DEPARTMENT OF DRUG ADMINISTRATION
National Medicines Laboratory
ANALYTICAL METHOD VALIDATION COMMITTEE

Metformin (Immediate Release) & Linagliptin Tablets

Analytical Profile No.: LMI 074/075/AP 030

Metformin (Immediate Release) and Linagliptin Tablets contain not less than 90 % and not more than 110 % of the stated amount of Metformin and Linagliptin.

1. Identification:

1.1. Metformin HCl: In the assay, the principle peak in the chromatogram obtained with the sample solution should correspond to the peak in the chromatogram obtained with the reference standard solution of Metformin HCl.

1.2. Linagliptin: In the assay, the principle peak in the chromatogram obtained with the sample solution corresponds to the peak in the chromatogram obtained with the reference standard solution of Linagliptin.

Tests:

2. Dissolution:

2.1 Metformin Hydrochloride:

2.1.1 Dissolution Parameters:

Apparatus:	Paddle
Medium:	900 ml, 0.1 N HCl
Speed and time:	50 rpm for 45 minutes
Temperature:	37 ± 0.5 °C

Withdraw a suitable volume of the medium and filter.

Determine by UV-Vis Spectroscopy

2.1.2 Test Solution: Dilute a suitable volume of filtrate with dissolution medium to make a solution having concentration similar to that of reference solution.

2.1.3 Reference Solution: A 0.02 mg/ml solution of Metformin Hydrochloride RS in dissolution medium. (20 ppm)

2.1.4 Procedure: Measure the absorbance of test solution and reference solution at 232 nm using dissolution medium as blank. Calculate the percentage release of Metformin Hydrochloride in each tablet at specified time.

2.1.5 Limit: Not less than 70 % (D) of the stated amount of Metformin Hydrochloride.

2.2 Linagliptin: *Determine by Liquid chromatography*

2.2.1 Dissolution Parameters: Proceed as directed under Metformin

2.2.2 Test Solution: Filter the resulting solution through 0.2 µ membrane filter. Use the filtrate.

2.2.3 Reference Solution: A 0.0025 mg/ml solution of Linagliptin RS in dissolution medium (2.5 ppm). Filter the resulting solution through 0.2 µ membrane filter.

2.2.4. Chromatographic System: Proceed as described under the Assay of Metformin and Linagliptin.

2.2.5 Procedure: Separately inject 20 µl of reference solution, test solution and blank solution (dissolution medium) and obtain the respective chromatograms. The test is not valid unless the column efficiency is not less than 2000 theoretical plates, tailing factor is not more than 2.0 and the relative standard deviation of five replicate injections is not more than 2.0 %. Measure the peak responses and calculate the % release of the drug.

2.2.6 Limit: Not less than 80 % (D) of the stated amount of Linagliptin.

3. Uniformity of content (Linagliptin): *Determine by Liquid Chromatography*

3.1 Test solution: Weigh 10 tablets and transfer individually to 100 ml volumetric flask. Add 60 ml of mobile phase and sonicate for 15 minutes to disperse the tablet, cool to room temperature and make up the volume with mobile phase. Dilute if necessary with mobile phase to make a solution having similar concentration to that of reference solution. Filter the resulting solution through 0.2 µm membrane filter.

3.2 Reference Solution: A 0.025 mg/ml solution of Linagliptin RS in mobile phase (25 ppm). Filter the resulting solution through 0.2 µ membrane filter.

3.3 Chromatographic system and Procedure: Proceed as directed under Assay.

3.4 Limit: 85 % - 115 % of the stated amount.

4. Assay: *Determine by Liquid Chromatography*

4.1 Buffer: 0.02 M phosphate buffer pH 4.

Weigh 0.675 g of Potassium dihydrogen orthophosphate in a 250 ml volumetric flask, add about 200 ml water to dissolve it completely. Make up the volume with water and adjust the pH to 4.0 with 10 % OPA.

4.2 Test solution: Weigh individually 20 tablets and crush them into fine powder. Weigh accurately a quantity of powder equivalent to 250 mg of Metformin HCl and transfer into 100 ml volumetric flask, add 70 ml of mobile phase, sonicate for 15 minutes to dissolve, cool and make volume to 100 ml with same solvent. Filter the final solution through 0.2 µm membrane filter. (2500 ppm)

4.3 Reference Solution:

4.3.1 Metformin HCl: Weigh accurately about 50 mg Metformin HCl reference standard and transfer into 20 ml volumetric flask and sonicate to dissolve with mobile phase.

4.3.2 Linagliptin: Weigh accurately about 12.5 mg Linagliptin reference standard and transfer it into 100 ml volumetric flask. Add about 70 ml of mobile phase, dissolve with the aid of sonicator and make up the volume to 100 ml with same solvent.

4.3.3 Mix Reference Solution: Transfer 2 ml Linagliptin reference solution to Metformin HCl solution and make up the volume to mark with mobile phase. Filter the final solution through 0.2 µm membrane filter.

4.4 Chromatographic system:

Column:	a stainless steel column 15 cm x 4.6 mm, packed with octadecylsilane bonded to porous silica (5 µm)
Flow rate:	1.0 ml per min
Wavelength:	265 nm (Metformin), 295 nm (Linagliptin)
Detector:	PDA Detector
Injection volume:	20 µl
Column temperature:	25 °C
Mobile phase:	a mixture of 70 volumes of 0.02 M phosphate buffer pH 4.0 and 30 volumes of acetonitrile

4.5 Procedure: Separately inject 20 µl of reference solution, test solution and blank solution and obtain the respective chromatograms. The test is not valid unless the column efficiency is not less than 2000 theoretical plates, tailing factor is not more than 2.0 and the relative standard deviation of five replicate injections is not more than 2.0 %. Resolution between two peaks should not be less than 2.0. Measure the peak responses and calculate the amount of Metformin HCl and Linagliptin in each tablet.

5. Other tests: As per pharmacopoeial requirements.