

# Hygromycin B, Sterile

## 1 Contents

Components	HY-K1051-10 mL	HY-K1051-50 mL
Hygromycin B, Sterile (50 mg/mL)	10 mL	50 mL

## 2 Introduction

Hygromycin B is an aminoglycosidic antibiotic purified from *Streptomyces hygroscopicus*. It acts by binding to the 70S subunit of the bacterial ribosome and inhibiting protein synthesis, leading to the death of bacteria, fungi and mammalian cells.

Resistance to Hygromycin B is conferred by the *E.coli* hygromycin resistance gene (hyg or hph). Therefore, Hygromycin B can be used to select and maintain prokaryotic and eukaryotic cells transfected with the hygromycin resistance gene. Low purity hygromycin can cause different toxic effects, so it is crucial to grasp its purity in the experiment.

## 3 General Protocol

1. The working concentration of Hygromycin B varies with cell type, media, growth conditions and cell metabolic rate.
2. Before stable transfected cell lines can be selected, the optimal Hygromycin B concentration needs to be determined by performing a kill curve titration.
  - 1) Seed the parental cell line in a suitable culture plate at a cell density of 20-25% and incubate for 24 hours at 37°C.
  - 2) Remove medium and then add medium with various concentrations of Hygromycin B (such as 50, 100, 250, 500, 750, and 1000 µg/mL) and incubate at 37°C.

- 3) Refresh the selective medium every 2-3 days and observe the percentage of surviving cells over time.
- 4) Determine the lowest concentration of antibiotic that kills a large majority of the cells within 7-10 days. This concentration should be used for selection of a stable transfected cell line.

## 4 Storage

Store at 4°C for 2 years

## 5 Precautions

1. Pay attention to aseptic operation to avoid contamination.
2. The working concentration of this product is set for the serum-containing basal medium. When used in a serum-free medium, the dosage should be appropriately reduced to avoid cytotoxicity.
3. This product is harmful to the human body. Please be careful when handling it, and avoid direct contact with the human body or inhalation of the body.
4. This product is for R&D use only, not for drug, household, or other uses.
5. For your safety and health, please wear a lab coat and disposable gloves to operate.