## Frequently Asked Questions

# **G-418 Sulfate**

#### 1. What is the difference between G-418 sulfate and Geneticin®?

They have the same chemical structure ( $C_{20}H_{40}N_4O_{10} \times 2H_2SO_4$ ). G-418 sulfate is the generic compound while Geneticin® was registered as trademark of Life Technologies Corporation in 1983.

#### 2. What is the mechanism of action of G-418 Sulfate?

G-418 Sulfate, like other aminoglycoside antibiotics, inhibits protein synthesis. It affects the 80S ribosomes and inhibits the elongation cycle.

#### 3. What is the relationship between the use of G-418 and neomycin resistance genes?

G-418 is an analog of neomycin sulfate, and has similar mechanism as neomycin. G-418 is commonly used in laboratory research to select genetically engineered cells. Neomycin cannot be used to select mammalian cells. Genes to confer neomycin resistance are known as neomycin resistance genes.

#### 4. Could be G-418 activity blocked?

Yes. There are two genes involved in G-418 resistance. The most common resistance gene associated with this antibiotic is the neomycin phosphotransferase II gene (*nptII*, Gb. DQ449903). This gene originates from transposon Tn5 has been included in many vectors. NPTII protein phosphorylates G-418, avoiding G-418 binding to the ribosome, providing resistance to the antibiotic.

Other neomycin phosphotransferase is known: neomycin phosphotransferase I (*nptl*). It was found in transposon Tn903 and confers resistance against G-418 in *Saccharomyces cerevisiae* and kanamycin in *Escherichia coli*.

Although there are many proteins that detoxify kanamycin, these phosphotransferases detoxify neomycin and kanamycin. The inclusion of these genes in different plasmids are indicated as neo/ kan genes. Promoters driven their expression are different in bacteria and mammals.

#### 5. What is the difference between neomycin and kanamycin?

They are aminoglycoside antibiotics: neomycin has a 4,5-di-substituted deoxystreptamine ring while kanamycin has a 4,6-di-substituted deoxystreptamine ring. Neomycin is used as selective pressure in eukaryotes and kanamycin is usually used in prokaryotes.



#### 6. Which is the optimal G-418 working concentration for selection?

Optimal concentration of G-418 should be determined for each cell line, media, vector and growth conditions. Recommended concentration ranges of G-418 selection are: mammalian cells (100-2000  $\mu$ g/mL), plant cells (10-100  $\mu$ g/mL), yeasts (500-1000  $\mu$ g/mL). For more info: <a href="https://cell-lines.toku-e.com/Antibiotics\_14.html">https://cell-lines.toku-e.com/Antibiotics\_14.html</a>

### 7. Can Gentamicin sulfate be used instead of G-418 for neomycin resistance selection in mammalian cells?

No. As selective agents gentamycin is a bactericidal antibiotic useful for selection of vectors including gentamicin resistance cassette (Gm<sup>R</sup>) while G-418 is used both in prokaryotes and eukaryotes to select vectors that include neomycin resistance cassette (Neo<sup>R</sup>). In some cases, gentamycin is used in mammalian cell cultures to avoid bacterial contamination.

#### 8. What is the difference between Geniticin® and Gentamycin?

Both antibiotics are the same drug class: aminoglycoside antibiotics. But they are different compounds. The next table compares both antibiotics.

	G-418/ Geniticin®	Gentamycin
CAS	108321-42-2	1405-41-0
Structure	HO H <sub>2</sub> N NH <sub>2</sub> OH OH OH OH OH	OH OH NH2 OH NH2 OH OH NH2
Formula	C <sub>20</sub> H <sub>40</sub> N <sub>4</sub> O <sub>10</sub> x 2H <sub>2</sub> SO <sub>4</sub>	C <sub>21</sub> H <sub>43</sub> N <sub>5</sub> O <sub>7</sub> x H <sub>2</sub> SO <sub>4</sub>
Molecular Weight	692.71 g/mol	575.67 g/mol
Action Spectrum	Bacteria, yeast, protozoans,	Bacteria (Pseudomonas aeruginosa,
	higher plant, and	Staphylococcus aureus) and some
	mammalian cells.	mycoplasmas

