

G418 Selective Antibiotic, Sterile

1 Contents

Components	HY-K1056-10 mL	HY-K1056-50 mL
G418 Selective Antibiotic, Sterile (50 mg/mL)	10 mL	50 mL

2 Introduction

G418 selective antibiotic is an aminoglycoside antibiotic produced by *Micromonospora rhodorangea*. It acts by binding to the 80S subunit of the bacterial ribosome, thus inhibiting protein synthesis in both prokaryotic and eukaryotic cells.

Resistance to G418 selective antibiotic is conferred by the *E. coli* APH (3) I and APH (3) II resistance genes. The gene becomes APH (3) II on Tn5 and APH (3) I on Tn601. Therefore, G-418 selective antibiotic can be used to select and maintain prokaryotic or eukaryotic cells carrying the G418 resistance gene.

3 General Protocol

1. The working concentration of G418 selective antibiotic varies with cell type, media, growth conditions and cell metabolic rate.

Cell	Transposon	Gene	Concentration (mg/L)	Remark
Slime mold	Tn5	APH (3) II	10	Cells grow in culture
			35	Cells attached to freeze-dried bacteria
Mammalian	Tn5	APH (3) II	400	Used to filter
			200	Used to maintain
Plant	Tn5	APH (3) II	10	Used to filter
Yeast	Tn601	APH (3) I	500	Used to filter
			125	Used to maintain

2. Before stable transfected cell lines can be selected, the optimal G418 selective antibiotic 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 the cells for 24 hours at 37°C.

2) Remove medium and then add medium with various concentrations of G418 selective antibiotic (such as 0, 50, 100, 200, 400, 800, 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

-20°C 2 years

5 Precautions

1. Minimize repeated freeze-thaw cycles.
2. Pay attention to aseptic operation to avoid contamination.
3. 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.
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.