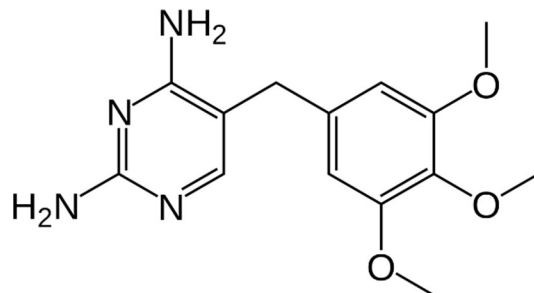


Trimethoprim
80 mg in Co-Formulation Tablets
with 400 mg Sulfamethoxazole
(A screening method for sulfamethoxazole in this combined formulation is available in
GPHF Minilab Manual Volume II, Method 6.39, pp. 184-187)

Structure:



Molecular Formula and Mass: C₁₄H₁₈N₄O₃ - 290.323

Category: Antibacterial

Sample:

Grind one tablet and dissolve in 100 mL of methanol. Shake for at least 10 min and filter. Dilute 1 mL of the stock solution with 2 mL of methanol. Final concentration of sample solutions = 0.267 mg/mL, which is the required concentration representing 100%.

Standards:

High Standard:

The high limit is 115%; therefore the concentration of the high standard = 0.267 mg/mL × 1.15 = 0.307 mg/mL. Weigh approximately 30.7 mg of standard and dissolve it in 100 mL methanol. If you weighed 30.6 mg of standard, dissolve it in: 30.6 mg ÷ 0.307 mg/mL = 99.7 mL of methanol. This makes the high standard solution concentration equal to 0.307 mg/mL.

Low Standard:

The low limit is 85%; therefore the concentration of the low standard = (0.267 mg/mL × 0.85) = 0.227 mg/mL. Dilute 3.00 mL of high standard to 4.00 mL by adding 1.00 mL of methanol. This gives a concentration of 0.307 mg/mL × 3.00 mL ÷ 4.00 mL = 0.230 mg/mL, which is 86.2%.

Spotting:

Spot on the 5 × 10 cm silica gel TLC aluminum plate with 3.00 µL aliquots as follows:

Left spot	low standard (85%) = 0.690 µg
Center Spot	100% sample = 0.801 µg
Right Spot	high standard (115%) = 0.921 µg

Development:

Mix 24.0 mL of ethyl acetate and 8.00 mL of methanol. Develop the plate in a small glass chamber with approximately 20.0 mL of this solution until the solvent front reaches within 1 cm of the top of the TLC plate.

(R_f = 0.21)

Detection:

UV: Dry the plate and observe under ultraviolet light at 254 nm. Observe the intensities and the sizes of the spots.

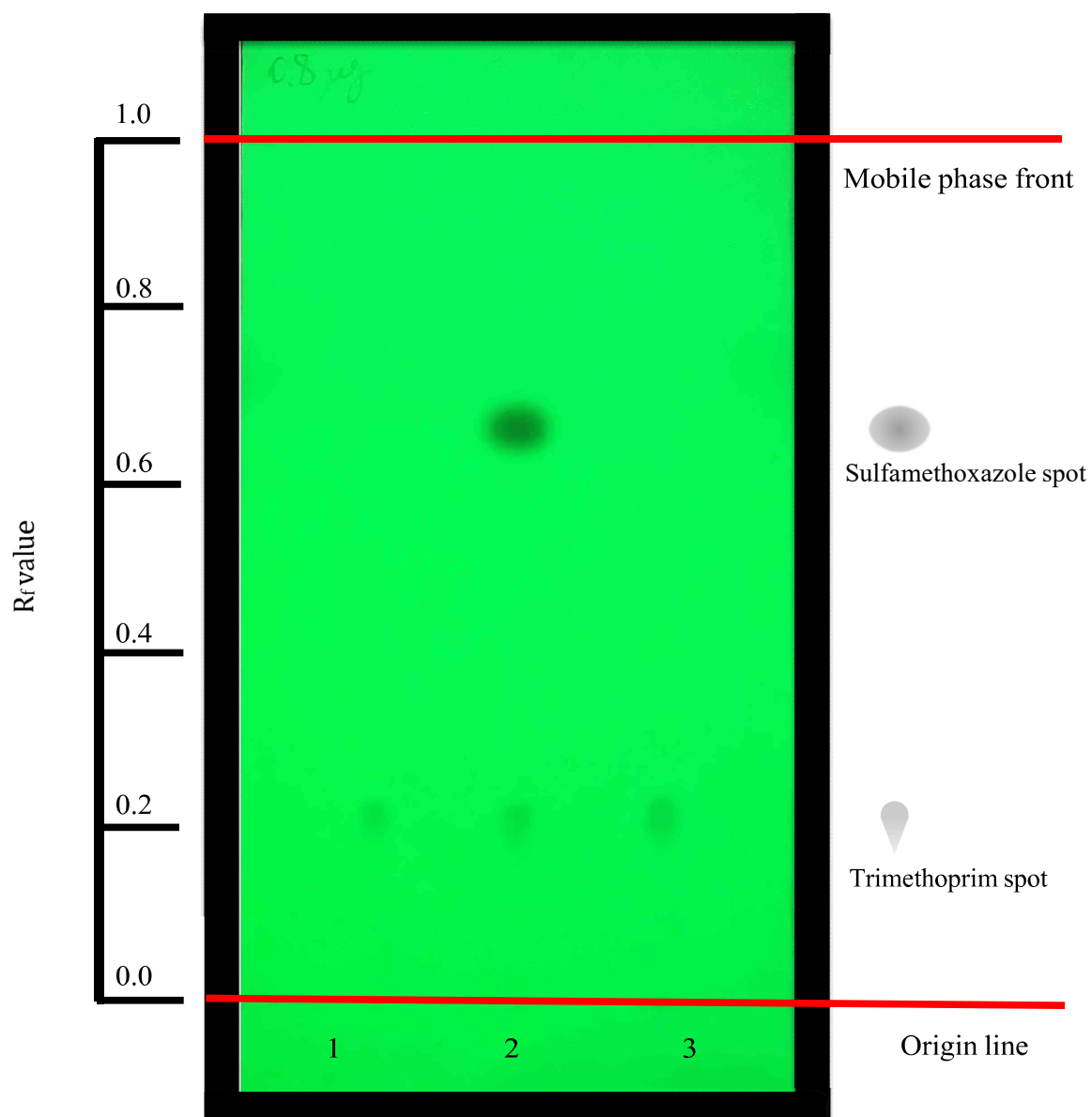


Plate observed under ultraviolet light at 254 nm.

Lane 1: Low standard (85%) = 0.690 μg

Lane 2: 100% sample = 0.801 μg

Lane 3: High standard (115%) = 0.921 μg

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