Government of Nepal
Ministry of Health and Population
Department of Drug Administration
National Medicines Laboratory
Quality and Method Validation Section

Analytical Profile of Bedaquiline Tablets

Analytical Profile No.: Bedaq 081/082 AP 168

Bedaquiline Tablets contain not less than 90.0% and not more than 110.0% of the stated amount of Bedaquiline.

Usual Strength: 100 mg

1. Identification:

In the Assay, the principal peak in the chromatogram obtained with the test solution corresponds to the peak in the chromatogram obtained with the reference solution.

2. Dissolution: *Determine by liquid chromatography*

2.1 Dissolution Parameters:

Apparatus: Basket

Medium: 900 ml 0.01 N Hydrochloric Acid

Speed and Time: 150 rpm and 45 minutes

2.2 Test Solution: After completion of the test withdraw a specimen from the dissolution medium, dilute if necessary.

2.3 Reference Solution: Weigh 35.0 mg of Bedaquiline fumarate WS accurately and transfer in 50 ml of a completely dried volumetric flask. Add 10 ml of N, N-dimethylformamide, and sonicate to dissolve. Make up the volume with the dissolution medium, and mix. Dilute 5 ml of the solution to 25 ml with the dissolution medium. Further, dilute 5 ml of the solution to 25 ml with the dissolution medium.

2.4 Procedure: Measure the absorbance of sample preparation at a wavelength of about 280 nm using UV spectrophotometry taking the dissolution medium as blank.

2.5 Limit: NLT 70 % (Q) of the stated amount.

3. Assay: *Determine by liquid chromatography*

3.1 Diluent: Prepare a mixture of 900 ml of methanol, and 100 ml of dichloromethane, add 0.5 ml of trifluoroacetic acid, and mix well.

3.2 Test solution: Weigh the content of 20 tablets and calculate the average weight. Weigh the powder equivalent to 100 mg of Bedaquiline in 500 ml of dry volumetric flask, add 400 ml of diluent, and sonicate for 30 minutes to dissolve with intermittent shaking at intervals of 5 minutes. Cool the sample solution to room temperature then, make up the volume with diluent and mix. Then centrifuge the sample for 5 minutes at 3000 rpm. Dilute 5 ml of supernatant liquid to 50 ml with diluent. Mix and filter the solution through 0.25-micron filter paper.

3.3 Reference solution: Weigh accurately about 30.5 mg of Bedaquiline fumarate WS and transfer to a 250 ml completely dried volumetric flask. Dissolve in 180 ml of diluent with the aid of ultrasound for 20 min at an interval of 5 min to dissolve. Dilute to the volume with diluent. Dilute 10 ml of the solution to 50 ml with the diluent and, mix, and filter through a 0.25-micron filter paper.

3.4 Chromatographic system:

Column: C18 (4.6mmX 150-mm, 5µm)

Flow rate: 1.5 ml/min

Wavelength: 225 nm

Injection volume: 10 µl

Column Temperature: 45ºC

Sample Temperature: 20ºC

Mobile phase A: Transfer 1 ml of trifluoroacetic acid to 1000 ml water, mix, and filter it with 0.45-micron filter paper.

Mobile phase B: Methanol

Gradient program:

Time (in min)	Mobile phase A (% v/v)	Mobile phase B (% v/v)
0.01	30	70
12.0	30	70
13.0	10	90
16.0	10	90
17.0	30	70
22.0	30	70

3.5 Procedure: Inject the reference solution five times and test the solutions. 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, and the relative standard deviation for replicate injections is not more than 2.0%. Measure the peak responses. Calculate the content of Bedaquiline.

4. Other tests: As per Pharmacopoeial requirements.