Puromycin

Selection antibiotic; cell culture tested
Catalog code: ant-pr-1, ant-pr-5, ant-pr-5b
http://www.invivogen.com/puromycin

For research use only
Version 20C31-MM

PRODUCT INFORMATION

Contents:
Puromycin dihydrochloride is supplied as a sterile filtered solution at 10 mg/ml in HEPES buffer. This product is available in 3 pack sizes:

- **ant-pr-1:** 10 x 1 ml (100 mg)
- **ant-pr-5:** 50 x 1 ml (500 mg)
- **ant-pr-5b:** 1 x 50 ml (500 mg)

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

Notes:
- *Puromycin is stable for 3 months at room temperature.*
- *During storage a crystalline precipitate of puromycin may form. If this occurs, heat the product at 37 °C for 30 minutes until the crystalline precipitate disappears. The formation of a crystalline precipitate does not affect the activity of the product.*

QUALITY CONTROL

Each lot is thoroughly tested to ensure the absence of lot-to-lot variation.
- Purity: ≥ 98% (HPLC)
- Endotoxin level: < 5 EU/mg
- Physicochemical characterization (pH, appearance)
- Cell culture tested: potency validated in puromycin-sensitive and puromycin-resistant mammalian cell lines
- Non-cytotoxicity of trace contaminants: absence of long-term effects confirmed in puromycin-resistant cells

DESCRIPTION

Puromycin is a selection antibiotic that acts on both eukaryotic and prokaryotic cells. It is an aminonucleoside antibiotic produced by *Streptomyces alboniger*. It specifically inhibits peptidyl transfer on prokaryotic and eukaryotic ribosomes. This antibiotic inhibits the growth of Gram positive bacteria and many animal and insect cells. Puromycin is weakly active against Gram-negative microorganisms.
Resistance to puromycin is conferred by the *pac* gene, which encodes a Puromycin N-acetyl-tranferase (PAC) that has been isolated from a Streptomyces producer strain[1].

SAFETY CONSIDERATIONS

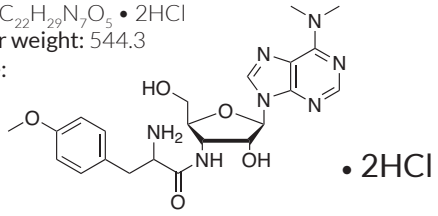
Puromycin is a harmful compound. Refer to safety data sheet for handling instructions.

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.

CHEMICAL PROPERTIES

CAS number: 58-58.2
Formula: $C_{22}H_{29}N_7O_5 \bullet 2HCl$
Molecular weight: 544.3
Structure:



SELECTION CONDITIONS

Escherichia coli
Puromycin is poorly active on *E. coli*. However, puromycin-resistant transformants can be selected in LB agar medium supplemented with 100-125 µg/ml of puromycin.

Mammalian cells
The working concentrations of puromycin for mammalian cell lines range from 1 to 10 µg/ml. In a starting experiment we recommend to determine optimal concentrations of antibiotic required to kill your host cell line. Puromycin quickly kills eukaryotic cells that do not contain the *pac* gene. Dying cells detach from the plates, allowing easy and early identification of transformant clones. Suggested working conditions for selection in some mammalian cells are listed below.

Cell line	Medium	Puromycin conc.	Ref.
B16 (Mouse melanocytes)	RPMI	1-2 µg/ml	2, 3
HEK293 (Human kidney cells)	DMEM	0.5-10 µg/ml	4, 5, 6
HeLa (Human uterine cells)	DMEM	1-10 µg/ml	6, 7
MEF (Mouse fibroblasts)	DMEM	1-5 µg/ml	6

1. Lacalle R. *et al.*, 1989. Molecular analysis of the pac gene encoding a puromycin N-acetyl transferase from Streptomyces alboniger. Gene. 79:375-80. **2. Furge KA. *et al.*, 2001.** Suppression of Ras-mediated tumorigenicity and metastasis through inhibition of the Met receptor tyrosine kinase. PNAS 98:10722-7. **3. Díaz J. *et al.*, 2014.** Rab5 is required in metastatic cancer cells for Caveolin-1-enhanced Rac1 activation, migration and invasion.. J Cell Sci. 127:2401-6. **4. Rössger K. *et al.*, 2013.** Reward-based hypertension control by a synthetic brain-dopamine interface. PNAS, 110:18150-5. **5. Schmitter D. *et al.*, 2006.** Effects of Dicer and Argonaute down-regulation on mRNA levels in human HEK293 cells. Nucleic Acids Res. 34:4801-15. **6. Kamer I. *et al.*, 2005.** Proapoptotic BID Is an ATM effector in the DNA-damage response Cell. 122:593-603. **7. Charnaux N. *et al.*, 2005.** RANTES (CCL5) induces a CCR5-dependent accelerated shedding of syndecan-1 (CD138) and syndecan-4 from HeLa cells and forms complexes with the shed ectodomains of these proteoglycans as well as with those of CD44. Glycobiology. 15:119-30.

TECHNICAL SUPPORT
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RELATED PRODUCTS

Product	Description	Catalog Code
Other selection antibiotics		
Blasticidin	Selection antibiotic for the *bsr* or BSD genes	ant-bl-1
G418	Selection antibiotic for the *neo* gene	ant-gn-1
Hygromycin B Gold	Selection antibiotic for the *hph* gene	ant-hg-1
Zeocin™	Selection antibiotic for the *Sh ble* gene	ant-zn-1
Plasmids encoding the *pac* gene		
pMOD2-Puro	Plasmid encoding a synthetic *pac* gene	pmod2-puro
pSELECT-puro-LacZ	LacZ-expression plasmid selectable with puromycin	psetp-lacz
pSELECT-puro-mcs	Expression plasmid selectable with puromycin	psetp-mcs
pUNO1-pac	Expression plasmid encoding the *pac* gene	puno1-pac