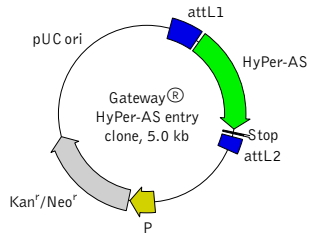


Gateway® HyPer-AS entry clone

The vector sequence has been compiled using the information from sequence databases, published literature, and other sources, together with partial sequences obtained by Evrogen. This vector has not been completely sequenced.



For vector sequence, please visit our Web site at <http://www.invitrogen.com/products/vectors.shtml>

Location of features

attL1 site: 14-113
 Kozak translation initiation site: 129-139
 HyPer-AS: 136-1572
 attL2 site: 1598-1697
 Kanamycin resistance gene: 2922-3716
 pUC origin of replication: 4301-4944

References

Kozak, M. (1987) "An analysis of 5'-noncoding sequences from 699 vertebrate messenger RNAs." *Nucleic Acids Res*, 15 (20): 8125-8148 / pmid: 3313277

Gateway® Technology. Ver. E. 22 Sept. 2003, 25-0522. <http://www.invitrogen.com/content/sfs/manuals/gatewayman.pdf> (visited on 18.06.2008)

Product	Cat.#	Size
Gateway® HyPer-AS entry clone	FP943	20 µg
Vector type	Gateway® entry clone	
Reporter	HyPer	
Reporter codon usage	Arabidopsis and Saccharomyces	
Promoter for HyPer	NO	
Host cells	prokaryotic	
Selection	kanamycin	
Replication	pUC ori	
Use	Transfer of HyPer-AS coding sequence into Gateway® destination vectors	

Vector description

Gateway® HyPer-AS entry clone is a vector containing HyPer gene variant with codon usage optimized for high expression in Arabidopsis and Saccharomyces. HyPer coding sequence is flanked by attL1 and attL2 sites allowing easy site-specific recombination. The Invitrogen Gateway® Technology provides a rapid and highly efficient way to transfer the HyPer gene into a number of Gateway® destination vectors for expression in different experimental systems.

To increase mRNA translation efficiency, Kozak consensus translation initiation site is generated upstream of HyPer coding sequence [Kozak 1987].

The vector backbone contains pUC origin of replication and kanamycin resistance gene (Kan^r) for propagation and selection in *E. coli*.

LR site-specific recombination

Please refer to Invitrogen Gateway® Technology description for detailed instructions regarding LR site-specific recombination reaction. In general, to transfer HyPer gene into the destination vector you will need:

- Purified plasmid DNA of Gateway® HyPer-AS
- A destination vector of choice
- Invitrogen LR Clonase™ II enzyme mix (Invitrogen Cat.# 11791-020)
- Proteinase K solution (supplied with the LR Clonase™ II enzyme mix)
- TE-Buffer, pH 8.0 (10 mM Tris-HCl, pH 8.0, 1 mM EDTA)
- Appropriate chemically competent *E. coli* host and growth media for expression
- Appropriate selective plates.

Propagation in *E. coli*

Suitable host strains for propagation in *E. coli* include DH5alpha, HB101, XL1-Blue, and other general purpose strains. Plasmid incompatibility group is pMB1/ColE1. The vector confers resistance to kanamycin (30 µg/ml) to *E. coli* hosts. Copy number in *E. coli* is about 500.

Notice to Purchaser:

The HyPer-related materials (also referred to as "Products") are intended for research use only.

Some elements of these materials may be covered by third party patents issued and applicable in certain countries. No license under these patents is conveyed expressly or by implication to the recipient of the materials. Users of these materials may be required to obtain a patent license depending upon the particular application and country in which the materials are received or used.

Invitrogen Gateway® Technology: please see Invitrogen Limited Use Label License N^o. 19: Gateway® Cloning Products.

MATERIAL SAFETY DATA SHEET INFORMATION: To the best of our knowledge, these products do not require a Material Safety Data Sheet. However, all the properties of these products (and, if applicable, each of their components) have not been thoroughly investigated. Therefore, we recommend that you use gloves and eye protection, and wear a laboratory coat when working with these products.