DEPARTMENT OF DRUG ADMINISTRATION
National Medicines Laboratory
ANALYTICAL METHOD VALIDATION COMMITTEE

Metronidazole and Diloxanide Furoate Suspension

Analytical Profile No.: MetrDilo L 076/077/AP 069

Metronidazole and Diloxanide Furoate Suspension contains not less than 90 % and not more than 110 % of the stated amount of Metronidazole and Diloxanide furoate.

1. Identification: In the Assay, the principle peak in the chromatogram obtained with the test solution corresponds to the peak in the chromatogram obtained with the reference solution.

Tests:

2. pH: 5 - 6.5

3. wt/ml: As per manufacturer's specification

4. Assay: *Determine by Liquid Chromatography*

4.1 Test Solution: Weigh accurately about 100 mg equivalent of Metronidazole and 125 mg of Diloxanide Furoate in 100 ml volumetric flask, make up the volume with mobile phase up to the mark and stir mechanically for 30 minutes. Further dilute 5 ml of this solution to 50 ml with same solvent, mix well. Filter the resulting solution through 0.2 µm membrane filter.

4.2 Reference Solution: Weigh accurately about 80.4 mg Metronidazole Benzoate RS and 62.5 mg of Diloxanide Furoate RS in a 50 ml volumetric flask, add about 30 ml of mobile phase and sonicate for about 15 minutes; cool to room temperature and make up the volume with same solvent. Further dilute 5 ml of this solution to 50 ml with same solvent, mix well.

4.3 Chromatographic system

Column:	25 cm x 4.6 mm, C18, 5 µm
Injection volume:	10 µl
Flow rate:	1.0 ml per minute
Column Temperature:	30 ºC
Wavelength:	241nm
Detector:	UV

Mobile phase: A mixture of 30 volumes of Buffer prepared by dissolving 1.625 g of Potassium dihydrogen phosphate and 0.3 g of Dipotassium hydrogen phosphate in 1000 ml of water, adjusting pH to 5.5 with orthophosphoric acid, 30 volumes of Acetonitrile and 40 volumes of Methanol.

4.4 Procedure: Inject the reference solution five times and test solution. The test is not valid unless the column efficiency is not less than 2000 theoretical plates, the tailing factor is not more than 2.0, the relative standard deviation for replicate injections in not more than 2.0 % and the resolution between Metronidazole and Diloxanide Furoate is not less than 2.

Calculate the content of Metronidazole and Diloxanide Furoate in the suspension.

5. Other tests: As per pharmacopoeial requirement.