ANALYTICAL METHOD VALIDATION FOR MIRTAZAPINE TABLETS USP

ANALYTICAL METHOD VALIDATION REPORT FOR MIRTAZAPINE TABLETS USP

Validation Analysis Records for Mirtazapine Tablets USP

OBJECTIVE: The efficacy & safety of a medicinal product can only be assured by analytical monitoring of its quality.

SCOPE: The scope of analytical validation is to ensure that the procedure under consideration is capable of giving reproducible and reliable results.

Product Name	Mirtazapine Tablets USP
Ingredient	Mirtazapine USP
Label Claim	Each film coated tablet contains
	Mirtazapine USP ----------7.5/15/30mg
Test Method	Liquid Chromatography
	Mirtazapine USP

Specificity (**Diluents Interference**)

Placebo Preparation:

A placebo solution was prepared same as the formulation except for the addition of the active ingredients. Here used as the placebo solution. Area at 290 nm, Observation Result: Nil

Conclusion for Specificity: We observed that at wavelength 290 nm there is no significant area for placebo (Diluents) for Mirtazapine tablets assay method. Therefore specificity of the method considered acceptable.

System Accuracy:

The system precision of the above method was carried out by taking area for six times of the sample preparation of exact weight.

Test data collection sheet:

Serial No.	Area of Mirtazapine
1.	
2.	
3.	
4.	
5.	
6.	
Mean	
% RSD	

Acceptance Criteria: RSD is not more than 2.0%.

Linearity/ Accuracy:

Definition:

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The Linearity of an analytical method is its ability to elicit test results that are directly, or by a well defined mathematical transformation, proportional to the concentration of the analyte in samples within a given range. Linearity is usually expressed in terms of the variance around the slope of the regression line calculated according to an established mathematical relationship from test results obtained by the analysis of sample with varying concentration of analyte.

Range:

Definition:

The Range of an analytical method is the interval between the upper & lower level of analyte that have been demonstrated with precision, accuracy & linearity using the method as written. The Range is normally expressed in same units as test results e.g. Percent or Parts per million, obtained by the analytical method.

Assay: Mirtazapine Tablet USP (Limit: 90.0 % to 110.0 % of the labeled amount).

Condition:

Column: Packing L1, 5µm (25 cm X 4.6 mm)

Column temperature: 40°C

Detector: UV 290nm

Flow rate: 1.5ml/min.

Injection volume: 10µl

Buffer Solution: Transfer about 18.0 g of Tetra methyl ammonium hydroxide pentahydrate to a 1000 mL volumetric flask and dissolve in about 750 mL of water. While stirring adjust with Phosphoric acid to pH of 7.4, dilute with water to volume and mix.

Diluents: A mixture of Acetonitrile and water (50: 50)

Mobile Phase: Prepare a filtered and degassed mixture of buffer solution, Acetonitrile, methanol and Tetrahyrofuran (65 : 15 : 12.5 : 7.5).

Standard Preparation:

Weigh accurately----------mg (30mg) of Mirtazapine WS in 100ml volumetric flask, add 20ml diluents and sonicate to completely dissolve and dilute to make up with diluents.

Sample Preparation:

Weigh and powdered of 20 tablets. Transfer a required quantity of the tablets powder in 100ml volumetric flask, add 20ml diluents and sonicate to completely dissolve and dilute to make up with diluents and shake.

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Chromatographic System:

Chromatograph the standard preparation, and record the peak response as directed for procedure: the column efficiency is not less than 7000 theoretical plates; the tailing factor is not more than 2.0; and the relative standard deviation for replicate injections is NMT 1.5 %.

Procedure:

Separately inject equal volumes (about 10 micro liters) of the standard preparation and the assay preparation into the chromatograph, record the chromatograms, and measure the response for the major peaks. Calculate the quantity, in % of Mirtazapine ($C_{17}H_{19}N_3$) in the portion of powder taken by the formula:

$$\frac{\text{Sample area} \times \text{WS Weight} \times \text{Potency of WS} \times \text{Average Weight} \times 100}{\text{Standard area} \times \text{Sample weight} \times 100 \times \text{Claim}} = \quad \%$$

Text data collection sheet:

S.No.	Standards	Area of Mirtazapine
1.	Standard-1	
2.	Standard-2	
3.	Standard-3	
4.	Standard-4	
5.	Standard-5	
6.	Standard-6	
7.	Mean	
8.	% RSD	

Samples	Sample Area Mirtazapine	Mean
Sample-A-01 80%		
Sample-A-02 80%		
Sample-A-03 80%		
Sample-B-01 90%		
Sample-B-02 90%		
Sample-B-03 90%		
Sample-C-01 100%		
Sample-C-02 100%		
Sample-C-03 100%		
Sample-D-01 110%		
Sample-D-02 110 %		
Sample-D-03 110%		
Sample-E-01 120 %		
Sample-E-02 120 %		
Sample-E-03 120 %		

Calculation:

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Data Collection:

Concentration (µg/ml)	Concentration in %	Corr. Coefficient	Sample Mean Area	% Recovery	Corr. Coefficient
	80	1.0			
	90				
	100				
	110				
	120				

Precision:

The precision of an analytical method is the degree of agreement among individual test results when the method is applied repeatedly to multiple sampling of a homogeneous sample. The precision of the analytical method is usually expressed as Standard deviation or relative standard deviation (coefficient of variation) of a series measurement. The precision may be measured of either the degree of reproducibility or of repeatability of the analytical method on the normal operating condition.

Precision: – Method precision

Mirtazapine Tablet USP (Limit: 90.0 % to 110.0 % of the labeled amount).

Analyst (I):

Condition:

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Detector: UV 290nm

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Injection volume: 10µl

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Diluents: A mixture of Acetonitrile and water (50 : 50)

Mobile Phase: Prepare a filtered and degassed mixture of buffer solution, Acetonitrile, methanol and Tetrahyrofuran (65 : 15 : 12.5 : 7.5).

Standard Preparation:

Weigh accurately----------mg (30 mg) of Mirtazapine WS in 100 ml volumetric flask, add 20 ml diluents and sonicate to completely dissolve and dilute to make up with diluents.

Sample Preparation:

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Weigh and powdered of 20 tablets. Transfer a required quantity of the tablets powder containing 30mg of Mirtazapine in 100ml volumetric flask, add 20ml diluents and sonicate to completely dissolve and dilute to make up with diluents and shake.

Chromatographic System:

Chromatograph the standard preparation, and record the peak response as directed for procedure: the column efficiency, the tailing factor is not more than 2.0; and the relative standard deviation for replicate injections is NMT 1.5 %.

Procedure:

Separately inject equal volumes (about 10 micro liters) of the standard preparation and the assay preparation into the chromatograph, record the chromatograms, and measure the response for the major peaks. Calculate the quantity, in % of Mirtazapine ($C_{17}H_{19}N_3$) in the portion of powder taken by the formula :

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Sample Dilutions:

(A) Take ----------mg of the sample and proceed as above.

(B) Take ----------mg of the sample and proceed as above.

(C) Take ----------mg of the sample and proceed as above.

(D) Take ----------mg of the sample and proceed as above.

(E) Take ----------mg of the sample and proceed as above.

(F) Take ----------mg of the sample and proceed as above.

Test Data Collection sheet:

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S.No.	Standards	Area of Mirtazapine
1.	Standard 1	
2.	Standard 2	
3.	Standard 3	
4.	Standard 4	
5.	Standard 5	
6.	Standard 6	
7.	Mean	
8.	% RSD	

Samples		Area of Mirtazapine	Mean
Sample A	T1		
	T2		
Sample B	T1		
	T2		
Sample C	T1		
	T2		
Sample D	T1		
	T2		
Sample E	T1		
	T2		
Sample F	T1		
	T2		

Calculation:

Table for Six Replicate Assays:

Sample Number	Estimated % Amount	Mean	Relative Standard Deviation (% RSD)
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Acceptance Criteria: NMT 2% (% of Relative Standard Deviation).

Intermediate Precision: – (Within laboratory variations such as different days, analyst & equipments):

Mirtazapine Tablet USP (Limit: 90.0 % to 110.0 % of the labeled amount).

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Condition:
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Column temperature: 40°C

Detector: UV 290 nm

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Flow rate : 1.5ml/min.

Injection volume : 10µl

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Diluents: A mixture of Acetonitrile and water (50 : 50).

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Weigh accurately----------mg (30mg) of Mirtazapine WS in 100ml volumetric flask, add 20ml diluents and sonicate to completely dissolve and dilute to make up with diluents.

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Chromatographic System:
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Procedure:
Separately inject equal volumes (about 10 micro liters) of the standard preparation and the assay preparation into the chromatograph, record the chromatograms, and measure the response for the major peaks. Calculate the quantity, in % of Mirtazapine ($C_{17}H_{19}N_3$) in the portion of powder taken by the formula:

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Sample Dilutions:
(A) Take ----------mg of the sample and proceed as above.

(B) Take ----------mg of the sample and proceed as above.

(C) Take ----------mg of the sample and proceed as above.

(D) Take ----------mg of the sample and proceed as above.

(E) Take ----------mg of the sample and proceed as above.

(F) Take ----------mg of the sample and proceed as above.

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Test Data Collection sheet:

S.No.	Standards	Area of Mirtazapine
1.	Standard 1	
2.	Standard 2	
3.	Standard 3	
4.	Standard 4	
5.	Standard 5	
6.	Standard 6	
7.	Mean	
8.	% RSD	

Samples		Area of Mirtazapine	Mean
Sample A	T1		
	T2		
Sample B	T1		
	T2		
Sample C	T1		
	T2		
Sample D	T1		
	T2		
Sample E	T1		
	T2		
Sample F	T1		
	T2		

Calculation:

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Table for Six Replicate Assays:

Sample Number	Estimated % Amount	Mean	Relative Standard Deviation (% RSD)
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Acceptance Criteria: NMT 2% (% of Relative Standard Deviation).

Table for Six Replicate Assays analyst by two different Analysts:

Test Data analyst -

Sample Number	Estimated % Amount	Mean	Relative Standard Deviation (% RSD)
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Test Data analyst:

Sample Number	Estimated % Amount	Mean	Relative Standard Deviation (% RSD)
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Acceptance Criteria: NMT 2 % (% of Relative Standard Deviation).

Robustness:

To demonstrate the analytical method is capable to yield reproducibility results under; small but deliberate variations in method parameters during normal usage such as composition & Flow rate of mobile phase.

Procedure:

Perform the robustness study by injecting single of resolution solution & standard solution for six times for the following parameters.

- Change in ratio of the mobile phase. Record the observation in below observation table.
- Change in Flow rate of mobile phase. Record the observation in below observation table.

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OBSERVATION TABLE:

Change in flow rate at 290 nm

Mobile phase				Flow rate	System suitability		
Buffer	**Acetonitrile**	**Methanol**	**Tetrahyrofuran**		**Retention Time**	**Theoretical Plates**	**Tailing Factor**
640ml	155ml	130ml	75ml	1.5ml/min			
650ml	150ml	125ml	75ml	1.5ml/min			
660ml	145ml	120ml	75ml	1.5ml/min			

Change in flow rate at 290 nm

Mobile phase				Flow rate	System Suitability		
Buffer	**Acetonitrile**	**Methanol**	**Tetrahyrofuran**		**Retention time**	**Theoretical Plates**	**Tailing Factor**
650ml	150ml	125ml	75ml	1.4ml/min			
650ml	150ml	125ml	75ml	1.5ml/min			
650ml	150ml	125ml	75ml	1.6ml/min			

Acceptance criteria:

Analytical method validation shall be robust (i.e. Theoretical Plates is not less than 2000 & tailing factor is not more than 2.0).

Analysed By/On: **Checked By/On:**