

## Spectinomycin 100 mg/ml EZ-Pak™ Protocol

### Introduction

Spectinomycin is an aminocyclitol antibiotic isolated from *Streptomyces spectabilis*. It is effective against gram-positive and gram-negative bacteria, but most commonly used against *Gonococcus* species. It has also been used to monitor cell fate during leaf development and as a selection marker in plants to display successful transformation of the *Spcr* gene. Spectinomycin functions by binding the 30s ribosomal subunit, inhibiting tRNA translation and protein synthesis. Resistance to this antibiotic is conferred through the *aadA* gene and mutated *rpsE* gene.

The Spectinomycin Dihydrochloride Pentahydrate EZ Pak™ is the fastest and easiest way to make a set amount of sterile spectinomycin dihydrochloride solution. The kit includes pre-weighed spectinomycin dihydrochloride powder, a sterile filter and a sterile container for the filtered solution. No need to calculate, simply add the stated amount of deionized H<sub>2</sub>O, filter, and pour into the labeled bottle for easy usage. The EZ Pak™ includes high quality GoldBio spectinomycin dihydrochloride and the sterile solution is ready for tissue culture, bacterial media or any number of uses.

### Materials

- 1 Bottle of Spectinomycin dihydrochloride powder
- 1 Sterile empty bottle for solution
- 1 Sterile Filter

### Method

#### Reconstitution

1. Warm Spectinomycin powder bottle to Room Temperature.
2. Add specified volume of dH<sub>2</sub>O to Spectinomycin powder bottle.

Product Catalog #	Volume of H <sub>2</sub> O to Add	Final Volume
<a href="#">S-140-EZ25</a>	23.5 ml	25 ml
<a href="#">S-140-EZ50</a>	47.0 ml	50 ml
<a href="#">S-140-EZ100</a>	94.0 ml	100 ml

3. Mix until all product goes into solution.
4. Sterile Filter:
  - a. For S-140-EZ25, EZ50 and EZ100
    - i. Remove vacuum filter from packaging.
    - ii. Attach vacuum hose according to instructions on filter packaging.
    - iii. Add solution to the upper cup of the filter.
    - iv. Apply vacuum pressure and let all the solution in the top cup flow through the filter into the bottom cup. Stop vacuum when all the solution is filtered.
    - v. Remove vacuum attachment from the filter and close the bottle with provided sterile cap.
    - vi. Tightly seal solution bottle and store at -20°C. Make aliquots if desired. Discard filter.
5. Use Spectinomycin at a final concentration of 100 µg/ml.