# METHOD DEVELOPMENT AND VALIDATION OF ROSUVASTATIN CALCIUM BY USING UV VISIBLE SPECTROSCOPY

Vijaya Durga Dumpala*, Datla Nishitha, Teluri Chinmayi Reddy,

Boosa Raja Shekar.

Department of Pharmaceutical Analysis,

Gokaraju Rangaraju College of Pharmacy, Hyderbad, Telangana, India.

## Abstract

The aim of study is to develop a new , rapid and highly sensitive UV Visible spectrophotometer method for development and validation for the estimation of Rosuvastatin calium.Chloroform was selected as the suitable solvent based on solubility studies.The lmax was found to be 243 nm.Linearity was established over a concentration range of 2-10µg/ml with a correlation coeffeicient of 0.999. Accuracy studies was showed a mean recovery of 100.19% with% RSD of 0.39% Precision, robustness and ruggedness were confirmed with %RSD values below 2%. The method demonstrated high sensitivity with LOD, LOQ values of 0.037 µg/ml and 0.116 µg/ml, respectively. Thus the method is validated as per ICH guidelines and is suitable for routine analysis of Rosuvastatin in Bulk and tablet dosage forms.

## Introduction

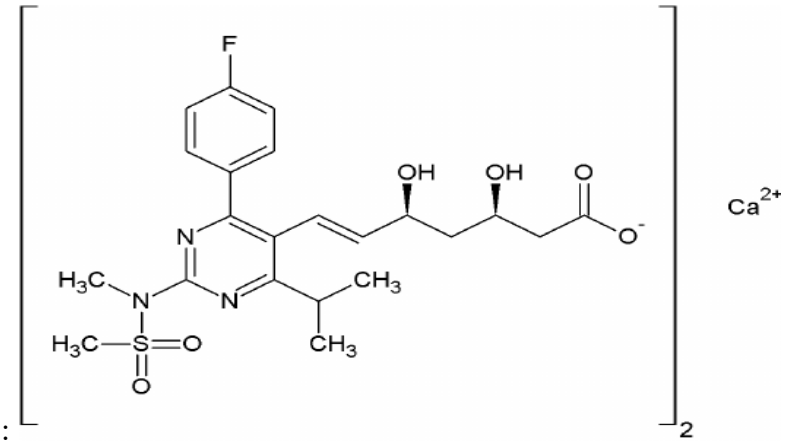
## Spectroscopy

It is a Branch of Science that deals with the study of the interaction of matter with light or Electromagnetic Radiation. It is the measurement and interpretation of Electromagnetic radiation absorbed or emitted when the molecules or atoms or ions of sample move from one energy state to another.Ultraviolet (UV) and visible radiation comprise only a small part of the electromagnetic spectrum, which includes such other forms of radiation as radio, infrared (IR), cosmic, and X rays.

## DRUG PROFILE

## Rosuvastatin Calcium

**Description:**
Rosuvastatin Calcium is a lipid-lowering agent that belongs to the statin class. It works by inhibiting HMG-CoA reductase, the enzyme responsible for cholesterol synthesis in the liver. It is used to treat high cholesterol and related conditions and to prevent cardiovascular disease.




**Chemical Structure of Rosuvastatin Calcium**

**Molecular Formula:** C44H54CaF2N6O12S2

**Solubility:** Practically insoluble in water, soluble in methanol and ethanol

**Melting Point:** Approx. 155°C (decomposes)

**$P^{Ka}$ Values:** 4.6 (carboxylic acid), 9.5 (hydroxyl group)

**Uses:**
Rosuvastatin is indicated for the treatment of primary hyperlipidemia and mixed dyslipidemia, including familial hypercholesterolemia (both heterozygous and homozygous types). It is also used in the prevention of cardiovascular events such as stroke and myocardial infarction in patients with elevated risk factors.

**Marketed Formulations:**

**Brand Names:** Crestor, Rosuvas, Rozavel

**Dose:** Commonly available in 5 mg, 10 mg, 20 mg tablet

## MATERIALS AND METHODS

### CHEMICALS USED:

| Chemicals Name | Supplier |
|---|---|
| Rosuvastatin calcium | Aurobindo pharma Ltd |
| Acetonitrile | Vijay enterprises |
| Chloroform | Vijay enterprises |
| Methanol | Vijay enterprises |

Table 4.1:Chemicals Name

**INSTRUMENTS USED:**

| Instrument name | Model name |
|---|---|
| Uv visible spectrophotometer | SHIMADZU (model UV-1800 |
| Analytical balance | SHIMADZU ATX224R |
| Ultra sonicator | Sonica®2200MH |

Table: 4.2 : Instruments Used

**METHOD DEVELOPMENT**

**Selection of solvent:**

Method development includes choosing an appropriate solvent for the procedure which can be the best for precise results, accuracy and cost efficient. By testing distilled water, methanol, n hexane, chloroform and acetonitrile. Drug is soluble in chloroform. Apparently chloroform is best for solubility.

**Solubility studies**:

Solubility studies help identify a **solvent or solvent system** in which the drug (like Rosuvastatin) is sufficiently soluble. This ensures **complete dissolution**, which is essential for accurate and consistent analysis.

**Selection of Wavelength**

*The absorbance of the solution containing* Rosuvastatin at 10µg/ml was determined in UV range 200-400nm using an appropriate blank. 10µg/ml was prepared by dissolving 10mg of Rosuvastatin in 100ml diluent (Stock A). 1ml was pipetted out and dissolved in 10ml diluent to obtain 10 µg/ml solution. The λmax was found to be 243nm.

**Preparation of Standard Solution**

- **Preparation of Rosuvastatin Standard Solution:** Weigh accurately about 10 mg and transfer in to a 100 ml volumetric flask. Add 30 ml of diluent and sonicate to dissolve. Dilute up to mark with diluent.) Filter through 0.45µm nylon membrane filter.
- **Preparation of Test Solution:** Determine the average weight of 10 tablets, Powdered it. Weigh accurately about 10 mg of Rosuvastatin calcium tablet powder (equivalent to 10 mg of Rosuvastatin calcium) and transfer in to a 100ml volumetric flask. Add 30 ml of diluent and sonicate to dissolve. Dilute up to mark with diluent. Filter the solution through 0.45µm nylon membrane filter.

## METHOD VALIDATION

**Specificity:**

- The specificity is established by recording the UV Spectrum from 200 nm to 400 nm for blank, Rosuvastatin shows maximum absorbance at **243.0 nm**, as confirmed by the spectrum overlay and individual spectrum data.
- standard, and Sample preparations. It is observed that there is no interference between the absorbance in the UV spectrum of Rosuvastatin shows maximum absorbance at **243.0 nm**, as confirmed by the spectrum overlay and individual spectrum data.

**Linearity**

- Linearity was determined by plotting concentration against corresponding absorbance. A standard solution was further diluted with buffer to obtain 2 µg/ml to 10 µg/ml range solutions. The calibration curves were constructed by plotting absorbance versus concentration and the regression equations were calculated.

**Accuracy:**

- The accuracy of the proposed method was assessed by recovery studies which were carried out at three different levels i.e 50%,100% and 150%. A known amount of standard drug solution was added to the pre analyzed sample solution at three different levels, was absorbance was recorded. The % recovery was then calculated.
- **Preparation of Rosuvastatin Standard Solution:** Weigh accurately about 10 mg and transfer in to a 100 ml volumetric flask. Add 30 ml of diluent and sonicate to dissolve. Dilute up to mark with diluent.) Filter through 0.45µm nylon membrane filter.
- **Preparation of Test Solution:** Determine the average weight of 10 tablets, Powdered it. Weigh accurately about 10 mg of Rosuvastatin calcium tablet powder (equivalent to 10 mg of Rosuvastatin calcium) and transfer in to a 100ml volumetric flask. Add 30 ml of diluent and sonicate to dissolve. Dilute up to mark with diluent. Filter the solution through 0.45µm nylon membrane filter.

**Precision:**

- **Intra-day precision:** standard stock solutions were taken in a 10 ml volumetric flasks and final volume was made made up to mark. The absorbances of these solutions were individually measured thrice within a day.
- **Inter day precision:** standard stock solutions were taken in 10 ml volumetric flasks and volume were made up to mark. The absorbances of these solutions were measured thrice in three days and recorded.

**Robustness:**

Small deliberate changes in the method like change in wavelength are made but there were no recognized change in the result and are within range as per ICH guidelines.

**Change in wavelength:**

**(normal experimental condition:243nm)**

Robustness conditions like wavelength minus (241nm), wavelength plus(245nm), were maintained and samples were injected in duplicate manner System sustainability parameters were not much affected and all the parameters were passed. %RSD was within the limit

**Ruggedness:**

Six test solutions of Rosuvastatin, were prepared as per the analytical method on different labs. These test solutions were analysed by a different analyst using different UV-Visible spectrophotometer. The % RSD of % drug release results of twelve test solutions (six samples from method precision and six samples from intermediate precision) was calculated.

**LOD & LOQ:**

LOD = 3.3$\sigma$ /S

LOQ = 10 /S,

Where,

σ is the standard deviation of the blank and

S is the slope of the calibration plot

**Assay:** Assay was performed with the Rosuvastatin Calcium formulation.

**Label claim**: 10.0 mg

**Results and Discussion**

**Solubility studies:**

**Table no. 5.1 solubility studies**

| **Solvent** | **Solubility** |
|---|---|
| Distilled water | Insoluble |
| N hexane | Insoluble |
| Acetonitrile | Soluble |
| Methanol | Insoluble |
| Chloroform | Soluble |

**Selection of wavelength:**

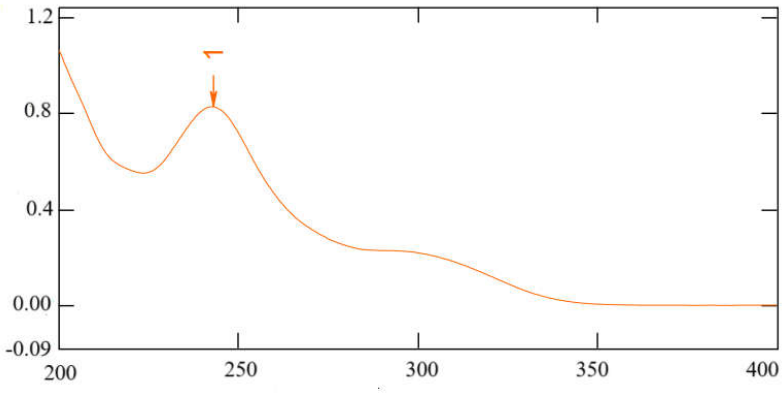



**Figure no:5.2. Selection of wavelength**

The λmax was found to be 243nm.

## METHOD VALIDATION

### 1.Specificity / Selectivity:

| **S.No.** | **Absorbance of Rosuvastatin in 243 nm** |
|---|---|
| 1 | 0.334 |
| 2 | 0.332 |
| 3 | 0.330 |
| 4 | 0.331 |
| 5 | 0.336 |
| **Mean** | **0.333** |
| **Standard Deviation (±)** | **0.002** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table 1: Selectivity results**

No absorption spectra obtained for blank preparations at 243 nm in the UV spectrum.

One Absorption spectra observed at 243 nm for Rosuvastatin standard and sample preparations.

**Remarks:**

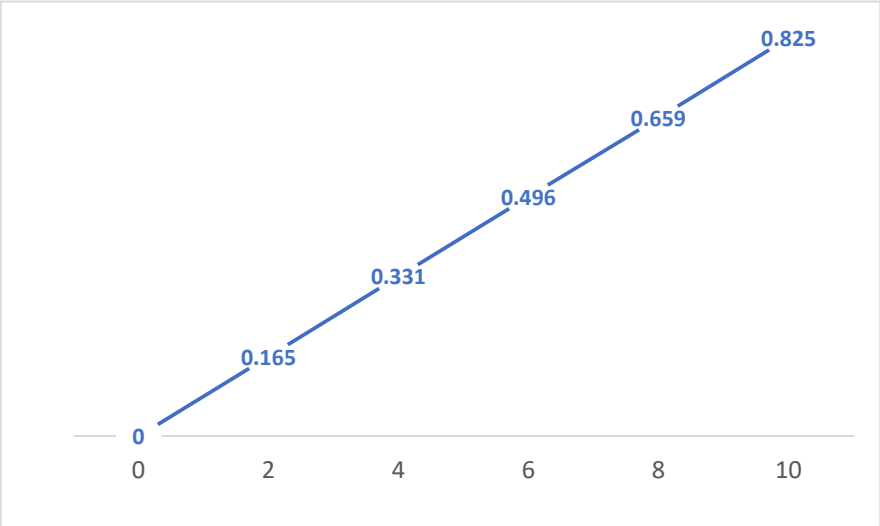
The method is specific.

### 2. Linearity

| Linearity Level | Sample | Absorbance | Correlation |
|---|---|---|---|

| | Conc (in mg/ml) | | Coefficient |
|---|---|---|---|
| Level – 1 | 0 | 0 | |
| Level – 2 | 2 | 0.165 | |
| Level – 3 | 4 | 0.331 | |
| Level – 4 | 6 | 0.496 | **0.999** |
| Level – 5 | 8 | 0.659 | |
| Level – 6 | 10 | 0.825 | |

**Table 5.2: Results of linearity of standard**




**3.Accuracy (% Recovery):**

| **Level of Addition** | **Recovery (%)** |
|---|---|
| First Level (Rec-50 %) | 100.17 |
| Third Level (Rec-100 %) | 100.59 |
| Fifth Level (Rec-150%) | 99.81 |
| **Mean** | 100.19 |
| **Standard Deviation (±)** | 0.3906 |
| **(%) Relative Standard Deviation** | 0.39 |

**Table –5. 3: Accuracy (%Recovery) – results**

**Remarks:**

The percentage recovery for Rosuvastatin at each level lies between 98.0% and 102.0%. % RSD at each recovery level is less than 2.0%.

The analytical method meets the pre-established acceptance criteria for recovery study as per protocol.

Hence, it is concluded that the method is accurate.

**4. Precision**

**Intraday Precision**

| **Sr. No.** | **Absorbance of Rosuvastatin in 243 nm** |
|---|---|
| 1 | 0.331 |
| 2 | 0.336 |
| 3 | 0.333 |
| 4 | 0.337 |
| 5 | 0.329 |
| **Mean** | **0.333** |
| **Standard Deviation (±)** | **0.003** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table.5.4 Intraday precision**

**Interday Precision**

| **Sr. No.** | **Absorbance of Rosuvastatin in 243 nm** |
|---|---|
| 1 | 0.338 |
| 2 | 0.335 |
| 3 | 0.337 |
| 4 | 0.336 |
| 5 | 0.335 |
| **Mean** | **0.336** |
| **Standard Deviation (±)** | **0.001** |
| **(%) Relative Standard Deviation** | **0.299** |

**Table – 5.5: Interday Precision**

**Remarks:** The % RSD of the twelve drug release results is found to be more than 2.0%Thus, the method is found to be precise.

**5.0 Robustness:**

| **S.No.** | **Absorbance of Rosuvastatin** | |
|---|---|---|
| **Standard** | **241nm** | **245nm** |
| 1 | 0.332 | 0.335 |
| 2 | 0.334 | 0.331 |
| **Mean** | **0.33** | **0.33** |
| **Standard Deviation (±)** | **0.00** | **0.00** |
| **(%) Relative Standard Deviation** | **0.33** | **0.65** |

**Table – 5.6: Robustness with change in wavelength**

| **Wavelength →** | **228nm** | **232nm** |
|---|---|---|
| **Sample** | **% Assay** | |
| Test solution | 99.34 | 99.57 |
| Avg % drug release result from method precision | 100.49 | 100.49 |
| **Mean** | **99.92** | **100.03** |
| **Standard Deviation (±)** | **0.81** | **0.65** |
| **(%) Relative Standard Deviation** | **0.82** | **0.65** |

**Table – 5.7: Results for change in wavelength**

Remarks: The % RSD of the six drug release results is found less than 2.0% and meets the pre-established acceptance criteria. Hence, it is concluded that the method is robust.

**6. Ruggedness**

I) Different Analysts

**Analyst – 1**

| **S.No.** | **Absorbance of Rosuvastatin in 243nm** |
|---|---|
| 1 | 0.332 |
| 2 | 0.335 |
| 3 | 0.331 |

| | |
|---|---|
| 4 | 0.336 |
| 5 | 0.330 |
| **Mean** | **0.333** |
| **Standard Deviation (±)** | **0.003** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table-5.8 Results for ruggedness by analyst-1**

**Remark:**

The % RSD of the five drug release results is found less than 2.0% and meets the pre-established acceptance criteria. Hence, it is concluded that the method is rugged.

**Analyst – 2**

| **Sr. No.** | **Absorbance of Rosuvastatin in 243 nm** |
|---|---|
| 1 | 0.332 |
| 2 | 0.335 |
| 3 | 0.331 |
| 4 | 0.336 |
| 5 | 0.330 |
| **Mean** | **0.333** |
| **Standard Deviation (±)** | **0.003** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table- 5.9 Results for ruggedness by analyst-2**

**Remark :**

The % RSD of the six drug release results is found less than 2.0% and meets the pre-established acceptance criteria. Hence, it is concluded that the method is precise.

**II) Different labs**

**Lab -I**

| **Sr. No.** | **Absorbance of Rosuvastatin at 243nm** |
|---|---|
| 1 | 0.336 |
| 2 | 0.333 |

| | |
|---|---|
| 3 | 0.334 |
| 4 | 0.338 |
| 5 | 0.334 |
| **Mean** | **0.335** |
| **Standard Deviation (±)** | **0.003** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table- 5.10 Results for ruggedness in Lab-I**

**Lab-II**

| **Sr. No.** | **Absorbance of Rosuvastatin at 243nm** |
|---|---|
| 1 | 0.332 |
| 2 | 0.335 |
| 3 | 0.331 |
| 4 | 0.336 |
| 5 | 0.330 |
| **Mean** | **0.333** |
| **Standard Deviation (±)** | **0.003** |
| **(%) Relative Standard Deviation** | **0.6** |

**Table- 5.11 Results for ruggedness in Lab-II**

**7.LOD and LOQ**

| | |
|---|---|
| **LOD Value** | **0.037** |
| **LOQ Value** | **0.116** |

**Table-5.12 Results of LOD and LOQ**

## Conclusion

The present study aimed to develop and validate a simple, accurate, precise, and robust UV-spectrophotometric method for the estimation of Rosuvastatin calcium in bulk and pharmaceutical formulations. The solubility studies revealed that Rosuvastatin is soluble in acetonitrile and chloroform, while it remains insoluble in distilled water, methanol, and n-hexane. This solubility profile guided the selection of acetonitrile as the suitable solvent for UV analysis. The wavelength of maximum absorbance (λmax) for Rosuvastatin was found to

be 243 nm, which was used throughout the method due to its sensitivity and lack of interference.

Validation of the developed method was carried out as per ICH guidelines. The method showed excellent specificity, as no absorbance was observed for blank solutions at 243 nm, confirming no interference from excipients or solvents. Linearity was established over the concentration range of 2–10 mg/mL with a correlation coefficient of 0.999, indicating a strong linear relationship between absorbance and concentration. Accuracy studies conducted through recovery tests at three levels (50%, 100%, and 150%) demonstrated recovery values between 98- 102%. and %RSD of only 0.39%, confirming that the method is highly accurate.

Robustness was demonstrated by small changes in wavelength (241 nm and 245 nm), with consistent results and %RSD values below 2%, proving the method's stability under slight variations. Ruggedness was assessed by different analysts and laboratories, and in all cases, %RSD values remained within acceptable limits, confirming that the method is reliable across different environments and operators. Furthermore, the method demonstrated good sensitivity with a low limit of detection (LOD) of 0.037 mg/mL and a limit of quantitation (LOQ) of 0.116 mg/mL.

In conclusion, the developed UV-spectrophotometric method for Rosuvastatin is simple, rapid, cost-effective, and highly reliable. It meets all the required validation criteria including specificity, linearity, accuracy, precision, robustness, ruggedness, LOD, and LOQ. Therefore, it can be confidently applied for the routine quantitative analysis of Rosuvastatin in bulk drug substances and pharmaceutical dosage forms in quality control laboratories.

**References**

Ambasana MA, Thakkar RS, Surabaya HT, Shah AK. (2010) ‘A new improved RP-HPLC method for assay of Rosuvastatin Calcium in tablets’, *Indian J Pharm Sci*, 72(5): 592–598.

Gajjar AK, Shah VD. (2010) ‘Simultaneous UV spectrophotometric estimation of Rosuvastatin and Ezetimibe in their combined dosage forms’, *Int J Pharm Pharm Sci*, 2(1): 131–138.

Gupta A, Mishra P, Shah K. (2009) ‘Simple UV spectrophotometric determination of Rosuvastatin Calcium in pure form and in pharmaceutical formulations’, *E-Journal of Chemistry*, 6(1): 89–92.

Hasumati RA, Rajput SJ, Dave JB, Patel CN. (2009) ‘Development and validation of two chromatographic stability-indicating methods for determination of Rosuvastatin in pure form and pharmaceutical preparation’, *Int J ChemTech Res*, 1(3): 677–689.

Indian Pharmacopoeia Commission. (2010) *Indian Pharmacopoeia*, Vol. III, Ghaziabad: Government of India, pp. 842–843.

Kumaraswamy G, Lalitha R, Vijayprakash K. (2016) ‘Validated spectrophotometric method for simultaneous estimation of Rosuvastatin Calcium and Aspirin in tablet dosage forms’, *Int J Pharm Chem Anal*, 3(2): 90–98.

Pandya CB, Channa Basavaraj KP, Chudasama JD, Mani TT. (2010) ‘Development and validation of RP-HPLC method for determination of Rosuvastatin Calcium in bulk and pharmaceutical dosage form’, *Int J Pharm Sci Rev Res*, 5(1): 82–86.

Patel B, Shah B, Gohil N. (2012) ‘Development and validation of spectrophotometric method for simultaneous estimation of Rosuvastatin Calcium and Aspirin in bulk and pharmaceutical dosage form’, *Int J Res Pharm Sci*, : 115–122.

Rajkondwar VV, Maini P, Vishwakarma M. (2009) ‘Characterization and method development for estimation and validation of Rosuvastatin Calcium by UV-visible spectrophotometry’, *Int J Theor Appl Sci*, 1(1): 48–53.

Ramnath Y, Lahare AN, Phuge AN, Gite AL, Jadhav AK. (2014) ‘A review on ultraviolet spectrophotometric determination of Rosuvastatin Calcium in marketed formulation’, *Int J Pure Appl Biosci*, 2(6): 169–177.

Sailaja B, Sravna K. (2016) ‘Analytical method development and validation for the estimation of Rosuvastatin Calcium in raw material and tablet formulation by UV spectrometric method’, *Saudi J Med Pharm Sci*, 2(1): 7–11.

Shyni Bernard, Mathew M, Joseph NJ. (2012) ‘Validated spectrophotometric method for the estimation of Rosuvastatin by hydrotropic solubilization’, *Asian J Pharm Health Sci*, 2(1): 273–277.

Tripathi KD. (2008) *Essentials of Medical Pharmacology*, 6th edn, New Delhi: Jaypee Brothers Medical Publishers Pvt. Ltd., pp. 612–616.

Tuljarani G, Sankar DG, Kadgapathi P, Suthakaran R, Satyanarayana B. (2010) ‘Visible spectrophotometric determination of Rosuvastatin in bulk and pharmaceutical formulations’, *Orient J Chem*, 26(2): 589–593.

Varghese SJ, Ravi TK. (2010) ‘Determination of Rosuvastatin and Ezetimibe in a combined tablet dosage form using high-performance column liquid chromatography and high-performance thin-layer chromatography’, *J AOAC Int*, 93(6): 1222–1227.

Garg S. (2014) ‘Oral novel spectrophotometric determination of Rosuvastatin Calcium in bulk and tablet dosage forms’, *Acta Velit*, 1(1): 20–24.