CALBIOCHEM®

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HYGROMYCIN B

Hygromycin B, an aminoglycoside antibiotic, inhibits the growth of prokaryotic and eukaryotic microorganisms and mammalian cells. It inhibits protein synthesis at the translocation step on the 70S ribosome and induces misreading of mRNA template.[1-3] Hygromycin B has been used to select mutants in a wide variety of cells including bacteria.[4,5] protozoans,[6] yeast,[7] fungi,[8-12] plants,[1,13-15] and mammalian cells.[16-18] Resistance to Hygromycin B is determined by a gene coding for a phosphotransferase that phosphorylates Hygromycin B, thereby making it inactive.[4,19] Hygromycin B is known to selectively penetrate cells that have been rendered permeable by virus infection.[20] This, combined with its potency in inhibiting translation, makes it an effective antiviral agent.[21-23]

The effective concentration required for selection varies depending on the type of cells, media, growth conditions, and the stage of the cell cycle. The effective dosage also depends on cell density. At higher cell densities somewhat greater doses are required to kill non-resistant cells. Hygromycin B can kill most bacterial cells under aerobic conditions at concentrations ranging from 10 to 1000 µg/ml. The sensitivity to Hygromycin B can be increased by increasing the pH of the medium. Also, the sensitivity appears to be greater at lower salt concentrations.

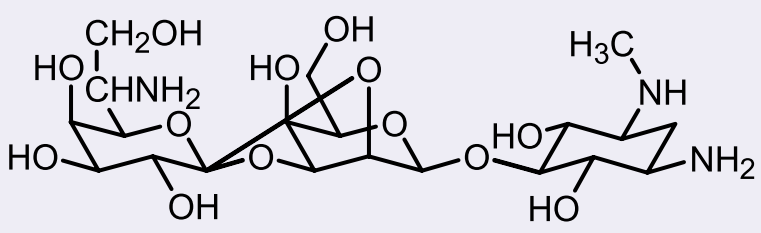
For most mammalian cell lines, the working concentration varies in the range from 50 to 1000 µg/ml. The killing of cells and their detachment from the plate, particularly at high cell density, may require a longer time compared to some other selection agents such as G 418. To generate a stable cell line expressing a protein of interest, one must determine the minimum concentration of Hygromycin B required to kill the untransfected host cell line. As natural resistance to Hygromycin B varies among various cell lines, it is best to generate a dose-response curve for each cell line used and then select the concentration that kills the most cells in a 7 to 10 day period.

Biological applications:

- Hygromycin B-resistance gene is useful in identification and selection of recombinant clones in a variety of cell types. A gene from *E. coli* encoding resistance to Hygromycin B can be isolated and cloned by recombinant DNA techniques into bacteria, yeast, or mammalian cells, making them Hygromycin B-resistant.
- Hygromycin B can be used as a screening agent for the transfer of a drug-resistant gene between prokaryotes and eukaryotes.
- Hygromycin B resistance may be used in construction of retroviral cloning vectors.
- Hygromycin B may be used as an antiviral drug in the treatment of murine coronaviral hepatitis.
- Hygromycin B may be used as an inhibitor of RNA translation.

CALBIOCHEM® now offers 5 forms of this frequently–used antibiotic selection agent!

$C_{20}H_{37}N_3O_{13}$ M.W. 527.5



O-6-Amino-6-deoxy-L-glycero-D-galacto-heptopyranosylidene-(1-2-3)-O-β-D-talopyranosyl(1-5)-2-deoxy-N^3-methyl-D-streptamine

PRODUCT INFORMATION

Product	Cat No.	Size
Hygromycin B, *Streptomyces* sp. Caramel-Colored Solution	**400051**	100 KU 250 KU 1 MU 5 MU 10 MU
Hygromycin B, *Streptomyces* sp. Caramel-Colored Solution Cell Culture-Tested	**400049**	100 KU 1 MU 5 MU 10 MU
Hygromycin B, *Streptomyces* sp. Off-White Lyophilized Solid Cell Culture-Tested	**400050**	100 mg 500 mg 1 g 5 g

Convenient and Economical Sterile-Filtered Solutions:

Hygromycin B, *Streptomyces* sp. 50 mg/ml in PBS Cell Culture-Tested	**400052**	5 ml 20 ml 50 ml
Hygromycin B, *Streptomyces* sp. 50 mg/ml in 25 mM HEPES, pH 7.4 Cell Culture-Tested	**400053**	5 ml 20 ml

Conversion of activity (Units/ml) to weight (mg/ml) of Hygromycin B
The following is a typical example for Cat. No. 400051 or 400049:
Product supplied as 1,000,000 Units with an activity of 405,670 units/ml
In-house technical information: Density = 1.13 g/ml; % Solids = 37.3

Calculation:
1,000,000 units at 405,670 units/ml

Volume = 2.465 ml
2.465 x 1.13 g = 2.785 g total weight (Hygromycin B + H_2O)
= 2,785 mg total weight
2,785 mg x 37.3% (% solids) = 1039.0 mg Hygromycin B
1039.0 mg Hygromycin B / 2.465 ml = **421.5 mg Hygromycin B/ml***

*The information provided on our product label reflects this ready conversion. You are provided with the number of mg Hygromycin B/ml.

Hygromycin B solution is stable for at least two years at +4°C and about one month at +37°C. It is sensitive to high concentrations of acids; however, a brief exposure to dilute acids does not affect its stability.

References:

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CALBIOCHEM® also offers a wide range of other antibiotics. For a complete listing ask for our Technical Bulletin "Drugs for Selection of Genetic Markers".

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