DEPARTMENT OF DRUG ADMINISTRATION
National Medicines Laboratory
ANALYTICAL METHOD VALIDATION COMMITTEE

MetforminSR &Sitagliptin Tablet

Analytical Profile No.: Met Sit 073/074/AP 016

Metformin SR &Sitagliptin Tablet contains not less than 90% and not more than 110% of the stated amount of Metformin and not less than 90% and not more than 110% of the stated amount of Sitagliptin.

1. Identification:

1.1. MetforminHCl:

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.

1.2. Sitagliptin:

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.

2.0 Dissolution: Determine by liquid chromatography.

2.1 Dissolution (Sitagliptin):

2.1.1 Dissolution Parameters:

- **Apparatus**: Basket
- **Medium:** 900 ml of Water
- **Speed and Time:** 100rpm & 30 minutes
- **Temperature:** 37˚C ± 0.5˚C

Withdraw a suitable volume of the medium and filter.

2.1.2 Chromatographic condition:

Determine by liquid chromatography, as described in the Assay

2.1.3 Test Solution: Use the filtrate.

2.1.4 Reference Solution:

Weigh accurately about 36.885 mg of Sitagliptin Phosphate Monohydrate WS eq. to 27.75 mg Sitagliptin and dissolve in water to produce 50 ml. Dilute 5 ml of this solution to 50 ml with water.

2.1.5 Procedure:

Use the chromatographic system as described in the Assay.Inject the reference solution and the test solution.Calculate the content of Sitagliptin.

2.1.6 Limit:

D. Not less than 75.0 % of the stated amount of Sitagliptin.

2.2 Dissolution (Metformin Sustained release tablet):

2.2.1 Dissolution Parameters: Determine by UV Spectroscopy

- **Apparatus:** Basket
- **Medium:** 1000 ml of phosphate buffer pH 6.8
- **Speed and Time:** 100 rpmand1 hour, 3 hours & 10 hours
- **Temperature**: 37˚C ± 0.5˚C

2.2.2 Test solution:

Dilute the filtrate, if necessary, with dissolution medium.

2.2.3 Reference solution:

Weigh accurately about 25 mg of Metformin hydrochloride WS and transfer into 100 ml volumetric flask. Dissolve with water and make up the volume to 100 ml with water. Dilute 2 ml of the standard solution to 100 ml with dissolution medium.

2.2.4 Procedure:

Measure the absorbance of the reference and test solution at about 232 nm. Calculate the content of Metformin.

2.2.6 Limit:

1 hr: 25 % to 50 % of the stated amount

3rdhr: 45% to 70 % of the stated amount

8thhr: NLT 80 % of the stated amount

3.0 Uniformity of the content (Sitagliptin):

Determine by liquid chromatography, as described in the Assay, using the following test solution and reference solution.

3.1 Test Solution:

Take a single tablet, crush and transfer to 100 ml volumetric flask with the help of 70 ml diluents. Sonicate for 30 min and diluted to 100 ml with diluents. Filter the solution and dilute 5 ml of filtrate to 50 ml with the diluent.Prepare similarly for 9 more tablets.

3.2Reference Solution:

Weigh accurately about 33 mg of Sitagliptin Phosphate Monohydrate WS eq. to 25 mg Sitagliptin in 50 ml volumetric flask and add 35 ml diluents. Dissolve by sonication and diluted to 50 ml with diluents. Dilute 5 ml resulting solution to 50 ml with diluents.

4. Assay:

4.1 Diluents: 5 volume of Acetonitrile: 95 volume of 0.1% v/v orthophosphoric acid in water.

4.2 Chromatographic system

- **Column:** C18, 250mm x 4.6 mm, 5 µm
- **Flow rate:** 1.0 ml/min
- **Injection volume:** 20 µl

- **Wavelength:** 205 nm
- **Detector:** UV Detector
- **Column temperature:** 30°C

Mobile phase: Buffer: Acetonitrile (75:25)

Buffer: Weigh 1.36gm KH_2PO_4, dissolve in 900ml water and add 2.5ml of triethylamine. Adjust to pH 3.5 with dilute orthophosphoric acid and dilute to 1000ml with water.

4.3Test Solution:

Weigh individually 20 tablets & crush the tablet into fine powder. Weigh accurately the powder eq. to 50 mg of Metformin HCl and transfer into 100 ml volumetric flask, add 70 ml of diluents & sonicate for 15 minutes to dissolve. After sonication, dilute to 100 ml with diluents and stir for 20 minutes, filter through filter paper and dilute 5 ml filtrate to 25 ml with diluents.

4.4Reference Solution (850 mg metformin+ 50 mg Sitagliptin Tablet):

4.4.1 Metformin HClReference solution:

Weigh accurately about 25 mg of Metformin HCl WS and transfer into 50 ml volumetric flask. Add about 35 ml of diluent and sonicate for about 10 minutes and make up the volume to 50 ml with diluents.

4.4.2SitagliptinReference solution:

Weigh accurately about 30 mg eq. of Sitagliptin from Sitagliptin Phosphate WS into separate 100 ml volumetric flask. Add about 70 ml of diluent and sonicate for about 10 minutes and make up the volume to 100 ml with diluents. Dilute 5 ml of this solution to 50 ml with diluents.

4.4.3Reference combination:

Pipette 5 ml of Metformin HCl standard solution &Sitagliptin standard solution into 25 ml volumetric flask and make up the volume to 25 ml with diluent.

Note: Adjust the concentration of the standard preparation and sample preparation depending upon the strength of Metformin and Sitagliptin Tablet.

4.5Procedure:

Inject the reference solution. The test is not valid unless the resolution between Metformin and Sitagliptin is not less than 2, the column efficiency is not less than 2000 theoretical plates, tailing factor is not more than 2.0 and the relative standard deviation for replicate injections in not more than 2.0% resolution between two peaks should be not less than 2.Inject the reference solution and the test solution.

Calculate the content of Metformin and Sitagliptin in the tablets.

5. Other tests: As per pharmacopoeial requirements.