UV SPECTROPHOTOMETRIC ANALYSIS AND VALIDATION OF ACYCLOVIR IN SOLID DOSAGE FORM

AKSHATA LASURE1*, AFAQUE ANSARI1, MALLINATH KALSHETTI1
D. S. T. S. Mandal’s College of Pharmacy, Solapur 413004 Maharashtra, India
Email: lasureakshata@gmail.com
Received: 20 Nov 2019, Revised and Accepted: 19 Jan 2020

ABSTRACT
Objective: A new, economical, sensitive, simple, rapid UV spectrophotometric method has been developed for the estimation of Acyclovir in pure form and pharmaceutical formulation.

Methods: This UV method was developed using distilled water as a solvent. In the present method, the wavelength selected for analysis was 254 nm. UV-Visible double beam spectrophotometer (Systronic 2201) was used to carry out the spectral analysis. The ICH guidelines were used to validate the method.

Results: The method was validated for linearity, range, accuracy, precision, robustness, LOD and LOQ. Linearity was found in the range of 5-30µg/ml. Accuracy was performed by using a recovery study. The amount of drug recovered was found to be in the range of 100.1-100.5 %. The % RSD value was found to be less than 2.

Conclusion: The proposed UV spectroscopic method was found to be accurate, precise, stable, linear, specific, and simple for quantitative estimation of acyclovir in bulk and pharmaceutical dosage form. Hence the present UV spectroscopic method is suitable for the routine assay of acyclovir in bulk and pharmaceutical formulations.

Keywords: Acyclovir, UV-Visible spectrophotometric method, Method validation

INTRODUCTION
One of the most frequently employed techniques in the pharmaceutical analysis is UV-Visible spectrophotometry. The amount of ultraviolet or visible radiation absorbed by a substance in a solution is measured by UV spectrophotometer [1].

Ultrasonicator (micro clean-103) was used to sonicate the formulation sample. Drug sample was weighed by using an electronic analytical balance (Shimadzu AY220).

Chemicals and reagents
Active pharmaceutical ingredient of Acyclovir is gifted as a sample from Aadhaar Life Science s Pvt. Ltd. Solapur. Marketed formulation of Acyclovir was procured from the local pharmacy.

Experimental work
Method development
Preparation of standard stock solution of acyclovir
10 mg of standard drug Acyclovir was accurately weighed and transferred into 10 ml volumetric flask and a sufficient amount of water was added into it and sonicated for 5 min, finally, volume was made up to the mark with the same solvent to make 1000µg/ml stock solution. From this 1 ml was again diluted to 10 ml to get a concentration of 100µg/ml of Acyclovir. From 100µg/ml solution 5 ml was again diluted to 10 ml to get a concentration of 50µg/ml.

Selection of wavelength
To determine the wavelength for measurement, Acyclovir (50µg/ml) solution was scanned in the range of 200-400 nm against distilled water as blank. Wavelength of maximum absorption was determined for the drug. Acyclovir showed maximum absorption at 254 nm.

Assay of acyclovir tablet
20 tablets weighed and powdered. The powder equivalent to 10 mg of acyclovir was weighed, transferred into 10 ml volumetric flask and dissolved in water. This solution was sonicated for 15 min and the final volume was made up to the mark with water. 1 ml of solution was transferred into 10 ml volumetric flask and diluted up to 10 ml with water. The absorbance of this solution was measured at 254 nm.
RESULTS AND DISCUSSION

Method validation

The method was validated for several parameters like Linearity, Accuracy, Precision, Robustness, Limit of Detection (LOD), Limit of Quantification (LOQ) and Specificity of Acyclovir tablet [6-9].

Linearity and range

The linear relation between absorbance and concentration of drug was evaluated using three replicates over concentration range 5-30 µg/ml by making the replicates (Table 1 and Fig. 3).

The wavelength for linearity was scanned at 254 nm. By taking six different concentrations for linearity the regression coefficient was found to be 0.997 i.e. in the limit of standard. Hence the linearity parameter was found to be validated.

Accuracy

Accuracy of the method was confirmed by recovery studies from marketed formulation at three different levels of standard i.e. 50%, 100%, 150% was done to confirm the accuracy of the developed method. The amount of acyclovir is calculated at each level and percentage recoveries were calculated (Table 2).

Precision

Precision of the developed method expressed in terms of the relative standard deviation of the absorbance. The solution was analyzed in 6 replicates for intra-day precision and in two successive days for inter-day precision. The % RSD value was found to be less than 2. Results confirmed that the precision of the method was found to be accepted. Precision results were given in Table 3 and Table 4 for intra and inter-day precision respectively.

Table 1: Results of linearity

| S. No. | Concentration (µg/ml) | Absorbance |
|--------|-----------------------|------------|
| 1      | 5                     | 0.291      |
| 2      | 10                    | 0.537      |
| 3      | 15                    | 0.764      |
| 4      | 20                    | 1.014      |
| 5      | 25                    | 1.214      |
| 6      | 30                    | 1.451      |

Table 2: Results of accuracy

| Name of drug | Recovery levels | Concentration (µg/ml) | Amount recovered | % Recovery with SD |
|--------------|-----------------|-----------------------|-----------------|--------------------|
| Acyclovir    | 50 %            | 10                    | 10.001          | 100.01±0.70        |
|              | 100 %           | 20                    | 20.001          | 100.03±0.13        |
|              | 150 %           | 30                    | 30.004          | 100.05±0.25        |
Table 3: Results for intra-day precision

| S. No. | Concentration (µg/ml) | Absorbance |
|--------|-----------------------|------------|
| 1      | 10                    | 0.538      |
| 2      | 10                    | 0.539      |
| 3      | 10                    | 0.537      |
| 4      | 10                    | 0.539      |
| 5      | 10                    | 0.537      |
| 6      | 10                    | 0.539      |
| SD     |                       | 0.000983   |
| %RSD   |                       | 0.182693%  |

Table 4: Results for inter-day precision

| S. No. | Concentration (µg/ml) | Absorbance (Day1) | Absorbance (Day2) |
|--------|-----------------------|-------------------|-------------------|
| 1      | 10                    | 0.538             | 0.537             |
| 2      | 10                    | 0.539             | 0.538             |
| 3      | 10                    | 0.537             | 0.539             |
| 4      | 10                    | 0.539             | 0.538             |
| 5      | 10                    | 0.537             | 0.537             |
| 6      | 10                    | 0.539             | 0.539             |
| SD     |                       | 0.000983          | 0.000894          |
| %RSD   |                       | 0.182693%         | 0.16625%          |

For intra-day and the inter-day precision relative standard deviation is in limit i.e. less than 2% hence parameter is validated.

Table 5: Results for robustness

| Wavelength | Concentration (µg/ml) | Absorbance (254 nm) | Absorbance (260 nm) |
|------------|-----------------------|---------------------|---------------------|
|            | 12µg/ml               | 0.612               | 0.613               |
|            | 12µg/ml               | 0.613               | 0.612               |
|            | 12µg/ml               | 0.611               | 0.611               |
|            | 12µg/ml               | 0.613               | 0.612               |
|            | 12µg/ml               | 0.612               | 0.611               |
|            | 12µg/ml               | 0.614               | 0.613               |
| Average    |                       | 0.613               | 0.612               |
| SD         |                       | 0.0011              | 0.00089            |
| % RSD      |                       | 0.179445            | 0.145425           |

By change in concentration and wavelengths i.e. 254 nm and 260 nm % RSD is less than 2% i.e. within the range. So parameter was validated.

Table 6: Results for ruggedness

| Concentration (µg/ml) | Analyst 1 | Analyst 2 |
|-----------------------|-----------|-----------|
| 15                    | 0.764     | 0.765     |
| 15                    | 0.762     | 0.764     |
| 15                    | 0.765     | 0.766     |
| 15                    | 0.764     | 0.763     |
| 15                    | 0.766     | 0.765     |
| 15                    | 0.765     | 0.762     |

By change in analyst and laboratory, there is no effect on absorbance with the same conditions (table 6). Hence, the parameter was validated.

Robustness

Robustness is a measure of its capacity to remain unaffected by small, but deliberate variations in method parameters and provides an indication of its reliability during normal usage. Robustness was carried out on two different instruments and also carried out by using two different analysts (table 5).

Ruggedness

The degree of reproducibility of test results of the same sample within different laboratories and different analysts under the same condition with the same concentration.

Limit of detection (LOD)

Limit of detection of an individual analytical procedure is the lowest amount of analyte in the sample which can be detected but not necessarily quantitated as an exact value. LOD was found to be 0.862.

Limit of quantitation (LOQ)

Limit of quantitation of an individual analytical procedure is the lowest amount of an analyte in the sample which can be quantified as an exact value. LOQ was found to be 2.612.

CONCLUSION

The proposed UV spectroscopic method is found to be accurate, precise, stable, linear, specific, and simple for quantitative estimation of acyclovir in the bulk and pharmaceutical dosage form. Hence the present UV spectroscopic method is suitable for the routine assay of acyclovir in bulk and pharmaceutical formulations.

ACKNOWLEDGMENT

The authors are thankful to the principal and the management, DSTS Manda's College of Pharmacy Solapur, for providing the necessary facilities for research.
FUNDING
Nil

AUTHORS CONTRIBUTIONS
All the authors have contributed equally.

CONFLICT OF INTERESTS
Declare none

REFERENCES
1. Padala NR, Baishakhi DA, Assaleh FH, Katakam PA, Rao BC. Uv-spectrophotometric estimation of acyclovir in bulk and pharmaceutical dosage forms. J Pharm Sci Innovation 2013;2:40-3.
2. Sadjadi SA, Regmi S, Chau T. Acyclovir neurotoxicity in a peritoneal dialysis patient: report of a case and review of the pharmacokinetics of acyclovir. Am J Case Rep 2018;19:1459-62.
3. Gandhi PK, Momin NS, Kharade SP, Konapure NP, Kuchekar BS. Estimation of acyclovir in the spectrophotometric estimation of acyclovir in pharmaceutical dosage forms pharmaceutical dosage forms. Indian J Pharm Sci 2006;68:516-7.
4. Muralidharan SA, Kabimani JP, Parasaruman SR, Sokkalingam AD. Development and validation of acyclovir HPLC external standard method in human plasma. Appl Pharmacokinetic Studies Adv Pharm 2014;33:1-5.
5. Chaudhari SA, Mannan AJ, Daswadkar SP. Development and validation of UV spectrophotometric method for simultaneous estimation of acyclovir and silymarin in niosome formulation. Der Pharmacia Lett 2016;8:126-33.
6. Ukpe AS, Johnson O. Spectrophotometric determination of acyclovir after its reaction with ninhydrin and ascorbic acid. J Appl Pharm Sci 2015;5:65-9.
7. Zendelovska D. Determination of acyclovir in human plasma samples by HPLC method with UV detection: application to a single-dose pharmacokinetic study. J Med Sci 2015;3:32-6.
8. Velivela SM, Konde AP, Mayasa AP, Nikunja BP. Method development and validation of acyclovir in rabbit plasma by RP-HPLC. J Pharm Res 2016;10:509-13.
9. ICH, Q2 (R1) Validation of analytical procedures: text and methodology. International conference on harmonization; 1996.