Amikacin

The method Amikacin Sulfate as Solution and Powder for Injection was published in the Minilab Manual, Supplement 2014 to Volume II, Method 6.73, pages 4-7. It was modified by elimination of the need for detection by exposure to iodine vapor, ninhydrin staining, and sulfuric acid staining as specified in the Minilab Manual. Amikacin spots on silica gel TLC aluminum plates were found to quench fluorescence under 254 nm UV light after mobile phase development and heating. Users may consider that elimination of the need for detection by iodine vapor and staining reagents makes this method safer, faster, and more convenient, especially for application in the field.

The modified method was developed using a product containing 500 mg of amikacin disulfate/2 mL of injection solution. In the Minilab Manual method for this product, the 100% working sample and standard solutions are prepared with a concentration of 3.75 mg amikacin free base/mL, and 2.00 µL are spotted (7.50 µg). In this modified method, 0.600 mL of the product was dissolved in a 10 mL volumetric flask with water to give a 100% sample solution with a concentration of 15.0 mg amikacin disulfate/mL [(0.600 mL× 500 mg/2.00 mL) ÷ 10.0 mL = 15.0 mg/mL]. To prepare the 100% standard solution with the same concentration of amikacin disulfate, 113 mg of commercial analytical grade amikacin free base standard (Sigma-Aldrich, No. PHR1654-1G) was diluted with 10.0 mL of water [113 mg x (782 g/mol amikacin disulfate/587 g/mol amikacin free base) ÷ 10.0 mL = 15.0 mg//mL]. Two uL of these solutions were spotted (30.0 ug).

The plate was developed with the same mobile phase as is in the Minilab Manual method: methanol- concentrated ammonium hydroxide (1:1). Instead of exposing the plate to iodine vapor or staining with sulfuric acid and ninhydrin as in the Minilab Manual method, the plate was heated on a hotplate (e.g., for 30 min at 180°C), and amikacin was detected as fluorescence quenching spots under 254 nm UV light as is shown in the photograph of a plate below.

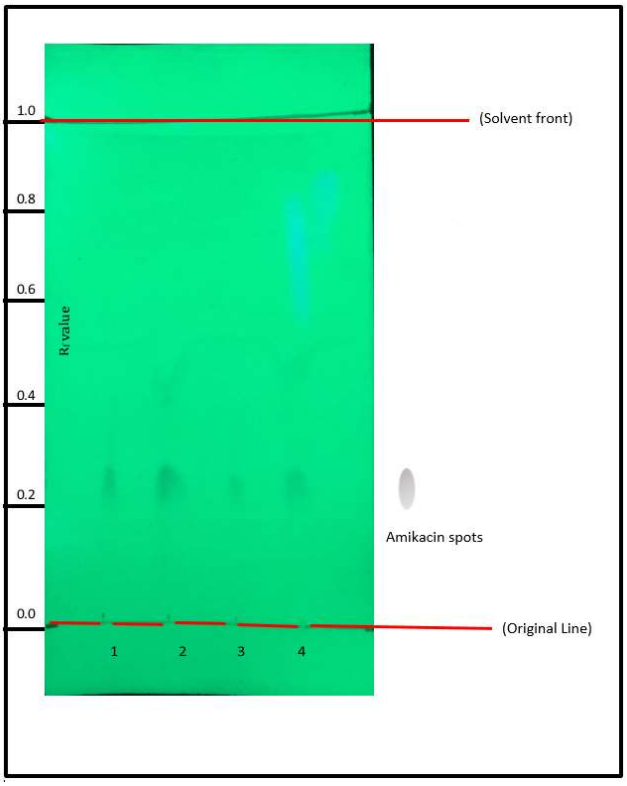
CHROMATOPLATE OBSERVED UNDER 254 NM UV LIGHT

Run No.1:
Upper working standard
representing 100% of total amikacin

Run No.2:
A product of good quality with
Acceptable amikacin content

Run No.3:
A product of poor quality with
Unacceptable low amikacin content

Run No.4:
Lower working standard
Representing 80% of total amikacin



(*A drug product of poor quality was simulated by diluting the 100% working sample solution of a drug product of good quality with water to one-third of the theoretical value.)

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