ANALYTICAL METHOD VALIDATION COMMITTEE FOR NON PHARMACOPOEIAL PRODUCT

DEPARTMENT OF DRUG ADMINISTRATION

National Medicines Laboratory

Clobazam Mouth Dissolving Tablet

Analytical Profile No.: CLOB 075/076/AP041

Clobazam Mouth Dissolving Tablet contains not less than 90% and not more than 110% of clobazam of stated amount.

1. Identification:

The retention time of the major peak in the chromatogram of sample solution corresponds to that of the standard solution as obtained in assay.

2. Dissolution: Determine by thin layer chromatography.

2.1 Dissolution Parameters:

Apparatus: Paddle

Medium: 900ml of 0.1M Hydrochloric Acid

Speed and Time: **75rpm for 30 minutes**

Temperature: **37˚C ± 0.5˚C**

2.2 Chromatographic Condition: same as Assay

2.3 Test Preparation:

Withdraw a suitable volume of medium and filter. Dilute 10 ml of the filtrate to 20ml with dissolution medium. Filter it through 0.2 micron membrane filter.

2.4 Reference Preparation:

Weigh accurately about 55.5 mg of Clobazam WS into 100 ml volumetric flask. Add 45ml of Acetonitrile, sonicate to dissolve it then make up the volume with water. Dilute 1ml of resulting solution to 100ml with dissolution medium to obtain 0.00555mg/ml concentration solution. Filter it through 0.2 micron membrane filter.

2.5 Procedure: Proceed the process as described in assay method and calculate the percent release of Clobazam.

2.6 Limit:

D. NLT 80%

3. Uniformity of Content (if required):

3.1 Chromatographic Condition: same as Assay

3.2 Standard Preparation: same as Assay

3.3 Sample Preparation:

Weigh 10 tablets individually and place one tablet individually in 100 ml volumetric flask. Add 70 ml of mobile phase & sonicate for 15 minutes to dissolve. After sonication, dilute to 100 ml with same solvent. Dilute 10 ml of resulting solution to 20ml with mobile phase to obtain 0.05mg/ml concentration solution. Filter it through 0.2 micron membrane filter.

3.4 Procedure: Proceed the process as described in assay method and calculate uniformity of content.

3.5 Limit:

85-115% of stated amount

4. Assay: Determine by liquid chromatography

4.1 Chromatographic Condition:

Column: C18 (15 cm X 4.6 mm), 5µm

Flow rate: 1.0 ml/min

Injection volume: 20 µl

Wavelength: 230nm

Detector: UV

Column Temperature: 35˚ C

Mobile Phase: Acetonitrile:Water(45:55)

4.2 Test Preparation:

Weigh individually 20 tablets & crush the tablet into fine powder. Weigh accurately the powder equivalent to 5 mg of Clobazam into 100 ml volumetric flask, add 70 ml of mobile phase & sonicate for 15 minutes to dissolve. After sonication, dilute to 100 ml with same solvent. Filter the solution through 0.2 µm membrane filter.

4.3 Standard Preparation:

Weigh accurately about 50 mg of Clobazam WS into 100 ml volumetric flask. Dissolve with 70ml of mobile phase and make up the volume up to the mark with same solvent. Dilute 2ml of resulting solution to 20ml with mobile phase to obtain 0.05mg/ml concentration solution. Filter it through 0.2 micron membrane filter.

4.4 Procedure:

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 in not more than 2.0%.

Calculate the content of Clobazam per tablet.

4.5 Other tests: As per pharmacopoieal requirement.