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Research Article

**ANALYTICAL METHOD DEVELOPMENT AND VALIDATION FOR
THE QUANTITATIVE ESTIMATION OF NIMODIPINE IN BULK AND
MARKETED PHARMACEUTICAL DOSAGE FORMS BY RP-HPLC****Kuchuri Mounika^{1*}, Vijaya Kuchana², Pasupuleti Sunitha³, G. Kalyani⁴**^{1*}Department of Pharmaceutical Analysis, Teegala Krishna Reddy College of Pharmacy, Medbowli, Meerpet (V), Balapur (M), Ranga Reddy (Dist), Hyderabad – 500097, Telangana²Department of Pharmaceutical Chemistry, Principal and Professor, Teegala Krishna Reddy College of Pharmacy, Medbowli, Meerpet (V), Balapur (M), Ranga Reddy (Dist), Hyderabad – 500097, Telangana³Department of Pharmaceutical Analysis, Associate Professor, Teegala Krishna Reddy College of Pharmacy, Medbowli, Meerpet (V), Balapur (M), Ranga Reddy (Dist), Hyderabad – 500097, Telangana⁴Department of Pharmaceutical Chemistry, Associate Professor, Teegala Krishna Reddy College of Pharmacy, Medbowli, Meerpet (V), Balapur (M), Ranga Reddy (Dist), Hyderabad – 500097, Telangana**Abstract:**

Objective: The current investigation was pointed at developing and progressively validating novel, simple, responsive and stable RP-HPLC method for the measurement of active pharmaceutical ingredient and Marketed Pharmaceutical Dosage form of Nimodipine.

Methods: A simple, selective, validated and well-defined stability that shows isocratic RP-HPLC methodology for the quantitative determination of Nimodipine. The chromatographic strategy utilized Symmetry ODS (C₁₈) RP Column, 250 mm x 4.6 mm, 5µm, using isocratic elution with a mobile phase of Phosphate Buffer (0.02M) and Acetonitrile were consists of 48:52% v/v (pH-2.80). A flow rate of 1.0 ml/min and a detector wavelength of 248 nm utilizing the UV detector were given in the instrumental settings. Validation of the proposed method was carried out according to an international conference on harmonization (ICH) guidelines.

Results: LOD and LOQ for the two active ingredients were established with respect to test concentration. The calibration charts plotted were linear with a regression coefficient of R²>0.999, means the linearity was within the limit. Recovery, specificity, linearity, accuracy, robustness, ruggedness were determined as a part of method validation and the results were found to be within the acceptable range.

Conclusion: The proposed method to be fast, simple, feasible and affordable in assay condition. During stability tests, it can be used for routine analysis of the selected drug.

Key Words: Nimodipine, RP-HPLC, Method Development, Validation, Accuracy, Robustness.

Corresponding author:**Name:** Kuchuri Mounika,**Department:** Pharmaceutical Analysis**College:** Teegala Krishna Reddy College of Pharmacy, Medbowli, Meerpet (V), Balapur (M), Ranga Reddy (Dist), Hyderabad – 500097, Telangana, **Phone No:** +91 8074971548,**Mail Id:** kuchurimounika2000@gmail.com

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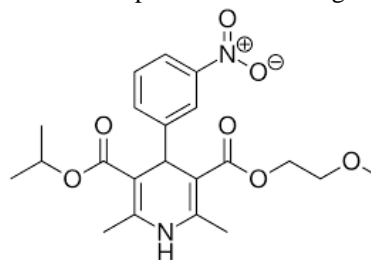
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INTRODUCTION:

Nimodipine is a calcium channel blocker used to improve neurological outcomes in patients with subarachnoid haemorrhage due to a ruptured intracranial aneurysm. Nimodipine is a 1, 4-dihydropyridine calcium channel blocker¹. It acts primarily on vascular smooth muscle cells by stabilizing voltage-gated L-type calcium channels in their inactive conformation. By inhibiting the influx of calcium in smooth muscle cells, Nimodipine prevents calcium-dependent smooth muscle contraction and subsequent vasoconstriction. Compared to other calcium channel blocking agents, Nimodipine exhibits greater effects on cerebral circulation than on peripheral circulation. Nimodipine is used to as an adjunct to improve the neurologic outcome following subarachnoid hemorrhage from ruptured intracranial aneurysm². For use as an adjunct to improve neurologic outcome following subarachnoid hemorrhage (SAH) from ruptured intracranial berry aneurysms by reducing the incidence and severity of ischemic deficits. Nimodipine is used to treat symptoms resulting from a ruptured blood vessel in the brain (subarachnoid hemorrhage)³. It works by increasing

MATERIALS AND METHODS

the blood flow to injured brain tissue. The IUPAC Name of Nimodipine is 3-O-(2-methoxyethyl) 5-O-propan-2-yl 2, 6-dimethyl-4-(3-nitro phenyl)-1, 4-dihydro pyridine-3, 5-dicarboxylate. The Chemical Structure of Nimodipine is shown in figure-1.

**Fig-1: Chemical Structure of Nimodipine**

Literature survey revealed that Nimodipine was determined in bulk form and marketed pharmaceutical dosage forms by RP-HPLC as well as in biological fluids using liquid chromatography and liquid chromatography mass spectrometric methods³¹⁻³⁴. In the present work the authors have developed a simple, rapid, precise, accurate and robust stability indicating liquid chromatographic method for the determination of Nimodipine in pure substances and marketed pharmaceutical dosage forms as per ICH guidelines⁴.

Table-1: List of Instrument used

S. No.	Instruments/Equipments/Apparatus
1.	Waters HPLC with Empower2 Software with Isocratic with UV-Visible Detector.
2.	ELICO SL-159 UV – Vis spectrophotometer
3.	Electronic Balance (SHIMADZU ATY224)
4.	Ultra Sonicator (Wensar wuc-2L)
5.	Thermal Oven
6.	Symmetry RP C ₁₈ , 5µm, 250mm x 4.6mm i.d.
7.	P ^H Analyzer (ELICO)
8.	Vacuum filtration kit (BOROSIL)

Table-2: List of Chemicals used

S.N.	Name	Specifications		Manufacturer/Supplier
		Purity	Grade	
1.	Doubled distilled water	99.9%	HPLC	Sd fine-Chem ltd; Mumbai
2.	Methanol	99.9%	HPLC	Loba Chem; Mumbai.
3.	Dipotassium hydrogen orthophosphate	96%	L.R.	Sd fine-Chem ltd; Mumbai
4.	Acetonitrile	99.9%	HPLC	Loba Chem; Mumbai.
5.	Potassium dihydrogen orthophosphate	99.9%	L.R.	Sd fine-Chem ltd; Mumbai
6.	Sodium hydroxide	99.9%	L.R.	Sd fine-Chem ltd; Mumbai
7.	Hydrochloric acid	96%	A.R.	Sd fine-Chem ltd; Mumbai
8.	3% Hydrogen Peroxide	96%	A.R.	Sd fine-Chem ltd; Mumbai

Selection of Wavelength:

The standard & sample stock solutions were prepared separately by dissolving standard & sample in a solvent in mobile phase diluting with the same solvent. (After optimization of all

conditions) for UV analysis. It scanned in the UV spectrum in the range of 200 to 400nm. This has been performed to know the maxima of Nimodipine, so that the same wave number can be

utilized in HPLC UV detector for estimating the Nimodipine⁵.

Preparation of Standard Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of Methanol and sonicate to dissolve and removal of air completely and make volume up to the mark with the same Methanol.

Further pipette 0.5ml of the above Nimodipine stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Procedure:

Inject the samples by changing the chromatographic conditions and record the chromatograms, note the conditions of proper peak elution for performing validation parameters as per ICH guidelines^{25,30}.

Preparation of Sample Solution:

Twenty tablets were taken and the average weight was calculated as per the method prescribed in I.P. The weighed tablets were finally powdered and triturated well. A quantity of powder of Nimodipine equivalent to 10mg were transferred to clean and dry 10 ml volumetric flask and 7 ml of HPLC grade methanol was added and the resulting solution was sonicated for 15 minutes. Make up the volume up to 10 ml with same solvent. Then 1 ml of the above solution was diluted to 10 ml with HPLC grade methanol. One ml (0.5 ml) of the prepared stock solution diluted to 10 ml and was filtered through membrane filter (0.45 μ m) and finally sonicated to degas⁶.

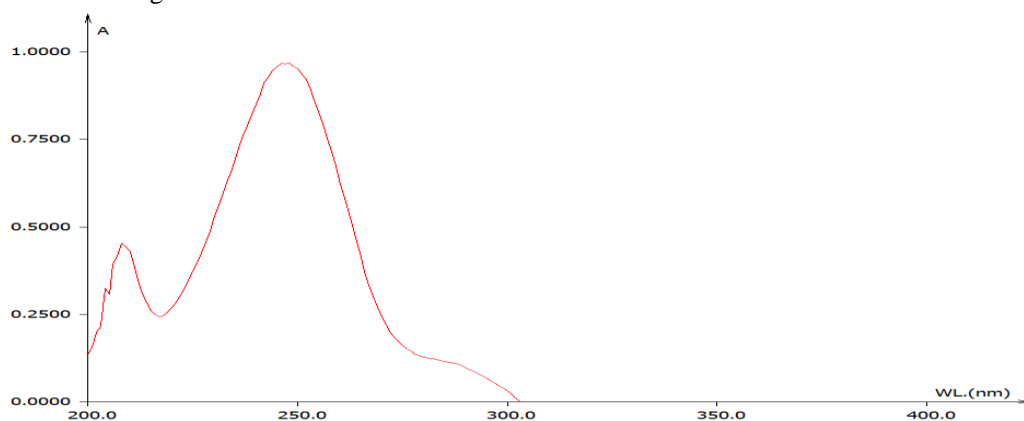


Fig-2: UV Spectrum for Nimodipine

Summary of Optimized Chromatographic Conditions:

The Optimum conditions obtained from experiments can be summarized as below⁸⁻¹⁰:

Table-3: Summary of Optimised Chromatographic Conditions

Mobile phase	Phosphate Buffer (0.02M): Acetonitrile = 48:52% v/v (pH-2.80)
Column	Symmetry ODS (C ₁₈) RP Column, 250 mm x 4.6 mm, 5 μ m
Column Temperature	Ambient
Detection Wavelength	248 nm
Flow rate	1.0 ml/ min.
Run time	08 min.

Preparation of 0.02M Potassium Dihydrogen Orthophosphate Solution:

About 2.72172grams of Potassium dihydrogen orthophosphate was weighed and transferred into a 1000ml beaker, dissolved and diluted to 1000ml with HPLC Grade water. The pH was adjusted to 2.80 with diluted orthophosphoric acid Solution.

Preparation of Mobile Phase:

480mL (48%) of above Phosphate buffer solution and 520mL of HPLC Grade Acetonitrile (52%) were mixed well and degassed in ultrasonic water bath for 15 minutes. The resulted solution was filtered through 0.45 μ m filter under vacuum filtration⁷.

Optimization of Chromatographic Conditions:

The chromatographic conditions⁶ were optimized by different means. (Using different column, different mobile phase, different flow rate, different detection wavelength & different diluents for sample preparation etc.

RESULTS AND DISCUSSION

Method Development and its Validation for Nimodipine by RP-HPLC

Method Development:

Selection of Wavelength:

While scanning the Nimodipine solution we observed the maxima at 248 nm. The UV spectrum has been recorded on ELICO SL-159 make UV – Vis spectrophotometer model UV-2450. The scanned UV spectrum is attached in the following page,

Temperature of Auto sampler	Ambient
Diluent	Mobile Phase
Injection Volume	20 μ l
Mode of Elution	Isocratic
Retention time	3.684 minutes

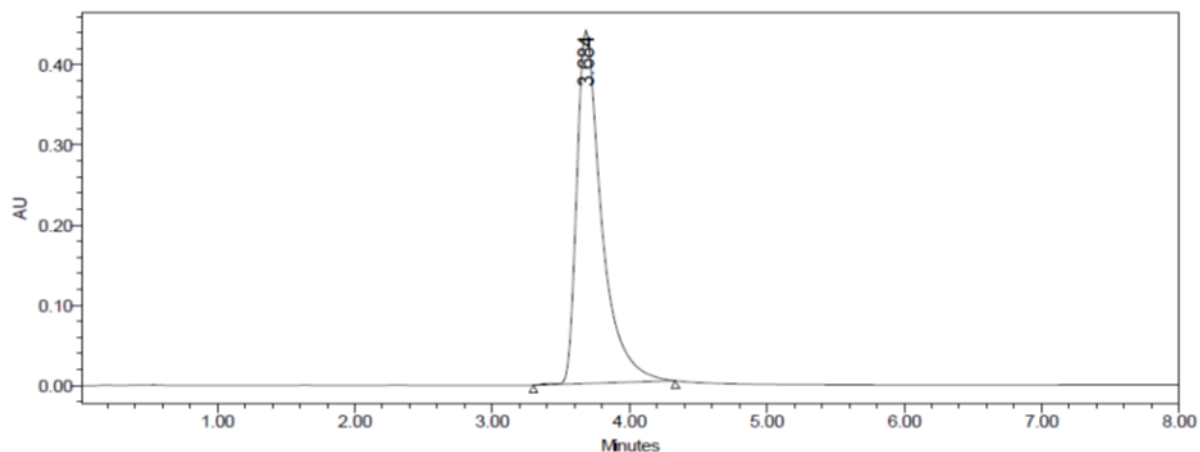


Fig-3: Chromatogram of Nimodipine in Optimized Chromatographic Condition

Validation of Analytical Method:

After the chromatographic and the experimental conditions were established, the method was validated by the determination of the following parameters such as specificity, system suitability, linearity, precision, accuracy, robustness, limit of detection (LOD) and limit of quantitation (LOQ) as per ICH Q2 (R1) guidelines¹¹⁻¹⁴.

1. Accuracy:

Preparation of Standard Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Further pipette 0.5ml of the above Nimodipine stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

For Preparation of 80% Standard Stock Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.4ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

For Preparation of 100% Standard Stock Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.5ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent¹⁵.

For Preparation of 120% Standard Stock Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.6ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Recovery Study:

To determine the accuracy of the planned technique, recovery studies were distributed by adds completely different amounts (80%, 100%, and 120%) of pure drug of Nimodipine were taken and extra to the pre-analyzed formulation of concentration 50 μ g/ml. From that proportion recovery values were calculated¹⁶⁻¹⁸. The results were shown in table-4.

Table-4: Accuracy Readings

Sample ID	Concentration ($\mu\text{g/ml}$)		Peak Area	% Recovery of Pure drug	Statistical Analysis
	Amount Added	Amount Found			
S ₁ : 80 %	40	40.141	502647	100.352	Mean= 100.3947% S.D. = 0.071319 % R.S.D.= 0.071038
S ₂ : 80 %	40	40.191	503214	100.477	
S ₃ : 80 %	40	40.142	502656	100.355	
S ₄ : 100 %	50	50.044	614215	100.088	Mean= 99.98533% S.D. = 0.183045 % R.S.D.= 0.183071
S ₅ : 100 %	50	49.887	612451	99.774	
S ₆ : 100 %	50	50.047	614254	100.094	
S ₇ : 120 %	60	60.192	728547	100.32	Mean= 100.311% S.D. = 0.408574 % R.S.D.= 0.407308
S ₈ : 120 %	60	59.939	725698	99.898	
S ₉ : 120 %	60	60.429	731211	100.715	

2. Precision:

2.1. Repeatability

Preparation of Nimodipine Product Solution for Precision:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.5ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Procedure:

The standard solution was injected for six times and measured the area for all six injections in HPLC⁹. The %RSD for the area of six replicate injections was found to be within the specified limits.

The exactitude of every technique was determined one by one from the height areas & retention times obtained by actual determination of six replicates of a set quantity of drug¹⁹. Nimodipine (API). The % relative variance was calculated for Nimodipine square measure bestowed within the table-5.

Table-5: Repeatability Readings

HPLC Injection Replicates of Nimodipine	Retention Time (Minutes)	Peak Area
Replicate – 1	3.649	5674158
Replicate – 2	3.684	5654715
Replicate – 3	3.687	5665841
Replicate – 4	3.688	5654578
Replicate – 5	3.688	5652284
Replicate – 6	3.687	5641487
Average		5657177
Standard Deviation		11369.72
% RSD		0.200979

2.2. Intermediate Precision/Ruggedness:

2.2.1. Intra-Day & Inter-Day:

The intra & inter day variation of the method was carried out & the high values of mean assay & low values of standard deviation & % RSD (% RSD < 2%) within a day & day to day variations for Nimodipine revealed that the proposed method is precise²⁰.

Procedure:

Analyst 1: The standard solution was injected for six times and measured the area for all six injections in HPLC. The %RSD for the area of six replicate injections was found to be within the specified limits.

Analyst 2:

The standard solution was injected for six times and measured the area for all six injections in HPLC. The %RSD for the area of six replicate injections was found to be within the specified limits.

Analyst-1/Intra Day/Day-1:**Table-6: Results of Ruggedness for Nimodipine Analyst 1**

S.No.	Peak Name	RT	Area (μV*sec)	Height (μV)	USP Plate Count	USP Tailing
1	Nimodipine	3.687	584968	65982	4985	1.42
2	Nimodipine	3.688	582479	66354	4876	1.46
3	Nimodipine	3.688	586236	67425	4896	1.48
4	Nimodipine	3.687	586985	65982	4986	1.47
5	Nimodipine	3.684	582679	65932	5016	1.45
6	Nimodipine	3.649	583989	65874	4987	1.43
Mean			584556			
Std. Dev.			1846.658			
% RSD			0.315908			

Analyst 2/Inter Day/Day-2:**Table-7: Results of Intermediate Precision Analyst 2 for Nimodipine**

S.No.	Peak Name	RT	Area (μV*sec)	Height (μV)	USP Plate count	USP Tailing
1	Nimodipine	3.649	598698	66985	5265	1.49
2	Nimodipine	3.684	596847	67458	5168	1.47
3	Nimodipine	3.687	596354	66985	5436	1.46
4	Nimodipine	3.688	598676	67854	5369	1.45
5	Nimodipine	3.688	596874	68521	5247	1.48
6	Nimodipine	3.687	598989	67898	5375	1.42
Mean			597739.7			
Std. Dev.			1168.098			
% RSD			0.195419			

3. Linearity & Range:

Preparation of Drug Solutions for Linearity:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Further pipette 0.5ml of the above Nimodipine stock solutions into a 10ml volumetric flask and dilute up to the mark with Mobile Phase²¹.

Preparation of Level – I (30ppm of Nimodipine):

Take 0.3ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – II (40ppm of Nimodipine):

Take 0.4ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – III (50ppm of Nimodipine):

Take 0.5ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – IV (60ppm of Nimodipine):

Take 0.6ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – V (70ppm of Nimodipine):

Take 0.7ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Procedure:

Inject each level into the chromatographic system and measure the peak area.

Plot a graph of peak area versus concentration (on X-axis concentration and on Y-axis Peak area) and calculate the correlation coefficient²².

The calibration curve showed good linearity in the range of 0-70 μ g/ml, for Nimodipine (API) with correlation coefficient (r^2) of 0.999 (Fig-4). A typical calibration curve has the regression equation of $y = 11266x + 50416$ for Nimodipine.

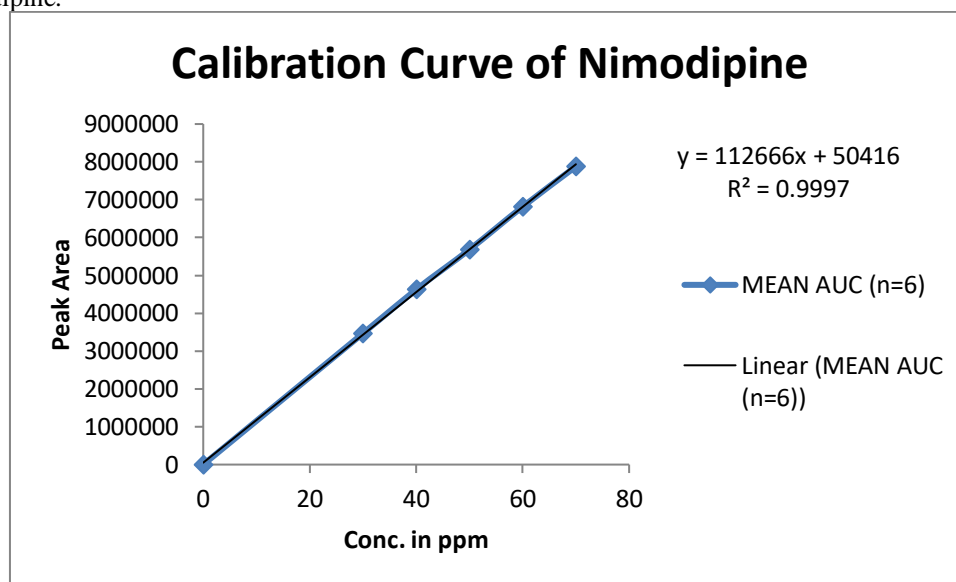


Fig-4: Calibration Curve of Nimodipine (API)

Table-8: Linearity Results

CONC.(μ g/ml)	MEAN AUC (n=6)
0	0
30	3465974
40	4626478
50	5682284
60	6815478
70	7878721

Linearity Plot: The plot of Concentration (x) versus the Average Peak Area (y) data of Nimodipine is a straight line.

$$Y = mx + c$$

$$\text{Slope (m)} = 112666$$

$$\text{Intercept (c)} = 50416$$

$$\text{Correlation Coefficient (r)} = 0.99$$

Validation Criteria: The response linearity is verified if the Correlation Coefficient is 0.99 or greater.

Conclusion: Correlation Coefficient (r) is 0.99, and the intercept is 50416. These values meet the validation criteria²³.

4. Method Robustness:

The robustness was performed for the flow rate variations from 0.9 ml/min to 1.1ml/min and mobile phase ratio variation from more organic phase to less organic phase ratio for Nimodipine. The method is robust only in less flow condition and the method is robust even by change in the Mobile phase $\pm 5\%$. The standard and samples of Nimodipine were injected by changing the conditions of chromatography. There was no significant change in the parameters like resolution, tailing factor, asymmetric factor, and plate count.

The analysis was performed in different conditions to find the variability of test results. The following conditions are checked for variation of results.

For Preparation of Standard Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.5ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Effect of Variation of Flow Conditions:

The sample was analyzed at 0.9ml/min and 1.1ml/min instead of 1ml/min, remaining conditions are same. 10 μ l of the above sample was injected and chromatograms were recorded.

Effect of Variation of Mobile Phase Organic Composition:

The sample was analyzed by variation of mobile phase i.e. Acetonitrile: Phosphate Buffer was taken in the ratio and 40:60, 30:70 instead of 35:65, remaining conditions are same. 10 μ l of the above sample was injected and chromatograms were recorded²⁴.

Table-9: Results for Robustness

Parameter Used for Sample Analysis	Peak Area	Retention Time	Theoretical Plates	Tailing Factor
Actual Flow rate of 1.0 mL/min	584624	3.649	1.42	4765
Less Flow rate of 0.9 mL/min	598676	3.687	1.49	4856
More Flow rate of 1.1 mL/min	612543	3.649	1.46	4965
Less organic phase	578642	3.688	1.49	4758
More organic phase	569896	3.684	1.47	4962

5. LOD & LOQ:

LOD: The detection limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be detected but not necessarily quantitated as an exact value²⁵.

$$\text{LOD} = 3.3 \times \sigma / s$$

Where

σ = Standard deviation of the response

S = Slope of the calibration curve

LOQ: The quantitation limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be quantitatively determined²⁶.

$$\text{LOQ} = 10 \times \sigma / S$$

Where

σ = Standard deviation of the response

S = Slope of the calibration curve

Table-10: Results of LOD & LOQ

SE of Intercept	48846.22527
SD of Intercept	109223.4801
LOD	3.199168
LOQ	9.694449

Observation: The Minimum concentration level at which the analyte can be reliable detected (LOD) & quantified (LOQ) were found to be 3.19 & 9.69 µg/ml respectively.

6. System Suitability Parameter:

System quality testing is associate degree integral a part of several analytical procedures. The tests square measure supported the idea that the instrumentation, physics, associate degree analytical operations and samples to be analyzed represent an integral system that may be evaluated intrinsically. Following system quality check parameters were established²⁷. The information square measured shown in Table-11 & 12.

Preparation of Standard Solution:

Accurately weigh and transfer 10 mg of Nimodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of Methanol and sonicate to dissolve and removal of air completely and make volume up to the mark with the same Methanol.

Further pipette 0.5ml of the above Nimodipine stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Procedure:

The standard solution was injected for six times and measured the area for all six injections in HPLC. The %RSD for the area of six replicate injections was found to be within the specified limits.

Table-11: Knowledge of System Suitability Parameter

S.No.	Parameter	Limit	Result
1	Asymmetry	$T \leq 2$	Nimodipine =0.98
2	Theoretical plate	$N > 2000$	Nimodipine =4782
3	Tailing Factor	$T < 2$	Nimodipine =1.49

Table-12: Results of System Suitability for Nimodipine

S.No.	Peak Name	RT	Area (µV*sec)	Height (µV)	USP Plate Count	USP Tailing
1	Nimodipine	3.644	584635	65847	4857	1.48
2	Nimodipine	3.645	582695	65421	4955	1.42
3	Nimodipine	3.644	587432	65369	4875	1.47
4	Nimodipine	3.662	589687	65748	4796	1.46
5	Nimodipine	3.660	582547	65398	4952	1.49
6	Nimodipine	3.660	589656	652418	4896	1.47
Mean			586108.7			
Std. Dev.			3275.654			
% RSD			0.558882			

7. Specificity:

Specificity can be determined by comparing the chromatograms obtained from the drugs with the chromatogram obtained from the blank solution. Blank solution was prepared by mixing the excipients in the mobile phase without drug. Drug solutions were prepared individually and the sample containing one drug was

also prepared. Now these mixtures were filtered by passing through 0.45 μ membrane filter before the analysis. In this observation no excipient peaks were obtained near the drug in the study run time. This indicates that the proposed method was specific²⁸.

The chromatograms representing the peaks of blank, Nimodipine and the sample containing the one drug was shown in following figures respectively.

Observation: In this test method blank, standard solutions were analyzed individually to examine the interference. The above chromatograms show that the active ingredient was well separated from blank and their excipients and there was no interference of blank with the principal peak. Hence the method is specific.

8. Estimation of Nimodipine in Pharmaceutical Dosage Form

Twenty pharmaceutical dosage forms were taken and the I.P. method was followed to work out the typical weight. On top of weighed tablets were finally pulverized and triturated well. A amount of powder cherish twenty five mg of medicine were transferred to twenty five cc meter flask, build and resolution was sonicated for quarter-hour, there once volume was created up to twenty five cc with same solvent. Then ten cc of the on top of resolution was diluted to a hundred cc with mobile part. The answer was filtered through a membrane filter (0.45 μ m) and sonicated to remove. The answer ready was injected in 5 replicates into the HPLC system and therefore the observations were recorded²⁹⁻³⁰.

A duplicate injection of the quality resolution was conjointly injected into the HPLC system and therefore the peak areas were recorded. The information square measure shown in Table-13.

ASSAY:

Assay % =

$$\frac{\text{AT}}{\text{AS}} \times \frac{\text{WS}}{\text{DS}} \times \frac{\text{DT}}{\text{WT}} \times \frac{\text{P}}{100} \times \text{Avg. Wt} = \text{mg/tab}$$

Where:

- AT = Peak space of drug obtained with check preparation
- AS = Peak space of drug obtained with normal preparation
- WS = Weight of operating normal taken in mg
- WT = Weight of sample taken in mg
- DS = Dilution of normal resolution
- DT = Dilution of sample resolution
- P = proportion purity of operating normal

Table-13: Recovery Data for Estimation of Nimodipine

Brand Name of Nimodipine	Labelled amount of Drug (mg)	Mean (\pm SD) amount (mg) found by the proposed method (n=6)	Assay % (\pm SD)
Nimpod 30mg Tablet (Intas Pharmaceuticals Ltd)	30mg	29.687 (\pm 0.486)	99.749 (\pm 0.398)

Observation: The amount of drugs in Nimodipine Tablet was found to be 29.687 (\pm 0.486) mg/tab for Nimodipine & % assay was 99.749 (\pm 0.398).

Forced Degradation Studies

Following convention was entirely clung to for constrained corruption of Nimodipine Active Pharmaceutical Ingredient (API). The API (Nimodipine) was subjected to pressure conditions in different approaches to watch the rate and degree of corruption that is probably going to happen over the span of capacity as well as after organization to body. This is one kind of quickened dependability contemplates that encourages us deciding the destiny of the medication that is probably going to occur after prolonged stretch of time stockpiling, inside a brief timeframe as contrast with the constant or long haul steadiness testing. The different debasement pathways²⁸ contemplated are Acid/corrosive hydrolysis, Alkali/fundamental hydrolysis, Thermal/warm Degradation, photolytic corruption/ Degradation and oxidative Degradation/corruption.

Results of Degradation Studies:

The results of the stress studies indicated the Specificity of the method that has been developed. Nimodipine was stable in photolytic and peroxide stress conditions. The result of forced degradation studies are given in the following table-14.

Table-14: Results of Forced Degradation Studies of Nimodipine API

Stress Condition	Time	Assay of active substance	Assay of degraded products	Mass Balance (%)
Acid Hydrolysis (0.1 M HCl)	24Hrs.	98.76	1.24	100.0
Basic Hydrolysis (0.1 M NaOH)	24Hrs.	98.63	1.37	100.0
Thermal Degradation (50 °C)	24Hrs.	93.98	6.02	100.0
UV (248nm)	24Hrs.	98.84	1.16	100.0
3 % Hydrogen Peroxide	24Hrs.	94.61	5.39	100.0

SUMMARY AND CONCLUSION:

The analytical method was developed by studying different parameters. First of all, maximum absorbance was found to be at 248nm and the peak purity was excellent. Injection volume was selected to be 20µl which gave a good peak area. The column used for study was Symmetry ODS (C18) RP Column, 250 mm x 4.6 mm, 5µm particle size because it was giving good peak. Ambient temperatures were found to be suitable for the nature of drug solution. The flow rate was fixed at 1.0ml/min because of good peak area and satisfactory retention time. Mobile phase is Phosphate Buffer (0.02M) and Acetonitrile were taken in the ratio of 48:52 % v/v (pH-2.80) was fixed due to good symmetrical peak. So this mobile phase was used for the proposed study. Methanol was selected because of maximum extraction sonication time was fixed to be 10min at which all the drug particles were completely soluble and showed good recovery. Run time was selected to be 8.0 min because analyze gave peak around 3.684min and also to reduce the total run time. The percent recovery was found to be 98.0-102 was linear and precise over the same range. Both system and method precision was found to be accurate and well within range. The analytical method was found linearity over the range of 30-70ppm of the Nimodipine target concentration. The analytical passed both robustness and ruggedness tests. On both cases, relative standard deviation was well satisfactory.

REFERENCES:

- <https://go.drugbank.com/drugs/DB00393>
- <https://en.wikipedia.org/wiki/Nimodipine>
- <https://pubchem.ncbi.nlm.nih.gov/compound/00004497>
- Journal of Pharmaceutical and Biomedical Analysis Volume 21, Issue 2, Pages 371-382, 1 November 1999.
- Tropical Journal of Pharmaceutical Research, © Pharmacotherapy Group, Volume: 8(5), Pg No: 449-454, October 2009.
- Rabi Sankar, Instrumental Method of Analysis, P-18-6, P-18-3.
- Lloyd R. Snyder *et al*, Practical HPLC Method Development, 2nd edition, P-503.
- Guidance for industry, Analytical Procedure and Method Validation, U.S. Department of Health and Human Services FDA, August 2000.
- Y. F. Cheng, T.H. Walter, Z. Lu, P. Iraneta, C. Gendreau, U. D. Neue, J. M. Grassi, J. L. Carmody, J. E. O' Gara, and R. P. Fisk, LCGC, Volume: 18(10), 1162, 2000.
- The United State Pharmacopeia 25/National Formulary 20, Ch. 1225, (The United State Pharmacopeia Convention, Inc., Rockville, Maryland, pg. 2256-2259, 2002.
- ICH Q2B: Validation of Analytical Procedure; Methodology (International Conferences on Harmonization of Technical requirements for the registration of Drugs for Human use, Geneva, Switzerland, May 1997.
- ICH Q2B: Validation of Analytical Procedure; Methodology (International Conferences on Harmonization of Technical requirements for the registration of Drugs for Human use, Geneva, Switzerland, Nov 2003.
- M. V. Gorenstein, J. B. Li, J. Van Antwerp, and D. Chapman, LCGC Volume 12(10), Pg no: 768-772, 1994.
- Matheson A.J., Noble S., Drugs, Volume 59, ISSN Number 4, Pg no: 829-835, 2000.
- Anttila S, Leinonen E: Duloxetine Eli Lilly. Curr Opin Investig Drugs.; Volume: 3 (8), Pg no: 1217-21, 2002.
- Gan TJ: Selective serotonin 5-HT₃ receptor antagonists for postoperative nausea and vomiting: are they all the same? CNS Drugs.; Volume; 19 (3), Pg no: 225-38, 2005.

17. Tan M: Granisetron: new insights into its use for the treatment of chemotherapy-induced nausea and vomiting. *Expert Opin Pharmacother*. Volume: 4(9), Pg no: 1563-71, 2003.
18. Ahuja S. In: *High Pressure Liquid Chromatography of Comprehensive Analytical Chemistry*. Elsevier Publications. 2006.
19. Principles and Methods. In: *Amesham Biosciences of Reversed Phase Chromatography*. 6-8.
20. Snyder LR, Kirkland JJ and Glajch JL. In: *Practical HPLC Method Development*, 2nd Ed, John Wiley and Sons Inc. Canada. 1997.
21. Mohammad T et al., HPLC Method Development and Validation for Pharmaceutical Analysis- A Review. *International Pharmaceutica Scientia*. 2012, 2(3), 14.
22. Snyder LR, Kirkland JJ and Glajch JL. In: *Practical HPLC Method Development*. 2nd ed, 2001.
23. Vibha G et al., Development and validation of HPLC method - a review. *International Research Journal of Pharmaceutical and Applied Sciences*. 2012, 2(4), 22-23.
24. Bliesner DM. In: *Validating Chromatographic Methods*. John Wiley & sons Inc. 2006, 88-92.
25. Validation of Analytical Procedures: Methodology. ICH-Guidelines Q2B, Geneva. 1996, 11. (CPMP/ICH/281/95).
26. Development and validation of HPLC method - A Review, Vibha Gupta et al, *International Research Journal of Pharmaceutical and Applied Sciences*, 2012; 2(4):17-25.
27. A Review: HPLC Method Development and Validation, Santosh Kumar Bhardwaj *et al. *International Journal of Analytical and Bioanalytical Chemistry*, accepted 20 November 2015.
28. Method Development: A Guide to Basics Quantitative & Qualitative HPLC, LC, GC chromatography.
29. Lalit V Sonawane* *Bioanalytical Method Validation and Its Pharmaceutical Application- A Review Pharmaceutica Analytical Acta* 2014, 5:3Center for Drug Evaluation and Research (CDER) Reviewer Guidance.
30. ICH Topic Q 2 (R1) Validation of Analytical Procedures: Text and Methodology.
31. Lubna B. Shaikh*1, Vishal V. Pande2, Deepak S. Musmade3 and Poonam P. Patil3, Development and Validation of RP-HPLC Method for Estimation of Process Related Impurity in Nimodipine Bulk and Formulation, *Scholars Research Library Der Pharmacia Lettre*, 2015, 7 (3): 287-290.
32. Xiaojun Shang, Suying Ma, Zheshen Li, Development and Validation of a RP-HPLC Method for Determination of Nimodipine in Sustained Release Tablets, *Journal of Chemistry*, Volume 2013, Article ID 612082, 4 pages, <http://dx.doi.org/10.1155/2013/612082>.
33. Kasture V.S.*, Pawar S.S., Patil P.P., Musmade D.S., Ajage R. K. and Gehlot Ganjendra, Development and Validation of Rp-Hplc Method for the Estimation of Process Related Impurities from Nimodipine Bulk and Formulation, *International Journal of Pharmacy*, 2014; 4(2): 189-195.
34. B. Rajani* and K. Mukkanti, Optimized and Validated RP-HPLC Method for the Estimation of Nimodipine in Tablet Dosage Form, *International Journal of Research In Pharmacy and Chemistry, IJRPC* 2014, 4(1), 105-109.