ANALYTICAL METHOD VALIDATION COMMITTEE FOR NON PHARMACOPOEIAL PRODUCT

DEPARTMENT OF DRUG ADMINISTRATION

Linagliptin Tablets

Linagliptin Tablets contain not less than 90 % and not more than 110 % of the stated amount of Linagliptin.

1. Identification:

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 linagliptin.

2. Dissolution Test:

2.1 Dissolution Parameter:

2.1.1 Medium : 900ml 0.1 M HCl
2.1.2 Apparatus : Basket
2.1.3 Rotation : 50 RPM
2.1.4 Temperature : 37˚C ± 0.5˚C
2.1.5 Time : 45 minutes

2.1.6. Dissolution Medium Preparation: Dilute 8.5 ml of conc. HCl with water to 1000 ml.

2.1.7 Standard Preparation:

Weigh accurately about 28 mg of working standard of linagliptin and transfer into 100 ml volumetric flask. Dissolve in the 0.1 M HCl and make up the volume to 100 ml with 0.1 M HCl. Pipette 2 ml of this solution and dilute to 20 ml with 0.1 M HCl. Again dilute 2 ml of the resulting solution to 20 ml with dissolution medium. Filter through 0.2 micron filter paper.

2.1.8. Sample preparation

Place 1 tablet in each dissolution vessel and run the apparatus as per above condition and collect the sample solution from each jar at specified time. After the completion of the dissolution, filter the resulting solution and again filter through 0.2 micron filter paper **for 2.5 mg linagliptin tablet. For 5 mg Linagliptin tablet, dilute 5 ml of the resulting filtrate to 10 ml with the dissolution medium and filter through 0.2 micron filter paper.**

2.1.9. Chromatographic system:

2.1.9.1 Column: 250 X 4.6 mm (C 18)
2.1.9.2 Flow rate: 1.0 ml/min
2.1.9.3 Wave length: 295 nm
2.1.9.4 Injection volume: 20 µl
2.1.9.5 Column Oven Temperature: 25˚c
2.1.9.5 Mobile phase

2.1.9.5.1 0.02 M Phospate buffer pH 4.0: Weigh accurately about 2.7 g of potassium dihydrogen orthophosphate and dilute to 1000 ml with water.

2.1.9.5.2 Mobile phase: Mix Acetonitrile and 0.02 M Phosphate buffer pH 4.0 in the ratio (30:70). Cool to room temperature and filter the solution through 0.2 micron filter paper using vacuum pump.

2.1.10. Procedure:

Inject 20 µl of standard preparation five/six times. 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 in not more than 2.0%. After the completion of the system suitability test parameter, inject 20 µl of each of the sample solution separately. Calculate the release of drug in the Linagliptin tablet by using following formula:

Calculation:

Linagliptin (%):

$$=\frac{Area\ of\ spl}{Area\ of\ std} \times \frac{conc.of\ std}{conc.of\ spl} \times std\ potency\ \% \times \frac{100-LOD/WC}{100}\ x\ 100\ \%$$

2.1.11. Tolerence Limit:

3. Assay:

3.1 Chromatographic system:
3.1.1 Column: 250 X 4.6 mm (C 18)
3.1.2 Flow rate: 1.0 ml/min
3.1.3 Wave length: 295 nm
3.1.4 Injection volume: 20 µl
3.1.5 Column Oven Temperature: 25˚c

3.2 Mobile Phase:
3.2.1 0.02 M Phosphoric acid Buffer pH 4.0: Weigh accurately about 2.7 g of potassium dihydrogen orthophosphate and dilute to 1000 ml with water.

3.2.2 Mobile phase: Mix Acetonitrile and 0.02 M Phosphate buffer pH 4.0 in the ratio (30:70). Cool to room temperature and filter the solution through 0.2 micron filter paper using vacuum pump.

3.3 Diluents: 0.1 M HCl

3.4 Standard Preparation:

Weigh accurately about 30 mg of working standard of linagliptin and transfer into 100 ml volumetric flask. Dissolve in the 0.1 M HCl and make up the volume to 100 ml with 0.1 M HCl.

Pipette 2 ml of this solution and dilute to 20 ml with 0.1 M HCl. Again dilute 2 ml of the resulting solution to 20 ml with 0.1 M HCl. Filter through 0.2 micron filter paper.

3.5 Sample Preparation:

Weigh individually 20 tablets and crush the tablet to fine powder. Weigh accurately the powder equivalent to 10 mg of linagliptin and transfer into 100 ml volumetric flask. Add about 70 ml of diluent, sonicate for about 10 minutes and cool the solution to room temperature and make up the volume to 100 ml with diluents. Centrifuge the solution. Dilute 3 ml of the resulting solution to 100 ml with diluent. Filter the solution with 0.2 micron filter paper.

3.6 Procedure

Inject 20 µl of standard preparation five/six times. 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 in not more than 2.0%. After the completion of the system suitability test parameter, inject 20 µl of each of the sample solution separately. Calculate the content of Linagliptin in each tablet by using following formula:

Linagliptin per tablet:

$$= \frac{Spl\ Peak\ Area}{Std\ Peak\ Area} \times \frac{conc\ of\ std}{conc\ of\ spl} \times std\ potency\ \% \times \frac{100 - LOD/WC}{100}\ \ x\ Average\ weight$$

4. Uniformity of content:

4.1 Chromatographic system: Same as Assay

4.2 Mobile Phase:

4.2.1 0.02 M Phosphoric acid Buffer pH 4.0: Weigh accurately about 2.7 g of orthophosphoric acid and dilute to 1000 ml with water.

4.2.2 Mobile phase: Mix Acetonitrile and 0.02 M Phosphate buffer pH 4.0 in the ratio (30:70). Cool to room temperature and filter the solution through 0.2 micron filter paper using vacuum pump.

4.3 Diluents: 0.1 M HCl

4.4 Standard Preparation:

Weigh accurately about 30 mg of working standard of linagliptin and transfer into 100 ml volumetric flask. Dissolve in the 0.1 M HCl and make up the volume to 100 ml with 0.1 M HCl. Pipette 2 ml of this solution and dilute to 20 ml with 0.1 M HCl. Again dilute 2 ml of the resulting solution to 20 ml with 0.1 M HCl. Filter through 0.2 micron filter paper.

4.5 Sample Preparation:

Weigh 10 tablet individually and transfer into 100 ml volumetric flask. Add about 70 ml of diluent, sonicate for about 10 minutes and cool the solution to room temperature and make up the volume to 100 ml with diluents. Centrifuge the solution. **Dilute 3 ml of the resulting solution to 25 ml with diluent for 2.5 mg linagliptin tablet. For 5 mg tablet, dilute 3 ml of the resulting solution to 50 ml with diluent.** Filter the solution with 0.2 micron filter paper.

4.6 Procedure

Inject 20 µl of standard preparation five/six times. 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 in not more than 2.0%. After the completion of the system suitability test parameter, inject 20 µl of each of the sample solution separately. Calculate the content of Linagliptin in each tablet by using following formula:

Linagliptin per tablet:

$$=\frac{Spl\ Peak\ Area}{Std\ Peak\ Area} \times \frac{conc\ of\ std}{conc\ of\ spl} \times std\ potency\ \% \times \frac{100-LOD/WC}{100}\ x\ Average\ weight$$