Blasticidin

Selection antibiotic; cell culture tested

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

http://www.invivogen.com/blasticidin

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[1], and two deaminase genes, *bsr* gene from *Bacillus cereus*[2], and *BSD* gene from *Aspergillus terreus*[3].

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-Gonalez J. *et al.*, 1990. Cloning and characterization of the gene encoding a blasticidin S acetyltransferase from *Streptoverticillum 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 signaling 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 oenothein B isolated from Epilobium angustifolium. J Immunol. 183:6754-66.

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RELATED PRODUCTS

Product	Description	Catalog Code
Other selection antibiotics		
G418	Selection antibiotic for the *neo* gene	ant-gn-1
Hygromycin B Gold	Selection antibiotic for the *hph* gene	ant-hg-1
Puromycin	Selection antibiotic for the *pac* gene	ant-pr-1
Zeocin™	Selection antibiotic for the *Sh ble* gene	ant-zn-1
Plasmids encoding the *bsr* gene		
pMOD2-Blast	Plasmid encoding a synthetic *bsr* 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