

Technical Data Sheet

pAcSecG2T Baculovirus Transfer Vector

Product Information

Material Number: 554797
Size: 20 µg in 20 µl

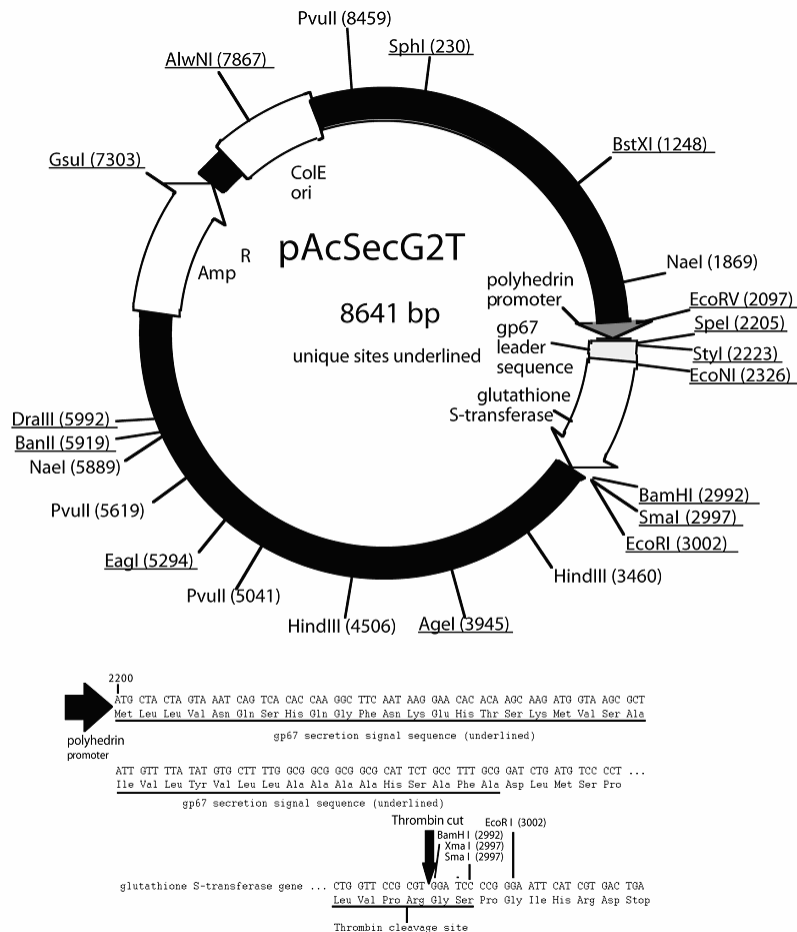
Description

The pAcSecG2T Baculovirus GST-fusion expression vector is a derivative of the pAcCL29 vector. Foreign genes may be inserted downstream of the glutathione S-transferase (GST) coding region into one of the available restriction enzyme sites (BamH I, Sma I or EcoR I). All foreign inserts **must be in frame with the GST open reading frame (ORF)**. The GST gene is preceded by an in-frame **gp67 signal sequence** to allow secretion of the GST-fusion protein into the supernatant. The signal sequence will be cleaved off during secretion while the GST tag will remain intact providing a fusion protein composed of GST and the foreign sequence. GST has a high affinity for reduced glutathione allowing single-step protein purification using glutathione agarose beads. After purification, the GST affinity tag can be removed by incubating the fusion protein in the presence of thrombin. This vector is a polyhedrin-locus derived vector and is recommended for use in conjunction with the BaculoGold™ DNA (Cat. No. 554739).

Preparation and Storage

Store undiluted at -20°C.

The plasmid DNA was purified by silica bead matrix and dissolved in TE buffer (10 mM Tris-HCl, pH 7.5; 1 mM EDTA).



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Application Notes

Application

Baculovirus	Routinely Tested
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Recommended Assay Procedure:

For expression 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 (DH5 α , HB101 or other suitable strain) under ampicillin selection. For generation of recombinant virus, perform a co-transfection of the pAcSecG2T vector and linearized Baculovirus DNA (BD BaculoGold™ viral DNA Cat. No. 554739) into a susceptible cell line (Sf9 or Sf21). The monoclonal antibody 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. For detailed procedures, refer to our online protocols or the Baculovirus Expression Vector System Manual, 6th edition on our web site at <http://www.bdbiosciences.com/pdfs/manuals/98-6088-1F.pdf>.

Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
554739	Linearized Baculovirus DNA	5 transfections	(none)
560129	Transfection Kit	5 transfections	(none)
560137	GST Purification Kit	1 box	(none)
554805	Purified Mouse Anti-Glutathione S-Transferase	0.1 mg	G172-1138

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.

References

- Davies AH, Jowett JB, Jones IM. Recombinant baculovirus vectors expressing glutathione-S-transferase fusion proteins. *Biotechnology*. 1993; 11(8):933-936. (Biology)
- Livingstone C, Jones I. Baculovirus expression vectors with single strand capability. *Nucleic Acids Res*. 1989; 17(6):2366. (Biology)