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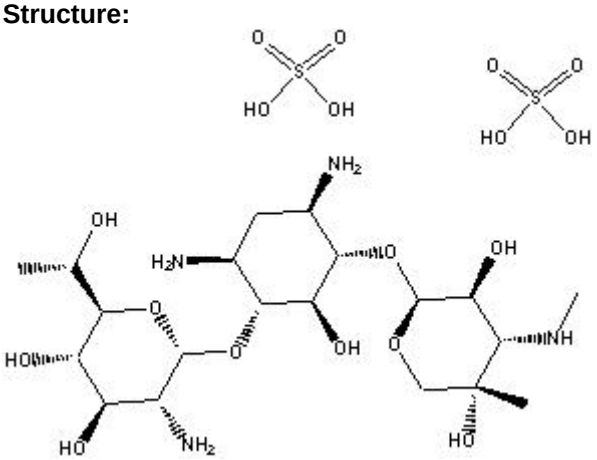
TECHNICAL INFORMATION

Catalog Number: 158782
Geneticin®, Disulfate Salt

Structure:



Molecular Formula: $C_{20}H_{40}N_4O_{10}\cdot H_2SO_4$
Molecular Weight: 692.7
CAS # 108321-42-2
Synonyms: Antibiotic G418; G418 Sulfate
Physical Appearance: White powder
Description: Geneticin is an aminoglycoside related to Gentamicin and is toxic to bacteria, yeast, higher plants, mammalian cells, protozoans and helminths. The resistance genes are located on transposons Tn601(903) and Tn5 and are bacterial in derivation, but they can be expressed in eucaryotic cells. By introducing these genes into cells, resistance to Geneticin is conferred. It is used for the selection of transfected mammalian cells, yeast, dictyostelium, plant and bacteria.
Certain transfection techniques in molecular biology and cell culture require the use of antibiotics to select for transfected cells. Geneticin® disulfate is an effective selecting agent utilized in killing procaryotic and eucaryotic cells. The mechanism of action occurs by inhibiting protein synthesis by binding to ribosomes of prokaryotic and eukaryotic cells, therefore killing the non-resistant cells. Due to this binding mechanism, cells will generally take several days to die. Resistance is conferred by the bacterial gene for aminoglycoside-3-phosphototransferase that can be expressed in eucaryotic cells. Cells may have variable resistance and can take up to one week to die; adherent cells may be more sensitive.
Characteristics:
High Potency - Potency is a measure of the amount of active ingredients per dry weight of material.
Low ED50 on Sensitive Cells - The lower the ED50 on sensitive cells, the greater the killing powder and stronger the selective pressure of Geneticin® disulfate.
High ED50 on Resistant Cells - The greater the ED50 on resistant cells, the higher the concentration of Geneticin® disulfate that can be used. This increases the selective pressure without adversly affecting the resistant cells.
Great Purity - Purity is an inverse measure of contaminants. Equal to potency plus percentage of inert ingredients.
Storage: The powder should be stored away from light. Avoid moisture uptake, as this can decrease the product's potency. Solutions should be aliquoted and stored frozen (-5 to -20°C). Solutions are generally stable for 3 to 6 months if kept frozen. Avoid multiple freeze-thaw cycles.
Working Concentration: The effective killing concentration of this antibiotic varies for cell type, media, growth conditions, metabolic rate of the cell, and stage of the cell cycle. Effective concentrations have been reported from 100 ug/mL to 5 mg/mL or greater. When using Geneticin® Disulfate in a new cell system or with a new lot with different potency, a full dose curve is suggested. Several points on the dose curve is suggested. Several points on the dose curve should be retested, as the potency (determined by a standard *B. subtilis* assay) may not exactly correlate to your system.
For Preparation of Stock Solution:
Using an analytical balance, weigh an appropriate amount of powder (using formula below). Reconstitute aseptically by adding the powder to distilled water (pH 5.6 to 7.0) to the desired concentration and sterile filter.
Notes: Do not filter cloudy solutions. This indicates that the powder may not yet be completely soluble and may result in the loss of powder on the filter, thereby decreasing activity in the resulting solutions.
Liquid media, NaCl, phosphate buffer, or organic solvents are not recommended for preparing stock solutions.
Stocks can be prepared either as simple weight/volume (w/v) or as active Geneticin® Disulfate (see formula below).
Working Concentrations:
Generally, initial selection of genetic tranformants requires a high concentration of Geneticin® Disulfate and a lower

concentration for maintenance. Growing conditions and other environmental factors will also have major influences on the amount of Geneticin® Disulfate needed to optimize selective pressures. Therefore, working concentrations may vary from cell line to cell line.

Cell Type	Concentration (active drug)	Application	Reference
Dictyostelium	a) 10 mg/L b) 30 mg/L	a) Cells grown in medium b) Cells plated on lyophilized bacteria	Hirth, et. al., *Proc. Natl. Acad. Sci.*, **v. 79**, 7356-7360 (1982).
Mammalian	a) 400 mg/L-1000 mg/L b) 200 mg/L	a) For Selection b) For Maintenance	Canaani and Berg, *Proc. Natl. Acad. Sci.*, **v. 79**, 5166-5170 (1982).
Plant	a) 25 mg/L-50 mg/L b) 10 mg/L	a) For Selection b) For Maintenance	Ursic, et. al., *Biochem. Biophys. Res. Comm.*, **v. 101:3**, 1031-1037 (1981).
Yeast	a) 500 mg/L b) 125 mg/L-200 mg/L	a) For Selection b) For Maintenance	Jimenez and Davies, *Nature*, **v. 287**, 869-871 (1980).
Bacteria	8 mg/L-16 mg/L	For Selection	Waitz, et. al., *Antimicrob. Agents Chemother.*, **v. 6:5**, 579-581 (1974).

Also Available:

Catalog Number	Description	Size
1672546 1672548	G418 Sulfate Solution	20 ml 50 ml