ejpmr, 2024, 11(7), 247-253

SJIF Impact Factor 7.065

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Review Article

ISSN 2394-3211

EJPMR

ANALYTICAL METHOD DEVELOPMENT AND VALIDATION FOR ESTIMATION OF AZELNIDIPINE IN BULK DRUG AND PHARMACEUTICAL DOSAGE FORM BY USING RP-HPLC

Akanksha D. Punekar* and Sonali A. Waghmare

Department of Pharmaceutical Quality Assurance, SND College of Pharmacy, Babhulgaon (Yeola), India.

***Corresponding Author: Akanksha D. Punekar**

Department of Pharmaceutical Quality Assurance, SND College of Pharmacy, Babhulgaon (Yeola), India.

Article Received on 13/05/2024 **Article Revised on 03/06/2024** **Article Accepted on 24/06/2024**

ABSTRACT

The development and validation of analytical methods for the estimation of pharmaceutical compounds play a crucial role in ensuring the safety and efficacy of drug products. Azelnidipine, a third-generation dihydropyridine calcium channel blocker, is known for its antihypertensive effects. This review article provides a comprehensive overview of the recent advancements in the development and validation of analytical methods for the estimation of Azelnidipine, with a specific focus on the RP-HPLC (Reverse Phase High-Performance Liquid Chromatography) method.

KEYWORDS: RP-HPLC, Drug, Azelnidipine, Methods, Analytical.

INTRODUCTION

Hypertension remains a significant global health concern, contributing to the burden of cardiovascular diseases. Calcium channel blockers (CCBs) have been integral in managing hypertension, with azelnidipine representing a notable member of the dihydropyridine class. Originally introduced for its vasodilatory effects, azelnidipines distinct pharmacological profile has garnered attention in the medical community.[1] Azelnidipine has gained popularity in the treatment of hypertension due to its unique pharmacological profile and reduced side effects compared to other calcium channel blockers. Accurate and precise estimation of Azelnidipine is essential for pharmacokinetic studies, bioequivalence assessments, and therapeutic drug monitoring. RP-HPLC, with its versatility and sensitivity, has emerged as a prominent technique for the bioanalysis of Azelnidipine. Azelnidipine is a calcium channel blocker used in the treatment of hypertension. It belongs to the dihydropyridine class of calcium channel blockers as shown in figure 1.[1,2] General information about Azelnidipine was shown in table 1.[2]

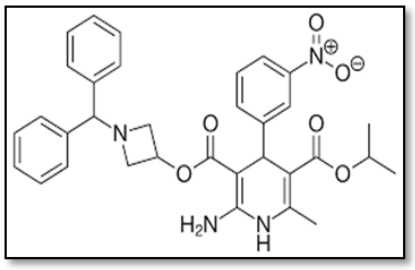

Figure 1: Structure of Azelnidipine.

Table 1: Drug Profile of Azelnidipine.

DRUG PROFILE	AZELNIDIPINE
Molecular Weight	582.646 g/mol
Molecular formula	$C_{33}H_{34}N_4O_6$
Drug Category	Calcium channel blocker
Chemical name	3-(1-benzhydrylazetidin-3-yl) 5-isopropyl 2-amino-6-methyl-4-(3-nitrophenyl)-1,4-dihydropyridine-3,5-dicarboxylate
Melting point	122 – 123°C
PKa	7.89
LogP	5.23
λ max	255 nm
Uses	Antihypertensive agent

Mechanism of Action

Azelnidipine blocks L-type calcium channels, particularly those in vascular smooth muscle cells. By inhibiting calcium influx through these channels, it reduces the contractility of smooth muscle and dilates peripheral arteries. The vasodilation results in a decrease in blood pressure as shown in figure 2.[3]

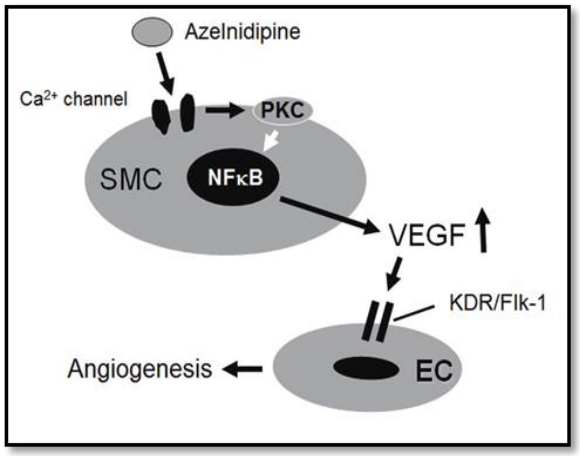

Figure 2: Mechanism of Action of Azelnidipine.

Pharmacokinetics

Absorption: Azelnidipine is rapidly absorbed after oral administration. However, its absorption is influenced by food, and taking it with a meal can increase its bioavailability.

Distribution: It is extensively bound to plasma proteins, mainly to albumin. The drug undergoes extensive distribution into tissues.

Metabolism: Azelnidipine undergoes extensive metabolism in the liver. The primary metabolite is azelnidipine dehydrate, which is pharmacologically active.

Elimination

The elimination half-life of azelnidipine is relatively long, leading to a once-daily dosing regimen. The drug and its metabolites are primarily excreted in the feces, with a smaller portion eliminated in the urine.[4-6]

Special Populations

Hepatic Impairment: Azelnidipine is metabolized in the liver, so hepatic impairment can affect its metabolism. Dose adjustments may be necessary in patients with liver dysfunction.

Renal Impairment: The drug is not significantly excreted through the kidneys, so dosage adjustments are generally not required in patients with renal impairment.

Drug Interactions: Azelnidipine can interact with other medications that affect cytochrome P450 enzymes, potentially leading to altered plasma concentrations.

Clinical Efficacy: Azelnidipine has been found to effectively lower blood pressure and is used in the management of hypertension. Its long half-life allows for once-daily dosing, contributing to patient compliance.[6,7]

RP-HPLC Technique

RP-HPLC is a powerful analytical technique widely used in the field of chromatography. It is a type of liquid chromatography that separates molecules based on their hydrophobicity, or more precisely, their affinity for a hydrophobic stationary phase. RP-HPLC is a versatile technique used in various scientific disciplines, including pharmaceuticals, biochemistry, environmental analysis, and food and beverage testing. It is particularly valuable for separating and quantifying complex mixtures of organic compounds.[8]

The fundamental principle of RP-HPLC revolves around the interaction between the analyte and the stationary phase. In reverse phase chromatography, the stationary phase is nonpolar, while the mobile phase is polar. This is in contrast to normal phase chromatography, where the stationary phase is polar.[9]

The stationary phase in RP-HPLC is typically a hydrophobic material, such as C18-bonded silica, where C18 refers to an 18-carbon alkyl chain. The mobile phase, on the other hand, is usually a mixture of water and an organic solvent, such as acetonitrile or methanol. The relative composition of these solvents can be adjusted to control the elution strength.[9,10]

Mechanism of RP-HPLC

The separation mechanism in RP-HPLC is based on the differential affinity of analytes for the stationary phase. Hydrophobic analytes interact more strongly with the nonpolar stationary phase, causing them to be retained longer, while hydrophilic analytes move more quickly through the column.[11]

Equipment

RP-HPLC systems consist of a high-pressure pump, a sample injector, a column containing the stationary phase, a detector, and a data analysis system as shown in figure 3. The high-pressure pump is essential to push the mobile phase through the column at a constant flow rate.[12]

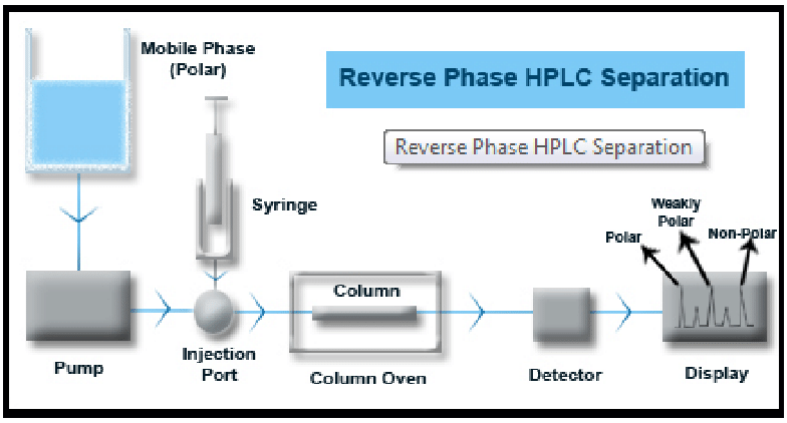

Figure 3: RP-HPLC Technique

Advantages

Wide Applicability: RP-HPLC can be applied to a broad range of compounds, from small organic molecules to large biomolecules.

High Sensitivity: The technique is highly sensitive, allowing for the detection of trace amounts of analytes.

Reproducibility: RP-HPLC provides reproducible results, making it a reliable method for routine analysis.

Resolution: It offers high-resolution separations, enabling the identification and quantification of individual components in a mixture.[13]

Method Development

Azelnidipine is a hydrophobic compound, so a reverse phase column with a nonpolar stationary phase, such as C18, is suitable. Knowledge of its pKa, solubility, and chemical stability aids in mobile phase selection.[14,15]

1. Selection of Chromatographic Column

Choose a reverse phase column with C18 packing material, as it provides good retention for hydrophobic compounds like Azelnidipine. Column dimensions (length, diameter) impact resolution and analysis time.[14]

2. Optimization of Mobile Phase

Mobile Phase Composition: A typical mobile phase for RP-HPLC includes a mixture of water (polar) and an organic solvent (nonpolar; acetonitrile or methanol). The percentage of organic solvent influences separation.[15]

3. pH Adjustment

Azelnidipine may exhibit pH-dependent solubility. Adjusting the pH of the mobile phase can impact retention time and peak shape. Incorporate buffers (e.g., ammonium acetate) to control pH and improve peak shape.

4. Flow Rate Optimization

Flow rate affects analysis time and resolution. Higher flow rates reduce analysis time but may compromise resolution.

5. Column Temperature

Temperature influences compound solubility and column efficiency. Optimize temperature for the best compromise between speed and resolution.

6. Injection Volume

The injected volume affects peak shape and resolution. Smaller volumes reduce band broadening but may limit sensitivity.

7. Detector Wavelength

Set the detector wavelength to the maximum absorbance of Azelnidipine. For UV detection, a wavelength around 360 nm is often suitable.

8. Gradient Elution Vs Isocratic Elution

Consider gradient elution for complex samples to improve separation. Isocratic elution may be sufficient for simpler samples.

9. Sample Preparation

Evaluate sample preparation techniques such as dilution, filtration, or solid-phase extraction to optimize sensitivity and minimize matrix effects.[16,19]

UV SPECTROSCOPIC METHOD

Selection of solvent

Prepare a mixture of Water and Methanol in the ratio 10:90 v/v respectively and mix. Sonicate to degas. Used as solvent for dissolving azelnidipine.[20]

Selection of Wavelength

An accurately weighed quantity about 27 mg of Azelnidipine standard was transferred to 250 mL volumetric flask. Add 200 mL of diluent, sonicate to dissolve and dilute up to the mark with diluent and mixed.[21]

Determination of λ Max

Between 400 and 200 nm, the standard solutions were scanned independently. Since the spectra at 255 nm shows strong absorbance, the azelnidipine λ max at 255 nm as shown in figure 4 was chosen for drug estimation in table 2.[22,23]

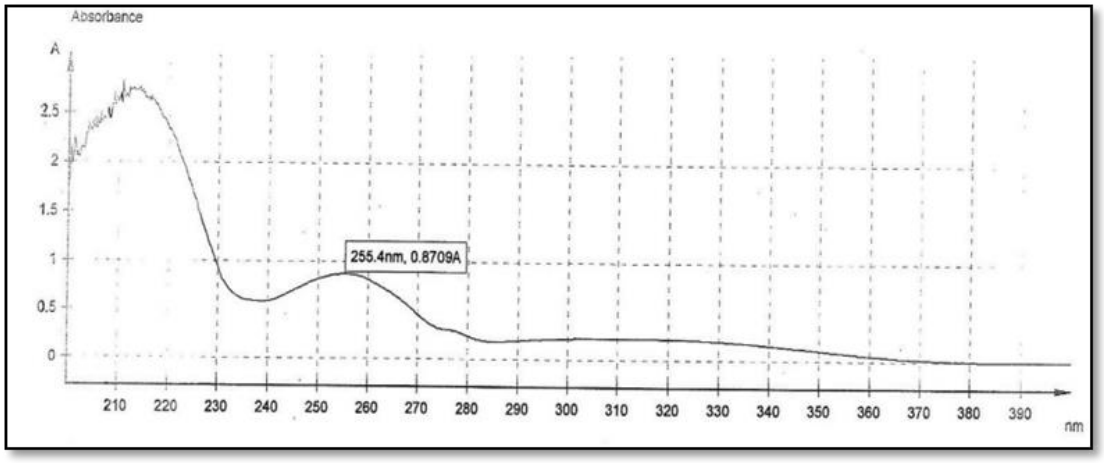

Figure 4: λ Max of azelnidipine.

Table 2: λ max of Azelnidipine.

Sr. No.	Wavelength (nm)	Absorbance
1	255	0.8709 A

Method Validation

In various studies, the method was validated for linearity, accuracy, precision and limit of detection, and limit and quantitation.[24-27]

Accuracy and precision

Recovery experiments determined the method's accuracy. The recovery studies were conducted six times, with a mean percentage recovery of 81.52 along with its standard deviation. It was discovered from the data that the additional recoveries of common medications were correct. Studies on intraday and interday variation proved the methods accuracy.[26] Six repeated injections of the standard and sample solutions were performed during the intraday trials. The response factor of the drug peaks and the percentage CV were computed and are shown in Table 3.[28] Six repeated injections of the standard and sample solutions were made for three days in a row as part of the interday variation investigations. The response factor of the drug peaks and the percentage of CV were computed and are shown in Table 3. The devised RP-HPLC method was found to be exact based on the data gathered.[29]

Table 3: Intraday and Interday precision studies of Azelnidipine.

Parameters	Intraday studies			Interday studies		
	5	15	30	5	15	30
	(mcg/ml)			(mcg/ml)		
Mean	4.6834	14.6643	29.4287	4.6094	14.6222	29.5055
SD	0.27	0.13	0.45	0.41	0.35	0.39
%CV	5.67	0.85	1.52	8.85	2.36	1.31
%Accuracy	93.6	97.7	98.1	92.19	97.48	98.35

Linearity

For working concentration, linearity was assessed in the 50% to 150% of azelnidipine range as shown in table 4. Azelnidipine working concentration is 40 ppm.[30-32]

Table 4: Linearity of Azelnidipine.

Level (%)	Concentration w.r.t sample (mg/mL)	Peak Area Injection - 1	Peak Area Injection - 2	Mean Peak Area
50	20	369854	371399	370627
75	30	558241	554928	556585
100	40	742160	743629	742895
125	50	930312	928634	929473
150	60	1123694	1099936	1111815
Correlation Co-coefficient (R)				0.9993
Slope of regression line				18423.674
%Y- intercept				0.02

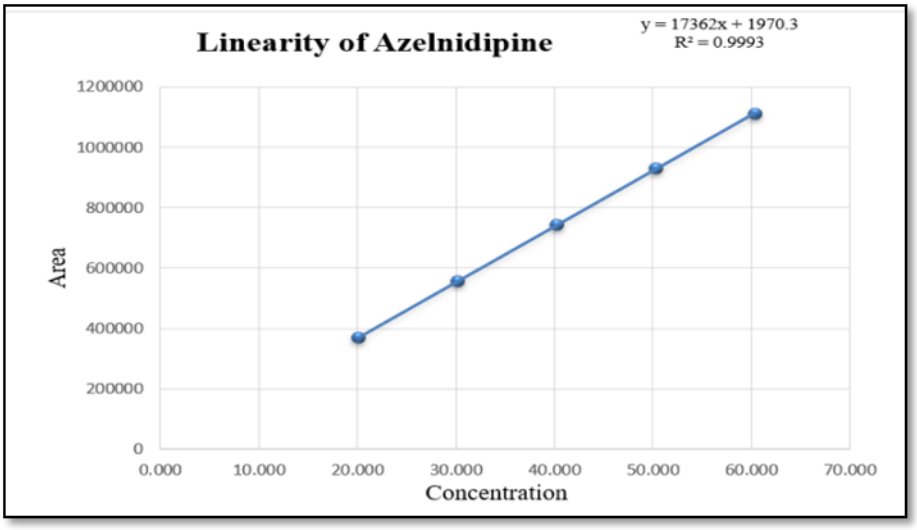

Figure 5: Linearity plot of Azelnidipine.

The information demonstrates that system suitability is met. The reaction is found to be linear, according to the data as shown in figure 5. The Y-intercept percent limit is within ± 2.0% of the operating levels corresponding Y-coordinate.[33,34]

Limit of Detection (LOD) and Limit of Quantification (LOQ)

By employing the developed RP-HPLC method to inject progressively lower concentrations of the standard solutions, LOD and LOQ of the produced method were ascertained.[35] The lowest concentration of the analyte (signal to noise ratio of 3) at which a detectable reaction is obtained is known as the LOD. It was discovered that azelnidipines LOD was 5 mcg/ml.[36] The lowest concentration of the analyte at which a reaction can be precisely measured is known as the limit of quantification, or LOQ. Azelnidipines LOQ was 11 mcg/ml, in that case.[37-39]

The following formula was used to determine LOD and LOQ in accordance with ICH regulations. LOQ = 10×σ /S and LOD = 3.3 × σ/S, where S is the calibration curves slope and σ is the response standard deviation.[40]

Solution stability

Both the standard and sample solutions were evaluated for five hours at room temperature to show that they were stable throughout the analysis process.[41] The findings demonstrate that the retention time and peak area of azelnidipine in both solutions stayed essentially unchanged, with no discernible degradation occurring within the specified time frame.[42-44] This suggests that the solutions were stable for a minimum of 5 hours, which was adequate to finish the analytical procedure.[45-47]

Research on the applicability of systems

For the standard solutions, the column efficiency, resolution, and peak asymmetry were computed.[48] The results showed that the system is suitable for analyzing this drug, and when the procedure is performed routinely, the system suitability parameters fall within a range of ±3% standard deviation.[49]

CONCLUSION

The development and validation of an analytical method for the estimation of Azelnidipine in both bulk drug and pharmaceutical dosage forms using Reverse Phase High Performance Liquid Chromatography (RP-HPLC) present a crucial advancement in pharmaceutical research. Through meticulous optimization and validation procedures, this method offers a reliable and accurate means of quantifying Azelnidipine, a widely used calcium channel blocker with therapeutic applications in hypertension and related cardiovascular conditions. The RP-HPLC method showcased in this review article demonstrates several key advantages. Firstly, it provides high sensitivity, specificity, and precision in detecting Azelnidipine within complex matrices, ensuring accurate quantification even at low concentrations. Secondly, the method offers excellent selectivity, allowing for the differentiation and quantification of Azelnidipine from potential impurities or excipients present in pharmaceutical formulations. Moreover, the robustness and reproducibility of the method across multiple analytical parameters underscore its reliability for routine quality control and regulatory compliance in pharmaceutical manufacturing.

Furthermore, the successful validation of this RP-HPLC method for Azelnidipine estimation in both bulk drug and pharmaceutical dosage forms validate its applicability across various stages of drug development and production. The method's ability to meet the stringent criteria outlined in regulatory guidelines ensures its suitability for use in pharmaceutical laboratories, facilitating the quality assessment and assurance of Azelnidipine-containing formulations. Overall, the development and validation of this RP-HPLC method for Azelnidipine estimation represent a significant contribution to analytical chemistry and

pharmaceutical sciences. By providing a robust, sensitive, and validated approach for quantifying Azelnidipine, this method enables researchers, analysts, and pharmaceutical manufacturers to ensure the quality, safety, and efficacy of Azelnidipine-based medications, ultimately benefiting patient health and well-being. Further research may focus on expanding the applicability of this method to other related compounds or exploring alternative analytical techniques to enhance efficiency and sensitivity.

REFERENCES

1. Oizumi K, Nishino H, Koike H, Sada T, Miyamoto M, Kimura T. Antihypertensive effects of CS−905, a novel dihydropyridine Ca2+ channel blocker. Jpn J Pharmacol, 1989; 51(1): 57-64. doi: 10.1254/jjp.51.57, PMID 2810942.
2. An H-M, Wang J-C. Determination of content and related substances of azelnidipine by HPLC. West China J Pharm Sci., 2006; 06.
3. Pan YF, Zhang JB, Ding J, Wang TM. Determination of azelnidipine tablets by HPLC. Qilu Pharm Aff., 2008; 07.
4. Kawabata K, Urasaki Y. Simultaneous determination of azelnidipine and two metabolites in human plasma using liquid chromatography-tandem mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci., 2006; 844(1): 45-52. doi: 10.1016/j.jchromb.2006.06.031, PMID 16854634.
5. Kawabata K, Samata N, Urasaki Y, Fukazawa I, Uchida N, Uchida E et al. Enantioselective determination of azelnidipine in human plasma using liquid chromatographytandem mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci., 2007; 852(1-2): 389-97. doi: 10.1016/j.jchromb.2007.01.050, PMID 17350354.
6. Zou JJ, Ji HJ, Zhou XH, Zhu YB, Fan HW, Xiao DW et al. Determination of azelnidipine by LC–ESI-MS and its application to a pharmacokinetic study in healthy Chinese volunteers. Pharmazie, 2008; 63(8): 568-70. PMID 18771003.
7. Ding L, Li L, Ma P. Determination of azelnidipine in human plasma by liquid chromatography–electrospray ionization-mass spectrometry. J Pharm Biomed Anal., 2007; 43(2): 575-9. doi: 10.1016/j.jpba.2006.07.011, PMID 16920318.
8. Patel JK, Patel NilamK. Validated stability-indicating RP-HPLC method for the simultaneous determination of azelnidipine and olmesartan in their combined dosage form. Sci Pharm., 2014; 82(3): 541-54. doi: 10.3797/scipharm.1312-14, PMID 25853066.
9. International Conference on Harmonization (ICH) of Technical Requirements for the Registration of Pharmaceuticals for Human Use, Validation of Analytical Procedure: Methodology, November 1996; (ICH – Q 2B): 1-8.
10. Bonfilio RBDAM, De Araujo MB, Salgado HRN. Recent applications of analytical techniques for quantitative pharmaceutical analysis: a review. WSEAS Trans Biol Biomed, 2010; 7(4): 316-38.
11. Mendham J, Denny RC, Thomas M, Vogel‟s text book of Quantitative Chemical Analysis. 6th ed. Pearson Education Limited, 2008; 29-39.
12. Zilker M, Sörgel F, Holzgrabe U. A systematic review of the stability of finished pharmaceutical products and drug substances beyond their labeled expiry dates. J Pharm Biomed Anal, 2019; 166: 222-35. doi: 10.1016/j.jpba.2019.01.016, PMID 30660807.
13. Chatwal GR, Anand SK. Instrumental methods of Chemical Analysis. 11th ed. Mumbai: Himalaya Publishing House, 2005; 1.1-2, 2.108-9: 2.151-2.153.
14. Kasture AV, Wadodkar SG, Mahadik KR, More HN. Pharmaceutical analysis instrumental methods; Nirali prakashan. 12th ed., 2005; 148-56.
15. Skoog D, Leqary J. Principle of instrumental analysis; Thomson Asia Pvt Ltd. Singapore. 54th ed., 2004; 3-8.
16. Skoog D, Holler F, Timothy A, Nieman N. Principles of instrumental analysis. 4th ed. London: Saunders College Publications, 1992; 1-2: 338-40.
17. Settle F; Handbook of Instrumental Techniques of Analytical Chemistry. 1st ed., 2004; 19-21: 609-17.
18. Corners KA. Textbook of pharmaceutical analysis, A wiley interscience publication. 1st ed., 1967; 475-8.
19. Kasture AV, Wadodkar SG, Mahadik KR, More HN. Textbook Pharm Anal. 13th ed., 2005; 1; II, Nirali prakashan, 47-56.
20. British Pharmacopoeia, 1993; II: 180-90.
21. Kakde RB, Kasture AV, Wadodkar SG. Indian J Pharm Sci., 2002; 64(1): 24-7.
22. Dyade GK, Sharma AK. Indian Drugs, 2001; 38(2): 75-8.
23. Sethi PD. Qualitativie Analysis of drugs in Pharmaceutical Formulations. 3rd ed., 1997; 182-4.
24. James S, James B. C; Encyclopedia of pharmaceutical technology. Vol. I. New York: Marcel Dekker, Inc., 1998; 217-24.
25. Sandy L. HPLC by open learning. London: John wiley & sons, 1991; 30-45.
26. Lough WJ, Wainer IWW. HPLC fundamental principles and practices. Blackie Academic & professional, 1991; 52-67.
27. Christian GD. Analytical chemistry. 4th ed. United Kingdom: John Wiley & Sons, 1986; 1-6.
28. Meyer Veronica R. Practical high-performance liquid chromatography. 2nd ed. 26(27). London: John wiley & sons, 1993; 40, 222: 246, 258.
29. Chatwal GR, Anand SK. Instrumental method of chemical analysis; Himalaya Publishing House. 11th ed., 2005; 2.634-8.
30. Scott RPW. Liquid chromatography for the analyst. Chromatogr Sci S., 1991; 1-30.
31. Westen A. HPLC and ce – principles and practice. Academic press, 1997; 1-21.

32. Wellington K, Scott LJ. Azelnidipine. Drugs, 2003; 63(23): 2613–2621: 2613-21. discussion 2623. doi: 10.2165/00003495-200363230-00004, PMID 14636080.
33. Sada T, Saito H. Pharmacological profiles and clinical effects of azelnidipine, a long-acting calcium channel blocker. Nihon Yakurigaku Zasshi, 2003; 122(6): 539-47. doi: 10.1254/fpj.122.539, PMID 14639008, 539-547.
34. PubChem. Azelnidipine [internet]. Available from: https://pubchem.ncbi.nlm [cited Jan 4 2022]. Available from: http://nih.gov/compound/Azelnidipine. Available from: https://pubchem.ncbi.nlm.nih.gov/compound/65948.
35. Matsumoto S. Effect of olmesartan / azelnidipine combination tablets on ambulatory blood pressure and cognitive function in the post-stroke patients. Stroke, 2020; 51(1): AWP458–AWP458. doi: 10.1161/str.51.suppl_1.WP458.
36. Shimada K, Ogihara T, Saruta T, Kuramoto K, REZALT Study Group. Effects of Combination olmesartan Medoxo mil Plus azelnidipine versus monotherapy with either Agent on 24-hour Ambulatory Blood Pressure and Pulse Rate in Japanese Patients with Essential hypertension:additional Results from the REZALT Study. Clin Ther., 2010; 1(5)(5): 861-81. doi: 10.1016/j.clinthera.2010.04.020, PMID 20685495, 861-881.
37. Saito I, Kushiro T, Zenimura N, Matsushita Y, Sagawa K, Hiramatsu K, et al. Olmesartan medoxomil and azelnidipine therapy in patients with hypertension and chronic kidney disease in Japan. J Nephrol, 2012; 25(5): 699-708. doi: 10.5301/jn.5000043, PMID 22020401, 699-708.
38. Daikuhara H, Kikuchi F, Ishida T. The combination of olmesartan and a calcium channel blocker (azelnidipine) or candesartan and a calcium channel blocker (amlodipine) in type 2 diabetic hypertensive patients: the OLCA study. Diabetes Vasc Dis Res., 2012; 1: 9(4). doi: 10.1177/1479164112447310, pages-280-286.
39. Patel JK, Patel NK. Validated stability-indicating RPHPLC method for the simultaneous determination of azelnidipine and olmesartan in their combined dosage form. Sci Pharm., 2014; 82(3): 541-54. doi: 10.3797/scipharm.1312-14, PMID 25853066, 541-554.
40. Amin A, Saad M, Amin M. Simultaneous determination of azelnidipine and olmesartan medoxomil in pharmaceutical dosage forms by UFLC method. J Pharm Sci Technol, 2017; 11: 6(2): 69-74.
41. Adepu R, Neelima PLMK, et al. A novel method for the simultaneous determination of azelnidipine and olmesartan in Human plasma by using liquid chromatographyelectro spray ionization tandem mass spectrometry and application to a pharmacokinetic study. Int J Pharm., 2017; 11: 7(3): 111-124.
42. Prabhakar D, Sreekanth J, Jayaveera KN, et al. Method development and validation of azelnidipine by RP-HPLC. Int J ChemTech Res., 2017; 10(10): 418-423.
43. Modi J, Patel SK, Parikh N, et al. Stability indicating analytical method development and validation for estimation of azelnidipine. World J Pharm Res., 2016; 5(2): 831-847.
44. Gore M, Dabhade PS. RP-HPLC method development and validation of azelnidipine. Int J Pharm Sci Res., 2016; 7(12): 5111-5114.
45. Ubale Sneha KMS, Bhavna H et al. Development and Validation of RPHPLC method for quantification of azelnidipine in Tablet. Int J Creat Res Thoughts, 2021; 9(7): 797-802.
46. Bharti TM, Rajput R, et al. Analytical method development and validation of assay for olmesartan medoxomil in formulated product by reverse phase ultraperformance liquid chromatography. Eur J Biomed Pharm Sci., 2016; 3(2): 215-222.
47. Chavda N, Kumar S. A Review article on Analytical Method Development for the combination of azelnidipine and telmisartan. Asian J Pharm Res Dev., 2021; 11(3): 227-34. doi: 10.52711/2231-5675.2021.00040, 227-240.
48. Kumar M, Chandra U, Garg A, Gupta P. A stability indicating RP-HPLC method validation for simultaneous estimation of azelnidipine and telmisartan in a fixed-dose combination. Int J Pharm Sci Drug Res., 2021; 13(3): 288-94. doi: 10.25004/IJPSDR.2021.130308, 288-294.
49. Ponnekanti K, Sunitha K. Development of HPLC stability demonstrating methodology for quantifying azelnidipine and telmisartan in tablets and bulk types: validation following ICH directives. Int J Appl Pharm., 2021; 13(5): 298-305. doi: 10.22159/ijap.2021v13i5.42099, 298-305.