

# Analysis of Apramycin Sulfate According to British Veterinary Pharmacopeia

Apramycin Sulfate is an aminoglycoside antibiotic that is used to treat colibacillosis and salmonellosis infections in calves, pigs, lambs and poultry. It is used as oral dosages as well as feed premixes and injectable formulations.

British Veterinary Pharmacopeia specifies tests to determine purity and quality of Apramycin Sulfate. Cation-exchange chromatography with post-column derivatization using Ninhydrin reagent is the recommended method for analysis of 3-hydroxyapramycin, lividamine, caerulomycin, 2-deoxystreptomycin and other related substances commonly found as impurities in Apramycin Sulfate.

Pickering Laboratories' post-column derivatization systems and patented Trione Ninhydrin reagent can be successfully used to analyze the purity of Apramycin Sulfate according to Veterinary Pharmacopeia.

## Method

### Analytical Conditions

**Column:** Dionex IonPac Fast Cation I column, 4 x 250 mm (Product # SP5391)

**Flow Rate:** 0.8 mL/min

**Column Temperature:** 30 °C

**Mobile Phase A:** Aqueous solution of 1.961% w/v of sodium citrate, 0.08% w/v of phenol and 0.5% v/v of thiodiglycol, adjusted to pH 4.25

**Mobile Phase B:** Aqueous solution of 4.09% w/v of sodium chloride, 3.922% w/v of sodium citrate and 0.08% w/v of phenol, adjusted to pH 7.4

**Injection Volume:** 20 µL

### Post-column Conditions

**Post-Column System:** Onyx PCX, Pinnacle PCX or Vector PCX

**Reagent 1:** Trione® Ninhydrin reagent

**Reactor Volume:** 1.0 mL

**Reactor Temperature:** 130 °C

**Flow Rate:** 0.5-1.0 mL/min

**Detection:** DAD 568 nm

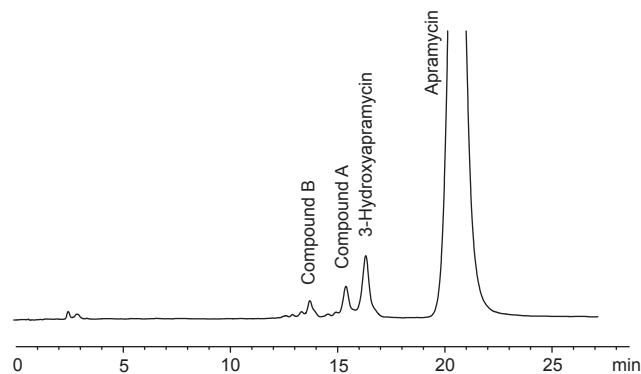


Fig 1. Chromatogram of British Pharmacopeia Chemical Reference Substance (BPCRS)

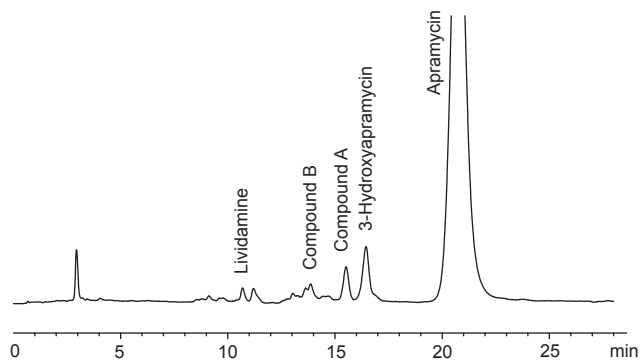


Fig 2. Chromatogram of commercially available Apramycin Sulfate

HPLC Program		
Time	Mobile Phase A %	Mobile Phase B %
0	75	25
3	75	25
9	0	100
30	0	100
30.1	75	25
40	75	25