Hygromycin B

Introduction

Hygromycin B, an aminoglycoside antibiotic derived from *Streptomyces hygroscopicus*, exerts its cytotoxic effects by disrupting protein synthesis in bacteria, fungi, and eukaryotic cells. Due to its broad-spectrum activity, it is widely employed for selecting both prokaryotic and eukaryotic transformed with hygromycin resistance gene.

Hygromycin B kills cells by binding to ribosomes and blocking protein synthesis. Resistance is conferred by the hygR gene, which produces an enzyme that inactivates the antibiotic through phosphorylation. To establish optimal working conditions, it is needed to perform a kill curve to identify the minimum concentration required to eliminate non-resistant cells.

Storage

Store desiccated at -20°C, stable for 2 years.

Protocol

1. **Preparation of stock solution**: Dissolve 50 mg Hygromycin B in 1 mL of sterile H_2O (or other suitable buffer) to make a 50 mg/mL stock solution. Then sterilize the stock solution with a 0.22 μm syringe filter. Aliquot and store the sterilized stock solution at -20°C, stable for 1 year.
2. **Recommended working concentration**: In a new experiment, it is suggested to obtain optimal concentrations by a kill curve. Recommended working concentrations for selection are listed below.
    1) For mammalian cell selection: 200 μg/mL.
    2) For plant cell selection: 20-200 μg/mL.
    3) For fungi selection: 200-1000 μg/mL.
    4) For bacteria selection: 20-200 μg/ml.

***Note:** The optimal concentration depends on the cell type.

Note

1. For your safety and health, please wear lab coats and gloves during the experiment.

2. For research use only. Not to be used in clinical diagnostic or clinical trials.

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