

# pFUSE2ss-CLIg-rk2

Plasmid featuring the constant region of the rabbit Ig kappa 2 light chain and the IL2 signal sequence

Catalog # pfuse2ss-rclk2

For research use only

Version 20J13-MM

## PRODUCT INFORMATION

### Content:

- 20 µg of pFUSE2ss-CLIg-rk2 plasmid provided as lyophilized DNA.

- 2 x 1 ml blasticidin at 10 mg/ml

### Storage and Stability:

- Product is shipped at room temperature.

- Lyophilized DNA should be stored at -20°C and is stable 3 months.

- Resuspended DNA should be stored at -20°C and is stable up to 1 year.

### Quality control:

- Plasmid construct has been confirmed by restriction analysis and sequencing.

- Plasmid DNA was purified by ion exchange chromatography.

## Materials required for antibody generation & isotype switching

- pFUSE2-CLIg plasmid that features the constant region of the kappa or lambda light chains. pFUSE2-CLIg plasmids are selectable with blasticidin.

- pFUSE-CHlg plasmid for the constant region of the heavy chain, this plasmid is selectable with Zeocin™ (sold separately, see RELATED PRODUCTS).

## GENERAL PRODUCT USE

pFUSE-CLIg and pFUSE-CHlg plasmids are designed to change a monoclonal antibody from one isotype to another, therefore, enabling the generation of antibodies with the same antigen affinity but with different effector functions (increased or reduced ADCC and CDC). Furthermore, they can be used to produce entire IgG antibodies from Fab or scFv fragments.

pFUSE-CHlg and pFUSE2-CLIg express the constant regions of the heavy (CH) and light (CL) chains, respectively. They contain a multiple cloning site (MCS) upstream of these constant regions to enable the cloning of the variable (VH and VL) regions of a given antibody. Transfection of mammalian cell lines with the recombinant pFUSE-CHlg and pFUSE2-CLIg pair allows to generate an IgG antibody that can be purified from the supernatant using the appropriate Protein A, Protein G or Protein L affinity chromatography.

pFUSE2ss plasmids contain the versatile hIL2 signal sequence for the generation of secretable immunoglobulin polypeptides, when the native Ig signal sequence is not available (such as VL derived from phage display library). Coexpression of the pFUSE2ss-CLIg (cloned with the VL region) with a pFUSEss-CHlg (cloned with the VH region) will allow for the expression of recombinant antibodies.

## Features of pFUSEss-CHlg and pFUSE2ss-CLIg plasmids

• **hEF1-HTLV prom** is a composite promoter comprising the Elongation Factor-1α (EF-1α) core promoter<sup>1</sup> and the R segment and part of the U5 sequence (R-U5') of the Human T-Cell Leukemia Virus (HTLV) Type 1 Long Terminal Repeat<sup>2</sup>. The EF-1α promoter exhibits a strong activity and yields long lasting expression of a transgene *in vivo*. The R-U5' has been coupled to the EF-1α core promoter to enhance stability of RNA.

• **MCS:** The multiple cloning site contains several restriction sites that are compatible with many other enzymes, thus facilitating cloning.

• **SV40 pAn:** The Simian Virus 40 late polyadenylation signal enables efficient cleavage and polyadenylation reactions resulting in high levels of steady-state mRNA<sup>3</sup>.

• **ori:** A minimal *E. coli* origin of replication to limit vector size, but with the same activity as the longer Ori.

• **CMV enh / hFerL prom:** This composite promoter combines the human cytomegalovirus immediate-early gene 1 enhancer and the core promoter of the human ferritin light chain gene. This ubiquitous promoter drives the expression of the blasticidin-resistance gene in mammalian cells.

• **IL2 ss:** The human IL2 signal sequence contains 20 amino acids (MYRMQLLSICIALSLALVTNS) and share common characteristics with signal peptides of other secretory proteins. The intracellular cleavage of the IL2 signal peptide occurs after Ser20 and leads to the secretion of the immunoglobulin chain.

• **EM2KC** is a bacterial promoter that enables the constitutive expression of the antibiotic resistance gene in *E. coli*. EM2KC is located within an intron and is spliced out in mammalian cells.

• **βGlo pAn:** The human beta-globin 3'UTR and polyadenylation sequence allows efficient arrest of the transgene transcription<sup>4</sup>.

## pFUSE2ss-CLIg-rk2 specific features

• **Rabbit IgKC2 (Ig Kappa 2 Light constant domain):** When cloning your VL of choice in the MCS, care must be taken to preserve the integrity of the kappa 2 light chain constant region and reading frame.

• **Bsr (blasticidin resistance gene):** Resistance to blasticidin is conferred by the bsr gene from *Bacillus cereus*. The same resistance gene confers selection in both mammalian cells and *E. coli*.

## References:

1. Kim DW, *et al.* 1990. Use of the human elongation factor 1 alpha promoter as a versatile and efficient expression system. *Mol Cell Biol.* 10(2):217-23. 2. Takebe Y, *et al.* 1988. SR alpha promoter: an efficient and versatile mammalian cDNA expression system composed of the simian virus 40 early promoter and the R-U5 segment of human T-cell leukemia virus type 1 long terminal repeat. *Mol Cell Biol.* 8(1):466-72. 3. Carswell S, & Alwine JC. 1989. Efficiency of utilization of the simian virus 40 late polyadenylation site: effects of upstream sequences. *Mol Cell Biol.* 9(10):4248-58. 4. Yu J, & Russell JE. 2001. Structural and functional analysis of an mRNP complex that mediates the high stability of human beta-globin mRNA. *Mol Cell Biol.* 21(17):5879-88.

## TECHNICAL SUPPORT

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## PROTOCOL

### Obtaining VH and VL sequences

To obtain the cDNA sequence of the VH and VL regions from an antibody producing hybridoma, total RNA or mRNA is extracted and reverse transcribed to cDNA. PCR is performed with 5' degenerate primers to anneal to the unknown VH and VL regions and the 3' primers designed to anneal to the "known" CH and CL regions. Alternatively 5' RACE can be used. The resulting amplicons must be sequenced.

### Plasmid resuspension

Quickly spin the tube containing the lyophilized plasmid to pellet the DNA. To obtain a plasmid solution at 1 µg/µl, resuspend the DNA in 20 µl of sterile H<sub>2</sub>O. Store resuspended plasmid at -20°C.

### Cloning into pFUSEss-CHIg and pFUSE2ss-CLIg

Once the VH and VL sequence are known, inserts for cloning into the plasmids can be generated. In pFUSE2ss-CLIg-rk2, the constant region of the rabbit kappa 1 light chain is preceded by a multiple cloning site containing six restriction sites: EcoRI, EcoRV, XhoI, NcoI, Acc65I and BamHI. Using EcoRI as the 5' cloning site ensures that the cloned VL will follow the hIL2 signal sequence without unwanted additional amino-acids. In pFUSE2ss-CLIg-rk2, use BamHI as the 3' cloning site for the VL in order to preserve the immunoglobulin kappa constant amino acid sequence.

### Notes:

- Using NcoI as the 3' cloning site will introduce amino acid changes that may not be suitable for some purposes.
- When generating the insert for VH, a XhoI restriction site must be introduced at the 3' end (for pFUSE-CHIg plasmid featuring the rabbit IgG (allele 3/2) heavy chain; see RELATED PRODUCTS). Using EcoRI as the 5' cloning site ensures that the cloned VH will follow the hIL2 signal sequence without unwanted additional amino-acids.

### Choice of strategies for the transfection

Cotransfect mammalian cells, such as 293 and CHO cells, with the recombinant plasmids pFUSE2ss-CLIg encoding the light chain and pFUSEss-CHIg encoding the heavy chain. Antibody production depends greatly on the ratio of heavy chain and light chain expression. Typically, pFUSEss-CHIg to pFUSE2ss-CLIg ratio of 2:3 is used to cotransfect mammalian cells. Since both plasmids share the same plasmid backbone, the appropriate heavy chain to light chain ratio can be easily determined by varying the quantities of plasmids.

OR

Transfect cells using a transfection agent, such as LyoVec™, with the plasmid coding for light chain and select the best clone. Following selection of the best clone, the plasmid coding for the heavy chain clone can be transfected into this clone.

Use blasticidin and Zeocin™ to select pFUSE2-CLIg and pFUSE-CHIg respectively.

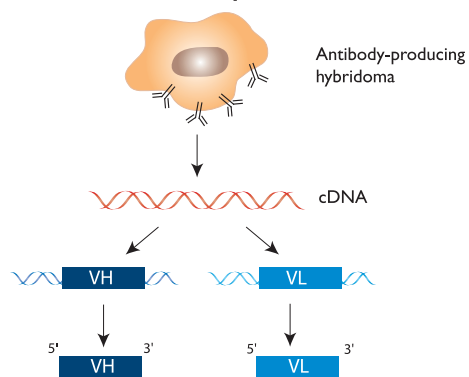
Antibody production can be analyzed by different techniques including SDS-PAGE, flow cytometry, ELISA, or a bioactivity assay.

### Antibody purification

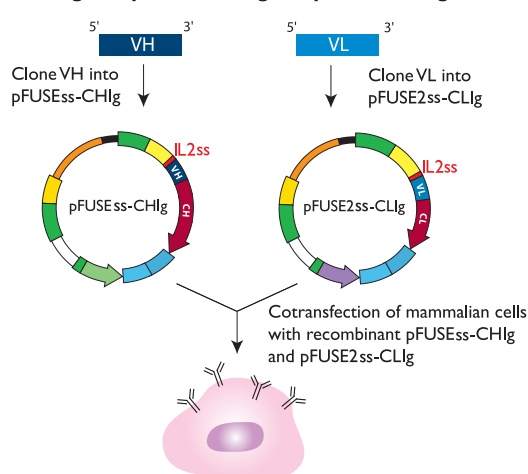
The resulting IgG antibody can be purified from the supernatant using the appropriate Protein A, Protein G or Protein L affinity chromatography.

## Antibody generation using pFUSE-CHIg & pFUSE2-CLIg

### 1- Obtention of VH and VL sequences



### 2- Cloning into pFUSEss-CHIg and pFUSE2ss-CLIg

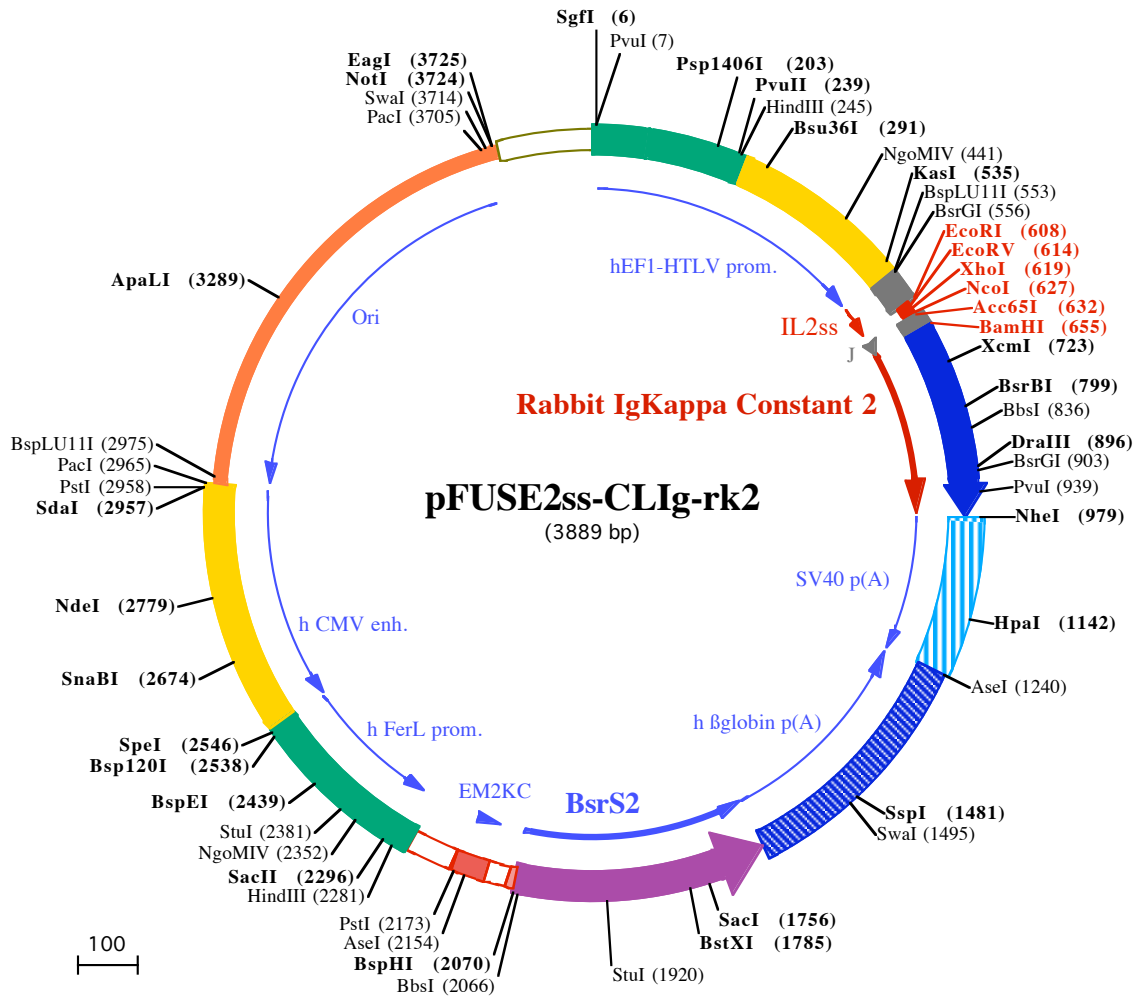


## RELATED PRODUCTS

Product	Catalog Code
pFUSEss-CHIg-rG	pfussess-rchg
LyoVec™	lyec-12
Protein L / Agarose	gel-protl-2
Protein G / Agarose	gel-agg-5
Blasticidin	ant-bl-1

### TECHNICAL SUPPORT

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PvuI (7)  
SgfI (6)  
1 GGATCTGCGATCGCTCCGGTGCCCGTCAGTGGCAGAGCGCACATCGCCACAGTCCCCGAGAAGTTGGGGGAGGGGTCGGCAATTGAACGGGTGCCTA  
101 GAGAAGGTGGCGCGGGTAAACTGGAAAGTGATGTCGTGTAAGTGGCTCCGCCTTTTCCGAGGGTGGGGGAGAACCCTATATAAGTGCAGTAGTCGCC

HindIII (245)  
Psp1406I (203) PvuII (239) Bsu36I (291)  
201 GTGAACGTTCTTTTTTCGCAACGGGTTTGCCGCCAGAACACAGCTGAAGCTTCGAGGGGCTCGCATCTCTCTTACAGCGCCCGCCGCCCTACCTGAGGCC  
301 GCCATCCACGCGGTTGAGTGCCTGTTGCGCCTCCCGCCTGTGGTGCCTCTGAAGTGCCTCCGCGTCTAGGTAAGTTTAAAGCTCAGGTCGAGACC

NgoMIV (441)  
401 GGGCCTTTGTCCGGCGCTCCCTTGAGCCTACCTAGACTCAGCCGGCTCTCCACGCTTGGCTGACCCTGCTTGTCTCAACTCTACGCTTTTGTTCGTTT

KasI (535) BsrGI (556)  
501 TCTGTTCTGCGCCGTTACAGATCCAAGCTGTGACCGCGCCCTACCTGAGATCAACATGTACAGGATGCAACTCCTGTCTTGCATTGCACTAAGTCTTGCA  
1 M Y R M Q L L S C I A L S L A

EcoRV (614) NcoI (627)  
601 CTTGTACGAAATTCGATATCTCGAGCACCATGGGTACCAAGGTGGAAATCAAACGGGATCCAGTTGCGCCTTCTGTCTCTCTTCCACCATCTAAGGA  
16 L V T N S 1 G T K V E I K 1 D P V A P S V L L F P P S K E

EcoRI (608) XhoI (619) Acc65I (632) BamHI (655)  
701 GGAGCTGACAACCTGGAACAGCCACCATCGTGTGCGTGGCGAATAAATCTATCCAGTGACATCACCGTACCTGGAAGTGGATGGCACCACCAACAG  
15 E L T T G T A T I V C V A N K F Y P S D I T V T W K V D G T T Q Q

XcmI (723) BsrBI (799)  
801 AGCGGCATCGAGAACAGTAAAACACCGCAGAGCCCCGAAGACAATACCTACAGCCTGAGCAGCACTCTGTCACTGACCAGCGCACAGTACAACAGCCACA  
49 S G I E N S K T P Q S P E D N T Y S L S S T L S L T S A Q Y N S H

BbsI (836) DraIII (896)  
901 GCGTGTACACCTGCGAGGTGGTCCAAGGCTCAGCCTCACCGATCGTCCAGAGCTTCAATAGGGGTGACTGCTAGAGCGAGCTAGCTCGACATGATAAGAT  
82 S V Y T C E V V Q G S A S P I V Q S F N R G D C •

BsrGI (903) PvuI (939) NheI (979)  
1001 ACATTGATGAGTTTGACAAACCACAACATAAGATGCAAGTGAAGAAATGCTTTATTTGTGAAATTTGTGATGCTATTGCTTTATTTGTGAAATTT

HpaI (1142)  
1096 GTGATGCTATTGCTTTATTTGTAACCATTATAAGTGAATAAACAAGTTAACAACAACAATTGCATTATTATGTTTCAGGTTTCAGGGGGAGGTGTG

AseI (1240)  
1196 GGAGGTTTTTAAAGCAAGTAAACCTCTACAAATGTGGTATGGAATTAATCTAAAATACAGCATAGCAAACTTTAACCTCAAATCAAGCCTCTAC  
1295 TTGAATCCTTTCTGAGGGATGAATAAGGCATAGGCATCAGGGGCTGTGCCAATGTGCATTAGCTGTTTGCAGCCTCACCTCTTTTCATGGAGTTAAG

SspI (1481)  
1395 ATATAGTGTATTTTCCCAAGGTTTGAACAGTCTTTCATTTCTTTATGTTTTAAATGCACTGACCTCCACATTCCTTTTATGTAATAATTCAGAAAT

SwaI (1495)  
1495 AATTTAAATACATCATTGCAATGAAAATAAATGTTTTTATTAGGCAGAAATCCAGATGCTCAAGGCCCTCATAATATCCCCAGTTTAGTAGTTGGACT  
1595 TAGGGAACAAAGAACCTTTAATAGAAATTGGACAGCAAGAAAGCGAGCTTCTAGCTTTAGTTCTGTTGACTTGGAGGGGATGAGTTCCTCAATGGTG  
141 • N R T Y K L P I L E E I T

SacI (1756) BstXI (1785)  
1695 GTTTTGACCAGCTTGCATTCTCAATGAGCACAAAGCAGTCAGGAGCATAGTCAGAGATGAGCTCTGCACATGCCACAGGGGCTGACCACCTGA  
126 T K V L K G N M E I L V F C D P A Y D S I L E R C M G C P S V V R I  
1795 TGGATCTGTCCACCTCATCAGAGTAGGGGTGCCTGACAGCCACAATGGTGTCAAAGTCTTCTGCCCGTTGCTCACAGCAGACCAATGGCAATGGCTTC  
93 S R D V E D S Y P H R V A V I T D F D K Q G N S V A S G I A I A E

StuI (1920)  
1895 AGCACAGACAGTGACCCTGCAATGTAGGCCTCAATGTGGACAGCAGAGATGATCTCCCAGTCTTGGTCTGATGGCCGCCGACATGGTCTTGTG  
60 A C V T V R G I Y A E I H V A S I I E G T K T R I A A G V H H K N

BspHI (2070)  
1995 TCCTCATAGAGCATGGTGTCTTCTCAGTGGCGACCTCCACCAGCTCCAGATCCTGCTGAGAGATGTTGAAGTCTTTCATGATGGCTCCTCctgtcagg  
26 D E Y L M T I K E T A V E V L E L D Q Q S I N F T K M

BbsI (2066)  
2094 agaggaagagaagaaggttagtacaattgCTATAGTGAGTTGTAATTACTATGCTTATGATTAATTGTCAAAGTGGCTGCAgggttcagatgtcc  
AseI (2154) PstI (2173)

HindIII (2281)  
2193 acttttctgcaactgccccatctctgcccacccttccaggcatagacagtcagtgacttacCAAACCTCACAGGAGGAGAAGCGAAGCTTGAGAC  
2293 AGACCCGCGGGACCGCGAACTGCGAGGGGACGTGGCTAGGGCGGCTTTTTATGGTGCGCCGGCCCTCGGAGGCAGGGCGCTCGGGGAGGCCTAGCGG

SacII (2296) NgoMIV (2352) StuI (2381)  
2393 CCAATCTGCGGTGGCAGGAGCGGGGCCGAAGCCGTGCTGACCAATCCGGAGCACATAGGAGTCTCAGCCCCCGCCCAAAGCAAGGGGAAGTCACG

BspEI (2439)

2493 **SpeI (2546)**  
**Bsp120I (2538)** CGCCTGTAGCGCCAGCGTGTGTGAAATGGGGCTTGGGGGGTGGGGCCCTGACTAGTCAAAACAAACTCCCATTGACGTCAATGGGGTGGAGACTTG

2593 **SnaBI (2674)**  
 GAAATCCCCGTGAGTCAAACCGCTATCCACGCCATTGATGTACTGCCAAAACCGCATCATCATGGTAATAGCGATGACTAATACGTAGATGTACTGCCA

2693 **NdeI (2779)**  
 AGTAGAAAGTCCATAAGGTCATGTACTGGGCATAATGCCAGGCGGGCCATTTACCGTCATTGACGTCAATAGGGGGCGTACTTGGCATATGATACACT

2793 TGATGTACTGCCAAGTGGGCAGTTTACCCTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACATACGTCATTATTG

2893 **PstI (2958)** **PacI (2965)**  
**SdaI (2957)** **BspLU11I (2975)**  
 ACGTCAATGGGGGGGGTTCGTTGGGCGGTGAGCCAGGCGGGCCATTTACCGTAAGTTATGTAACGCCTGCAGGTTAATTAAGAACATGTGAGCAAAGGC

2993 CAGCAAAGGCCAGGAACCGTAAAAAGCCGCTTGGTGGCGTTTTCCATAGGCTCCGCCCCCTGACGAGCATCACAAAAATCGACGCTCAAGTCAGA

3093 GGTGGCGAAACCCGACAGGACTATAAAGATACCAGCGTTTTCCCCTGGAAAGCTCCCTCGTGCCTCTCCTGTCCGACCCTGCCGTTACCGGATACCT

3193 **ApaLI (3289)**  
 GTCCGCCTTCTCCCTTCGGAAGCGTGGCGCTTCTCATAGCTCAGCTGTAGGTATCTCAGTTCGGTGTAGGTCGTTCCGCTCCAAGCTGGGCTGTGTG

3293 CACGAACCCCCGTTCCAGCCGACCGCTGCGCCTTATCCGGTAACTATCGTCTTGAGTCCAACCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCA

3393 CTGGTAACAGGATTAGCAGAGCGAGGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTACGGCTACACTAGAAGAACAGTATTTGGTAT

3493 CTGCGCTCTGCTGAAGCCAGTTACCTTCGAAAAAGAGTTGGTAGCTCTTGATCCGGCAAACAACACCGCTGGTAGCGGTGTTTTTTTGTGTTGCAAG

3593 CAGCAGATTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTCTACGGGTCTGACGCTCAGTGAACGAAAACACTCACGTTAAGGGATT

3693 **EagI (3725)**  
**PacI (3705)** **Swal (3714)** **NotI (3724)**  
 TGGTCATGGCTAGTTAATTAACATTTAAATCAGCGGCCGCAATAAAATATCTTTATTTTCATTACATCTGTGTGTTGGTTTTTTGTGTGAATCGTAACTA

3793 ACATACGCTCTCCATCAAACAAACGAAACAAACAAACTAGCAAATAGGCTGTCCCCAGTGAAGTGCAGGTGCCAGAACATTTCTCTATCGAA

# Blasticidin

Selection antibiotic; cell culture tested

Catalog code: ant-bl-05, ant-bl-1, ant-bl-5, ant-bl-5b

<http://www.invivogen.com/blastcidin>

For research use only

Version 20J13-MM

## PRODUCT INFORMATION

### Contents

Blasticidin hydrochloride is supplied as a sterile filtered solution at 10 mg/ml in HEPES buffer. It is available in 4 pack sizes:

- ant-bl-05: 5 x 1 ml (50 mg)
- ant-bl-1: 10 x 1 ml (100 mg)
- ant-bl-5: 50 x 1 ml (500 mg)
- ant-bl-5b: 1 x 50 ml (500 mg)

### Storage and stability

- Blasticidin is shipped at room temperature. Upon receipt it should be stored at 4 °C or at -20 °C. Avoid repeated freeze-thaw cycles.  
- The expiry date is specified on the product label.

**Note:** Blasticidin is stable for 2 weeks at room temperature.

## QUALITY CONTROL

Each lot is thoroughly tested to ensure the absence of lot-to-lot variation.

- Purity: ≥95% (HPLC)
- Endotoxin level: < 1 EU/mg
- Physicochemical characterization (pH, appearance)
- Cell culture tested: potency validated in blasticidin-sensitive and blasticidin-resistant mammalian cell lines
- Non-cytotoxicity of trace contaminants: absence of long-term effects confirmed in blasticidin-resistant cells

## BACKGROUND

Blasticidin is a selection antibiotic that acts on both eukaryotic and prokaryotic cells. It is a peptidyl nucleoside antibiotic isolated from the culture broth of *Streptomyces griseochromogenes*. It specifically inhibits protein synthesis in both prokaryotes and eukaryotes by inhibiting peptide bond formation in the ribosomal machinery. Three blasticidin resistance genes have been cloned and sequenced: an acetyl transferase gene, *bls* from a blasticidin producer strain<sup>1</sup>, and two deaminase genes, *bsr* gene from *Bacillus cereus*<sup>2</sup>, and *BSD* gene from *Aspergillus terreus*<sup>3</sup>.

Both *bsr* and *BSD* genes are used as dominant selectable markers for gene transfer experiments in mammalian and plant cells. Although blasticidin was developed as a selection agent for mammalian cells, it can also be used in *E. coli*.

## GENERAL GUIDELINES

Successful transfection is influenced by many factors. The health and viability of the cell line, the quality of the nucleic acid used, the transfection reagent, the duration of transfection, and the presence or absence of serum can all play a part.

## SAFETY CONSIDERATIONS

Blasticidin is a harmful compound. Refer to safety data sheet for handling instructions.

## SELECTION CONDITIONS

### - *Escherichia coli*

*E. coli* is poorly sensitive to blasticidin, but transformants resistant to blasticidin can be selected on low salt LB agar medium (pH 8) supplemented with 100 µg/ml blasticidin. High pH enhances the activity of blasticidin.

### - Mammalian cells

The working concentration of blasticidin for mammalian cell lines varies from 1 to 10 µg/ml, in a few cases up to 30 µg/ml. In a starting experiment we recommend to determine optimal concentrations of antibiotic required to kill your host cell line. After treatment, cell death occurs rapidly, allowing the selection of transfected cells with plasmids carrying the *bsr* or *BSD* genes in as little as 7 days post-transfection. Suggested concentrations of blasticidin for selection in some examples of mammalian cells are listed below.

Cell line	Medium	Blasticidin conc.	Ref.
CHO (Chinese hamster ovarian cells)	DMEM	5-10 µg/ml	4, 5
HEK293 (Human embryonic kidney cells)	DMEM	5-15 µg/ml	6, 7
HeLa (Human uterine cells)	DMEM	2.5-10 µg/ml	8, 9
Neuro2a (Mouse neuroblasts)	DMEM	30 µg/ml	10
THP-1 (Human monocytes)	RMPI	10 µg/ml	11

1. Perez-Gonzalez J. *et al.*, 1990. Cloning and characterization of the gene encoding a blasticidin S acetyltransferase from *Streptovorticillum* sp. *Gene*. 86:129-34.
2. Izumi M. *et al.*, 1991. Blasticidin S-resistance gene (*bsr*): A novel selectable marker for mammalian cells. *Exp.Cell Res.*197:229-33.
3. Kimura M. *et al.*, 1994. Blasticidin S deaminase gene from *Aspergillus terreus* (*BSD*): a new drug resistance gene for transfection of mammalian cells. *Biochim. Biophys. Acta.* 1219:653-9.
4. Dorgham K. *et al.*, 2009. An engineered CX3CR1 antagonist endowed with anti-inflammatory activity. *J Leukoc Biol.* 86(4):903-11.
5. LeBon L. *et al.*, 2014. Fringe proteins modulate Notch-ligand cis and trans interactions to specify signalling states. *eLife Sci.* 3:e02950.
6. Tomecki R. *et al.*, 2014. Multiple myeloma-associated hDIS3 mutations cause perturbations in cellular RNA metabolism and suggest hDIS3 PIN domain as a potential drug target. *Nucleic Acids Res.* 42:1270-90.
7. Edbauer D. *et al.*, 2004. Co-expression of nicastrin and presenilin rescues a loss of function mutant of APH-1. *J Biol Chem.* 279:37311-5.
8. Khandelia P. *et al.*, 2011. Streamlined platform for short hairpin RNA interference and transgenesis in cultured mammalian cells. *PNAS* 108:12799-804.
9. Lee HK. *et al.*, 2007. Application of beta-lactamase enzyme complementation to the high-throughput screening of toll-like receptor signaling inhibitors. *Mol Pharmacol.* 72:868-75.
10. Matsumoto G. *et al.*, 2011. Serine 403 phosphorylation of p62/SQSTM1 regulates selective autophagic clearance of ubiquitinated proteins. *Mol Cell.* 44:279-89.
11. Schepetkin IA. *et al.*, 2009. Immunomodulatory activity of oenothin B isolated from *Epilobium angustifolium*. *J Immunol.* 183:6754-66.

## TECHNICAL SUPPORT

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## RELATED PRODUCTS

Product	Description	Catalog Code
<b>Other selection antibiotics</b>		
G418	Selection antibiotic for the <i>neo</i> gene	ant-gn-1
Hygromycin B Gold	Selection antibiotic for the <i>hph</i> gene	ant-hg-1
Puromycin	Selection antibiotic for the <i>pac</i> gene	ant-pr-1
Zeocin™	Selection antibiotic for the <i>Sh ble</i> gene	ant-zn-1
<b>Plasmids encoding the <i>bsr</i> gene</b>		
pMOD2-Blast	Plasmid encoding a synthetic <i>bsr</i> gene	pmod2-blast
pSELECT-blasti-LacZ	LacZ-expression plasmid selectable with blasticidin	psetb-lacz
pSELECT-blasti-mcs	Expression plasmid selectable with blasticidin	psetb-mcs
pUNO1-bsr	Expression plasmid selectable with blasticidin	puno1-bsr

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