacific 65.6861.0633 Latin America/Caribbean 55.11.5185.9995

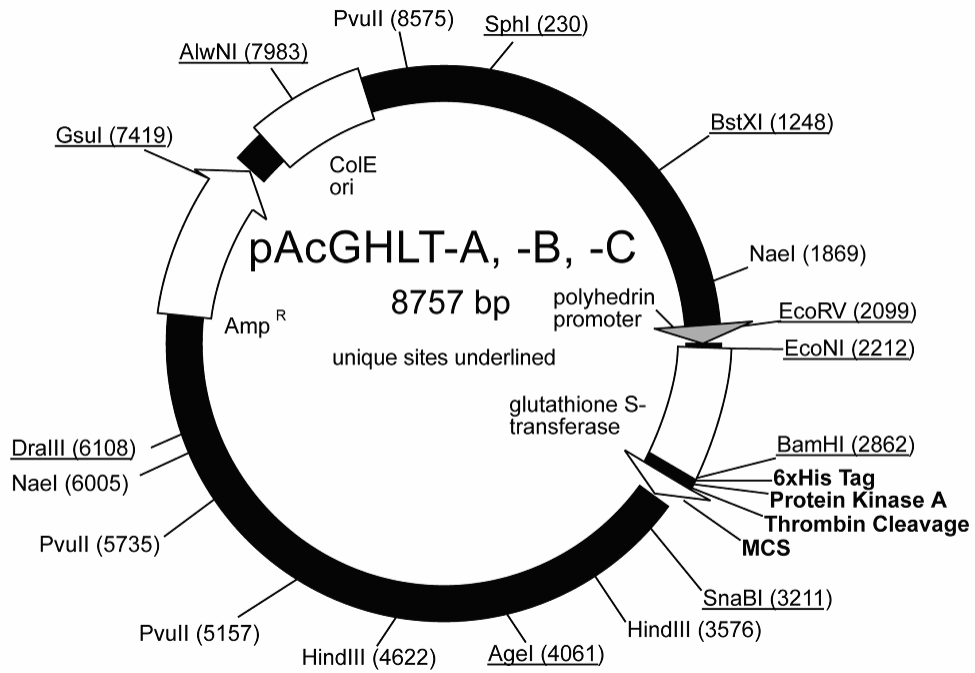
For country-specific contact information, visit www.bdbiosciences.com/how_to_order/

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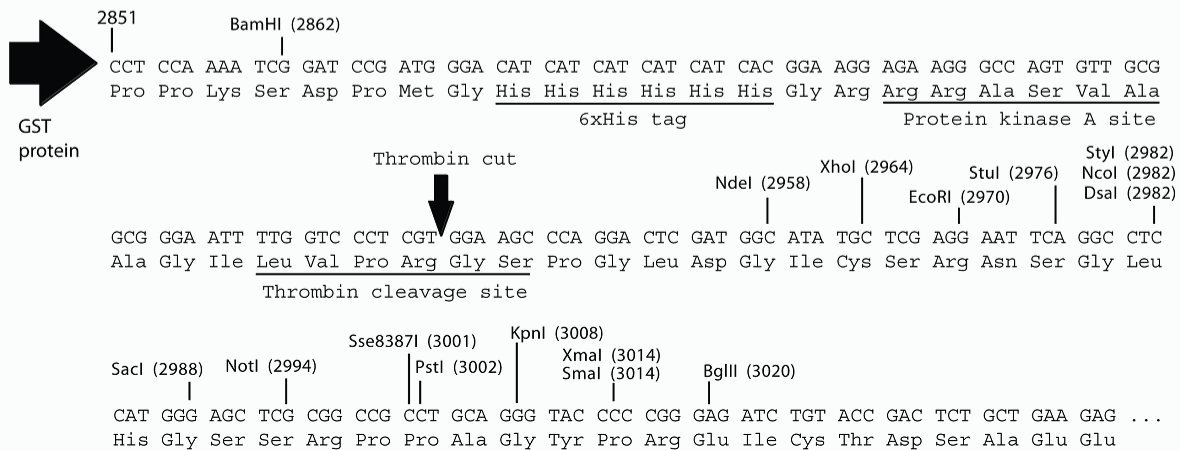
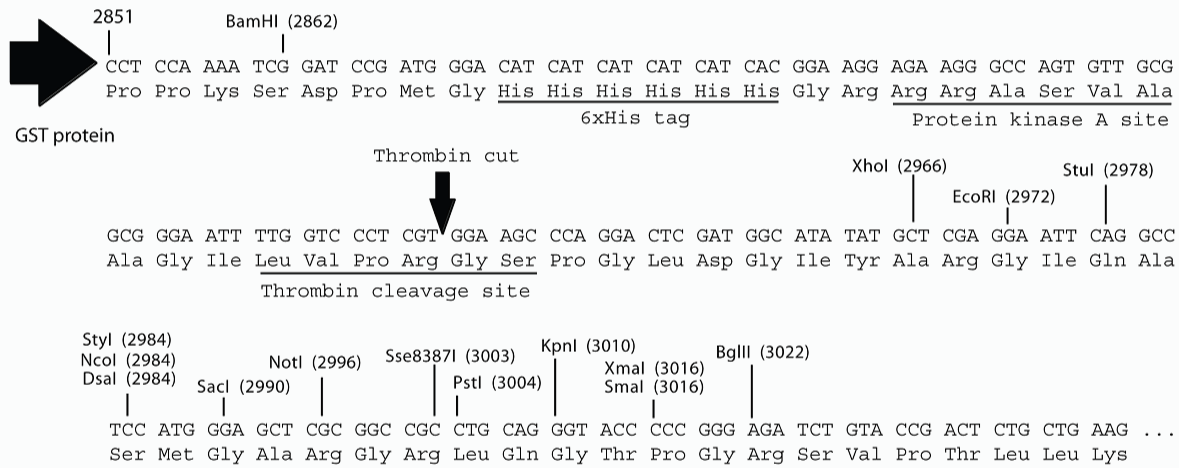
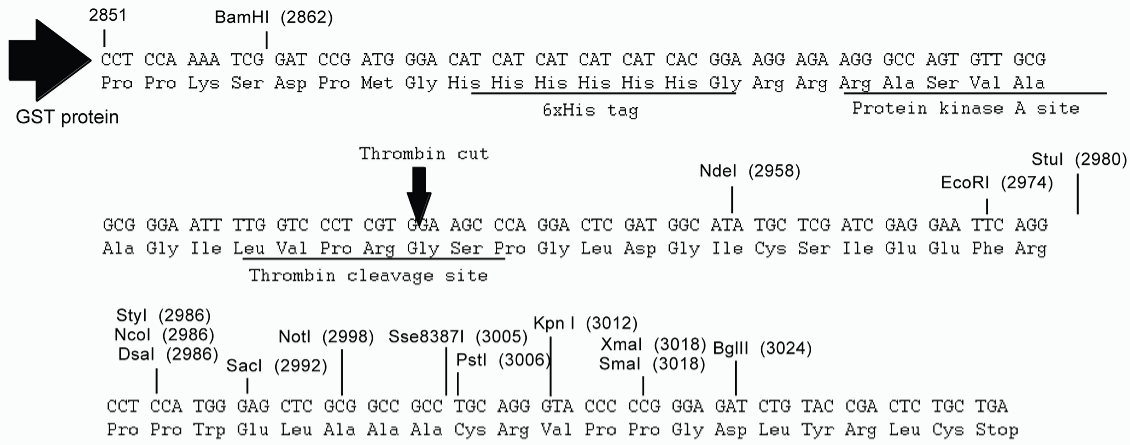




Plasmid Map of the Baculovirus Transfer Vector Set.

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.



Multiple cloning sites of the Baculovirus Transfer Vector Set.

- pAcGHLT-A, Component # 51-21460P (upper panel)*
- pAcGHLT-B, Component # 51-21461P (middle panel)*
- pAcGHLT-C, Component # 51-21462P (lower panel).*

References

Davies AH, Jowett JB, Jones IM. Recombinant baculovirus vectors expressing glutathione-S-transferase fusion proteins. *Biotechnology*. 1993; 11(8):933-936. (Biology)