Puromycin, dihydrochloride

MG9568

Technical literature is available at: www.mesgenbio.com. E-mail MesGen Technical Services if you have questions on use of this system: tech@mesgenbio.com

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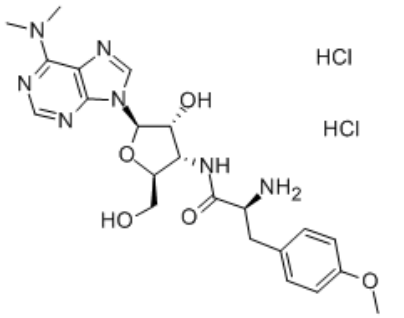
CAS : 58-58-2

Molecular weight : 544.43

Molecular formula : C22H29N7O5•2HCl

Melting point : 178-180°C

Synonyms : Stylomycin hydrochloride



Description

Puromycin is an aminonuclease antibiotic used for selection and maintenance of cell lines expressing a transfectedpac gene, whose product, puromycin-N-acetyl-transferase, inactivates puromycin via acetylation. This product has been cell culture tested and is recommended for selection of stably transfected cells following transfection with Santa Cruz Biotechnology shRNA or Lentiviral particles. BACKGROUND: Puromycin is an antibiotic substance produced by the soil actinomycete Streptomyces alboniger which induces apoptosis in cells by interfering with RNA function, leading to inhibition of protein synthesis. Toxic to both eukaryotic and prokaryotic cells. Believed to act as an acyl-tRNA analogue causing premature chain termination, it has been shown to arrest cells in G2/M phase.

Usage & method

For the selection of stably transfected cells following transfection with shRNA or Lentiviral particles, proceed with puromycin selection as follows :

• 48 hours post-transfection, aspirate the medium and replace with fresh medium containing puromycin at the appropriate concentration.

• Approximately every 2-3 days, aspirate and replace with freshly prepared selective media.

• Monitor the cells daily. Puromycin selection requires a minimum of 48 hours. Optimum effectiveness should be reached within 3-10 days.

• Assay transfected cells.

Titrating puromycin for selecting transfected cell lines :

• Plate 2 x 10^5 cells in each well of a 6-well plate containing 3 ml of the appropriate complete medium plus increasing concentrations of puromycin (i.e., 0, 1.0, 2.5, 5.0, 7.5, and 10.0 µg/ml)

• Replace with fresh selective medium after 2 days to remove dead cells.

• Examine the wells for viable cells every two days.

• Monitor the cells daily and observe the percentage of surviving cells. Optimum effectiveness should be reached in 1-4 days.

•The minimum antibiotic concentration to use is the lowest concentration that kills 100% of the cells in 3-5 days from the start of puromycin selection.

Solubility : Soluble in water (50 mg/ml), ethanol (~1 mg/ml), DMSO (~13 mg/ml), DMF (~14 mg/ml), and PBS, pH 7.2 (~10 mg/ml).

Storage condition

-20°C

For Research Use Only. Not For Use In Diagnostic Procedures.

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