ANALYTICAL METHOD VALIDATION REPORTS FOR CIPROFLOXACIN TABLETS BP 500 mg

ANALYTICAL METHOD VALIDATION/VERIFICATION PROTOCOL FOR CIPROFLOXACIN TABLETS USP 500 MG

Product Name	Ciprofloxacin Tablets 500 mg
Ingredient	Ciprofloxacin Hydrochloride
Label Claim	Each film coated tablet contains Ciprofloxacin Hydrochloride BP Eq. to Ciprofloxacin ------500 mg
Test Method	By Liquid Chromatography

Specificity (Diluents Interference):

Placebo Preparation:

A placebo solution was prepared same as the formulation except for the addition of the active ingredients. Here used the placebo solution area at 278 nm, Observation Result: Nil

Conclusion for Specificity:

We observed that at wavelength 278 nm there is no significant area for placebo (Diluents) for Ciprofloxacin 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.

Text data collection sheet:

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

Acceptance Criteria: RSD is not more than 2.0%**.**

Linearity/ Accuracy:

Definition:

The Linearity of an analytical method is its ability to elicit test results that are directly, or by a well

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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: (Limit: 90.0 % to 110.0 % of the labeled amount).
Chromatographic Condition:-

- a stainless steel column 25 cm X 4.6 mm, packed with octadecylesilane bonded to porous silica (5um),
- Flow rate 1.5ml per minute,
- spectrophotometer set at 278 nm,
- Injection volume 10 µl.
- Column temperature of 40°.

Mobile Phase: A mixture of 13 volumes of Acetonitrile and 87 volumes of a 0.245% w/v solution of Orthophosphoric acid the pH of which has been adjusted to 3.0 with triethylamine. Then degas and filter through with 0.45 µm filter paper.

Standard preparation:

Weigh accurately----------mg (about 116 mg) of Ciprofloxacin Hydrochloride working standard in 100ml volumetric flask add 50 ml of mobile phase and sonicate to completely dissolve, shake and makeup 100 ml with mobile phase & filter. Take 5.0ml and dilute to 25 ml with mobile phase & shake.

Sample Preparation:

Weigh accurately as required quantities of the sample powdered tablets in 100ml volumetric flask add 50 ml of mobile phase and sonicate to completely dissolve, shake and makeup 100 ml with mobile phase & filter. Take 5.0 ml and dilute to 25 ml with mobile phase & shake.

Chromatographic system:

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Inject the standard and sample solutions. If the tailing factor more than 2.0 and column efficiency is less than 2000 theoretical plates the test is not valid. The relative standard deviation for replicate injection is not more than 2.0 %. Inject the test solution and reference solution.

Test data collection sheet:

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

Acceptance Criteria: RSD is not more than 2.0%

Samples	Sample Area	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-01 120%		
Sample-E-01 120%		

Calculation:

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Data Collection:-

Concentration (µg/ml)	Concentration in %	Corr. Coefficient	Sample mean area	% Recover	Corr. Coefficient
	80.0	1.0			
	90.0				
	100.0				
	110.0				
	120.0				

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:

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Sample Dilutions: By …….

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

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

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

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

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

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

Test data collection sheet:

S.No.	Standards	Area
1.	Standard-1	
2.	Standard-2	
3.	Standard-3	
4.	Standard-4	
5.	Standard-5	
6.	Standard-6	

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7.	Mean	
8.	%RSD	

Acceptance Criteria: RSD is not more than 2.0%

Test Data Collection:-

Samples		Sample Area	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	% RSD
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Intermediate Precision –
(Within laboratory variations such as different days, analyst & equipments):
Analyst (II) Anurag Singh:-

Chromatographic Condition:-

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

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

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

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

Test data collection sheet:

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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	% RSD
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Table for Six Replicate Assays analyst by two different Analysts & days:

Test Data analyst by **Bhoopendra Singh-**

Table for Six Replicate Assays

Sample Number	Estimated % Amount	Mean	% RSD
Sample A			
Sample B			
Sample C			
Sample D			
Sample E			
Sample F			

Test Data analyst by **Anurag Singh-**

Table for Six Replicate Assays

Sample Number	Estimated % Amount	Mean	% RSD

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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.

Observation Table:-

Change ratio in the mobile phase at 278 nm

Mobile phase		Flow rate ml/min	System suitability		
Buffer	**Acetonitrile**		**Retention Time**	**Theoretical Plates**	**Tailing Factor**
875 ml	125 ml	1.5 ml/min			
870 ml	130 ml	1.5 ml/min			
865 ml	135 ml	1.5 ml/min			

Change in flow rate at 278 nm

Mobile phase		Change in Flow rate	System Suitability		
Buffer	**Acetonitrile**		**Retention time**	**Theoretical Plates**	**Tailing Factor**
870 ml	130 ml	1.4 ml/min			
870 ml	130 ml	1.5 ml/min			
870 ml	130 ml	1.6 ml/min			

Acceptance criteria:

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Analytical method validation shall be robust (i.e. Theoretical Plates is not less than 1500 & tailing factor is not more than 2.0).

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