ANALYTICAL METHOD VALIDATION COMMITTEE FOR NON PHARMACOPOEIAL PRODUCT
DEPARTMENT OF DRUG ADMINISTRATION

Analytical profile of Daclatasvir Tablets

Analytical Profile No.: DAC075/076/AP040
Daclatasvir tablet contains 90% to 110% of Daclatasvir of stated amount.

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 Daclatasvir.

2. Dissolution: Determine by liquid chromatography

2.1 Dissolution Parameters:

Medium:	1000 ml ofPhosphate buffer pH 6.8 with 0.75% brij 35
Apparatus:	Paddle
Speed and time:	75rpm, 45 minutes
Volume:	1000 ml

2.2 Chromatographic system:same as in Assay

2.3 Test Solution:

Withdraw a suitable volume of the sample after 45 minutes. Dilute 1ml of this solution to 20ml with methanol. Filter the final solution through 0.2 µm membrane filter.

2.4 Standard Solution:

Weigh accurately about 33.0 mg Daclatasvir dihydrochloride WS in 100 ml volumetric flask. Add about 70 ml of dissolution medium, sonicate to dissolve and make up the volume to 100 ml with dissolution medium. Dilute 5ml of resulting solution to 50ml with dissolution medium. Further dilute 2 ml of this solution to 20 ml with methanol. Filter the final solution through 0.2 µm membrane filter.

2.5Procedure:

Inject the reference solution five/six times and sample solutions. 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 for replicate injections isnot more than 2.0%. Measure the peak responses. Calculate the % release of Daclatasvir per tablet.

2.6 Limit:

D. NLT 80% of the stated amount.

3. Assay: Determine by liquid chromatography.

3.1Chromatographic system:

Column:	C18, 250*4.6 mm, 5 µm
Injection volume:	5 µl
Flow rate:	1.0 ml/min
Column Temperature:	25°C
Detector:	UV 315nm
Mobile Phase:	Methanol: Water (98:02)

3.2 Test Solution:

Weigh individually 20 tablets & crush the tablet into fine powder.Weigh accurately the powder eq. to 30 mg of Daclatasvir in 100ml volumetric flask, add 70 ml of methanol, sonicate to dissolve with intermittent shaking at 20°C to 25°C temperature and make volume to 100 ml with same solvent.Stir for 10 minutes. Dilute 5ml of resulting solution to 50ml with same solvent. Further dilute 10ml of this solution to 25ml with same solvent. Filter the final solution through 0.2 µm membrane filter.

3.3Standard Solution:

Weigh accurately about 33.0 mg Daclatasvir dihydrochloride WSin 100 ml volumetric flask. Add about 70 ml of methanol and sonicate for about 5 minutes and make up the volume to 100 ml with same solvent. Dilute 5ml of resulting solution to 50ml with methanol.Further dilute 10ml of this solution to 25ml with methanol. Filter the final solution through 0.2 µm membrane filter.

3.4Procedure:

Inject the reference solution five/six times and sample solutions. 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 for replicate injections isnot more than 2.0%. Measure the peak responses. Calculate the content of Daclatasvir per tablet.

3.5 Other tests: As per pharmacopoieal requirement.

NOTE: 110 mg Daclatasvir dihydrochloride eq. to 100 mg Daclatasvir