IJBPAS, December, 2025, 14(12): 6187-6199

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

DEVELOPMENT AND VALIDATION OF A UV SPECTROSCOPIC METHOD FOR OLANZAPINE ANALYSIS IN PHARMACEUTICAL DOSAGE FORMS – A MULTIVARIATE CALIBRATION APPROACH

MYTHREYAN T, SEETHARAMAN R* AND MANIKANDAN K

Department of Pharmaceutical Analysis, SRM College of Pharmacy, SRM Institute of Science and Technology, Kattankulathur, Chengalpattu District, Tamil Nadu - 603203, India

***Corresponding Author: Dr. Seetharaman R: E Mail: seetharr@srmist.edu.in; seerampharm@gmail.com**

Received 15th July 2024; Revised 20th Sept. 2024; Accepted 14th Nov. 2024; Available online 1st Dec. 2025

https://doi.org/10.31032/IJBPAS/2025/14.12.8344

ABSTRACT

The objective of the current study is to establish a multilinear regression procedure wholly precise, straightforward, sensitive, and legitimate for the assessment of Olanzapine in commercial tablet dose forms. The multivariate calibration approach was put into effect through generating equations through linear regression analysis, exploiting the correlated relationship amongst absorbance as well as concentration at five unique wavelengths each of which were meticulously selected. The maximum wavelength of olanzapine turned out to be the value of 226 nm. The outcomes were analysed through statistical analysis. A trend towards linearity emerged throughout the concentration range of 7-13 µg mL^{-1}, with acceptable regression coefficient. The relative standard deviation scores for intra-day and inter-day precision have been established to be 0.42 and 0.39, as well. The assay was performed and resulted in an assortment between 99.20 % to 99.80 % w/w.

Keywords: Olanzapine, Antipsychotic, UV spectrophotometry, Multivariate calibration, Assay, ICH guidelines

INTRODUCTION

Olanzapine is a thienobenzodiazepine synthetic derivative with antipsychotic, antiemetic, and antinausea properties and its structure is presented in **Figure 1**. Olanzapine collaborates to the following receptors with a high affinity that acts as a

specialized monoaminergic antagonist. muscarinic M1-5, histamine H1, serotonin, dopamine, histaminergic, and alpha-1-adrenergic receptors. It faintly binds to beta-adrenergic, benzodiazepine, and type A gamma-aminobutyric acid receptors. This medication appears to block 5-HT2 and 5-HT3 serotonin receptors, which has antinausea and antiemetic properties. Olanzapine's antipsychotic effect is thought to be mediated through antagonizing dopamine D2 receptors, which have quick ligand-receptor dissociation kinetics and reduce extrapyramidal symptoms. Olanzapine may also increase appetite. Molecular formula and molecular weight were found to be $C_{17}H_{20}N_4S$, and 312.4 g/mol **[1]**. Literature survey reveals various methods for UV **[2-6]**, HPLC **[7-14]**, HPTLC **[15-21]** for estimating the olanzapine.

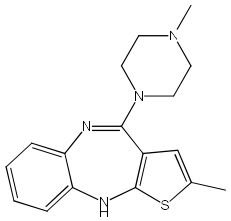

Figure 1: Structure of olanzapine

The suggested multi variate calibration (MVC) technique provides higher confidence in results as it directly evaluates Olanzapine and has been attested with greater accuracy and precision than a classical UV-Visible assay. This technique is more cost-effective, direct, and rapid than other methods and can be used for bulk drugs and various dosage forms. This multivariate standardization method simplifies the individual result and converts it into an "m" value as a reliant variable. Within optimized conditions, this analytical technique would provide excellent sensitivity, resolving power, expeditiousness, and cost-effectiveness for a validated quantification of olanzapine. The absorbance of an analyte (X), i.e., Olanzapine, is scanned at 5 different absorbances (λ = 222, 224, 226, 228, and 230nm); the following formula can then be applied for any preferred wavelength.

The following equations can be generated for each chosen wavelength if the absorption of a sample(x) is determined at several wavelengths (λ), namely at 222, 224, 226, 228, and 230 nm **[22]**.

$A_{\lambda 222} = a \times C_x + k_1$ …… (1)

$A_{\lambda 224} = b \times C_x + k_2$ …… (2)

$A_{\lambda 226} = c \times C_x + k_3$ ……..…..... (3)

$A_{\lambda 228} = d \times C_x + k_4$ ……...…. (4)

$A_{\lambda 230} = e \times C_x + k_5$ ……..……...…. (5)

Whereas,

- A_λ = Absorbance of the sample;
- a, b, c, d, e = Slope of the straight regression functions of a sample;
- k_1, k_2, k_3, k_4, k_5 = Intercept of the straight regression;
- C_x = Concentration of the sample

Rearranging the five equations above results in:

$A_T = a \times C_x + b \times C_x + c \times C_x + d \times C_x + e \times C_x + K_T$……. (6)

Equation (6) might be modified to read:

$A_T = C_x (a + b + c + d + e) + K_T$ ……. (7)

Whereas,

- A_T = Sum of the absorbances acquired
- K_T = Sum of intercepts of regression equation

Equation can be used to determine the sample's (X) concentration in a solution.

$$Cx = \frac{A_T - K_T}{(a+Ab+c+d+e)} \quad \text{…… (8)}$$

MATERIALS AND METHODS

Materials

Analytical grade materials were used for all "chemicals and reagents" in this experiment. Each film-coated tablet in a marketed formulation contains 10 mg of Olanzapine from two brands that were selected and purchased from a retail pharmacy.

Instrumentation

• UV- Visible double beam spectrophotometer (LABINDIA UV 3092 model)

• Soniclean Sonicator (model 106 T, Thebarton, Australia)

• Analytical balance (AS 245, Mettler Toledo, India)

METHOD DEVELOPMENT

Solubility

In Methanol, Olanzapine was completely soluble. So, Methanol was employed as the study's solvent throughout the research.

Reference Samples

Olanzapine was kindly supplied by Ideal Analytical and Research Institution (Pondicherry, India).

Preparation of Standard solution

The standard solution of Olanzapine reference standard was prepared by dissolve 50 mg of drug in 50 ml of the solvent to acquire 1000 µg mL^{-1} with the use of sonicator. To achieve concentrations in ranges from 7 to 13 µg mL^{-1}, the solution is diluted.

Preparation of sample solution

Olanzapine tablet were weighed, put to a mortar, and ground into a powder. Next, accurately measured amounts of 5 mg of olanzapine were fully dispersed in 50 millilitres of methanol with a sonicator, filtrated, and diluted for further research.

Selection of wavelength for MVC

The working standardized solutions of olanzapine had been checked against methanol, a blank solution with maximum absorbance at 226 nm, over the wavelength that range from 200 to 400 nm. The wavelength for the MVC method was therefore in the vicinity of these absorption maxima, or 222, 224, 226, 228, and 230 nm.

METHOD VALIDATION

The prepared method was validated per ICH guidelines for accuracy, precision, linearity **[23]**.

Linearity

Olanzapine stock solution was dissolved in the solvent to produce concentrations between 7-13 µg mL^{-1} . The amplitudes of these solutions were measured around 226 nm, i.e., 222, 224, 226, 228, and 230 nm, in the spectrum mode, to reduce instrumental fluctuations and increase correlation.

Limit of Detection and Limit of Quantification

According to the slope of the calibration curve and the standard deviation of the responses for a specific wavelength, the following formulas were used to determine the limit of detection (LOD) and limit of quantification (LOQ) for olanzapine.

$$\text{LOD} = \frac{3.3 \text{ x standard deviation}}{\text{Slope}} \quad \text{..... (10)}$$

$$\text{LOQ} = \frac{10 \text{ x standard deviation}}{\text{Slope}} \quad \text{..... (11)}$$

Precision

Intra-day and inter-day precisions were measured three times daily and three distinct days (inter-day precision) and (intra-day precision) accordingly, using replicate measurements on homogeneous solutions containing 10 µg mL^{-1}.

Accuracy

At 80, 100, and 120 percent of the pre-analyzed sample solutions, the methodology's accuracy for olanzapine was tested, and the recovery values' percentages were estimated.

Assay

Accurately weigh 50 mg of olanzapine in a quantity of olanzapine pills, then add 50 ml of methanol to it and sonicate it for 20 minutes. Add sufficient methanol and make up to 100 mL. To make a solution with a 10 µg mL^{-1} concentration, the solution that was obtained above has been filtrated and diluted with methanol.

RESULTS AND DISCUSSION

A 10 µg mL^{-1} solution at 226 nm, exhibits highest amplitude with methanol as the solvent and presented in **Figure 2**.

Linearity

As demonstrated in **Figure 3**, the linearity was measured at 222, 224, 226, 228 and 230 nm for various concentrations between 7 to 13 µg mL^{-1}. **Table 1** presents the observed results in tabular form. In the chosen concentration range, it was discovered that all of the standard curve were linear. **Figures 4-8** and **Table 2** provide the calibration graphs, and regression analysis, respectively.

Precision

Figure 9 displays the Olanzapine system precision spectra. **Figure 10** displays the interday Olanzapine precision spectra. The intraday precision spectra were represented in **Figure 11** for Olanzapine. Olanzapine's interday and intraday system precision RSDs were calculated. It was discovered to be less than 2%, demonstrating the accuracy of the approach method. At the chosen wavelengths, the SD and % RSD values

were also determined and are shown in **Table 3**. Comparing the values acquired from other accuracy approaches, the suggested method exhibits good precision.

Accuracy

At 80, 100, and 120%, olanzapine accuracy tests were conducted. The Olanzapine overlay spectra are shown in **Figure 12**. **Table 4** displays the accuracy results, and it was determined that the results were within acceptable bounds.

Assay of marketed formulations

The recommended UV method was used to investigate the quantity of olanzapine in tablet formulation. The spectrum of tablet was obtained for three replicates. After extraction and filtration, there was no appreciable decrease in the pharmaceutical formulation's excellent analytical recovery values. The results are provided in the **Table 5** for olanzapine, which demonstrates that the new strategy out performs the earlier ones.

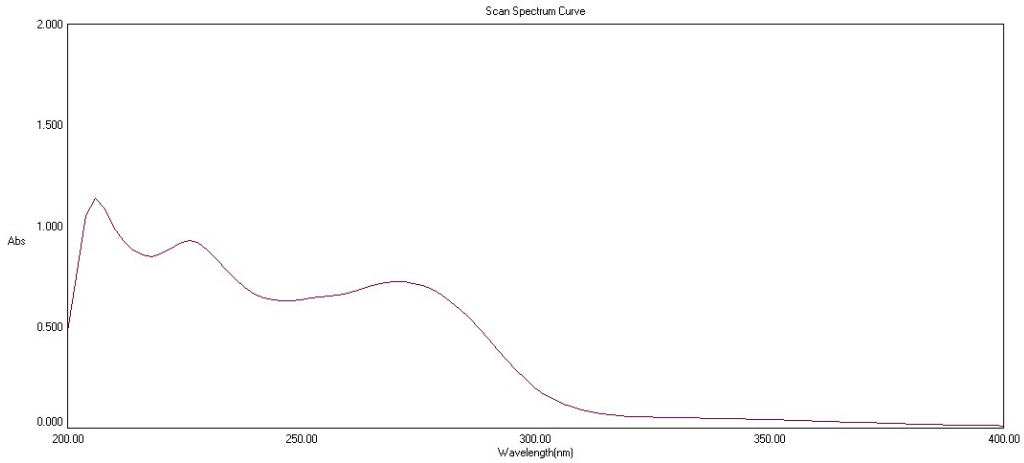

Figure 2: UV spectrum of 13 µg mL^{-1} olanzapine showing λ_{max} at 226 nm

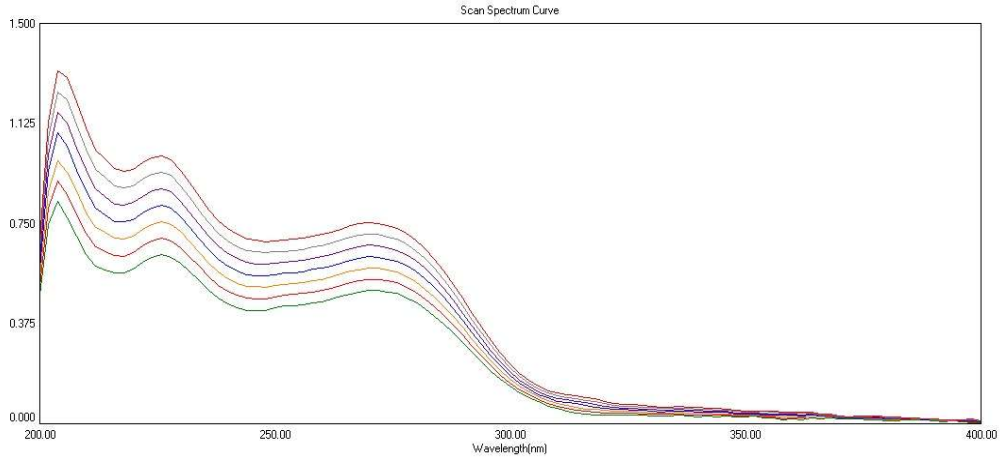

Figure 3: Linearity spectrum of Olanzapine (7 to 13 µg mL^{-1}) using methanol as blank

Table 1: Multivariate UV calibration data at five selected wavelengths

Concentration (µg mL^{-1})	Absorbance at				
	222nm	224nm	226nm	228nm	230 nm
7	0.604	0.621	0.635	0.623	0.594
8	0.669	0.681	0.696	0.681	0.657
9	0.727	0.744	0.758	0.739	0.715
10	0.787	0.812	0.820	0.797	0.773
11	0.850	0.870	0.882	0.865	0.831
12	0.913	0.932	0.943	0.928	0.884
13	1.004	0.990	1.004	0.990	0.942

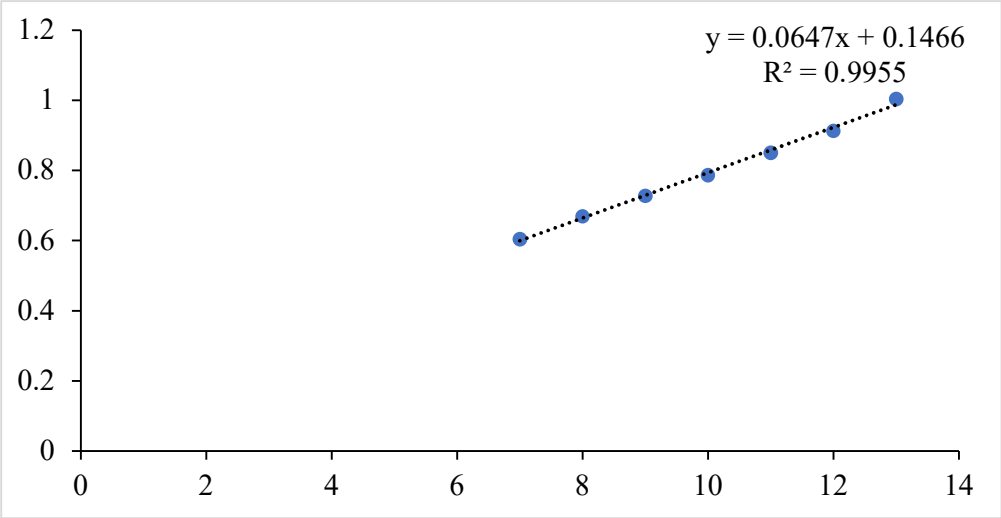

Figure 4: Calibration curve at 222 nm

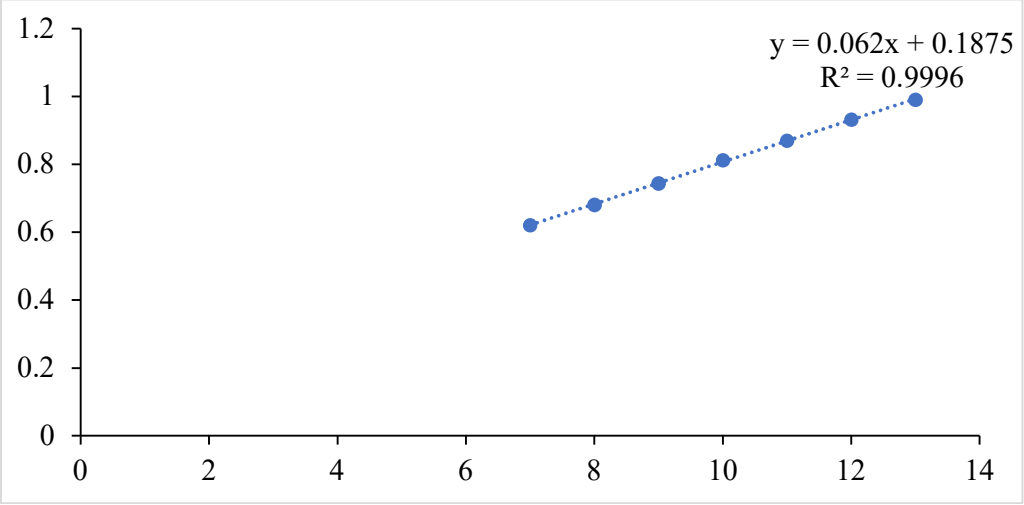

Figure 5: Calibration curve at 224 nm

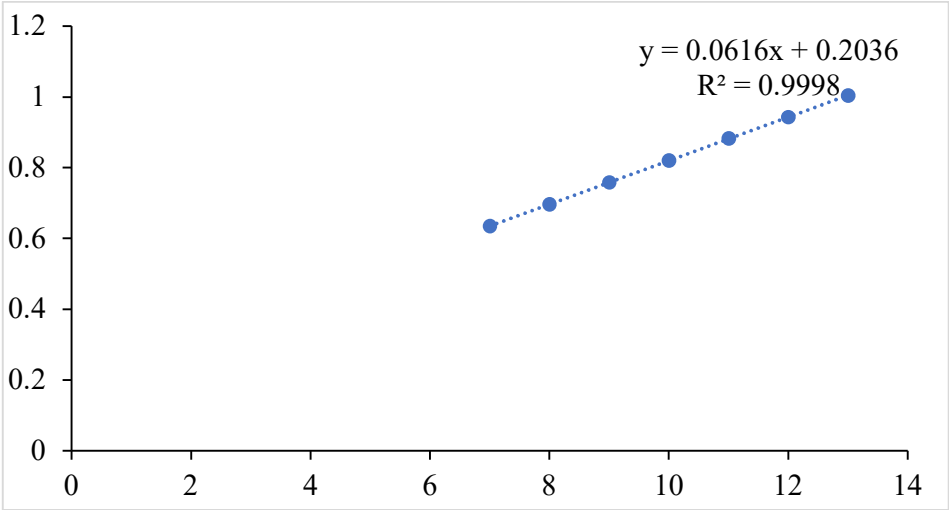

Figure 6: Calibration curve at 226 nm

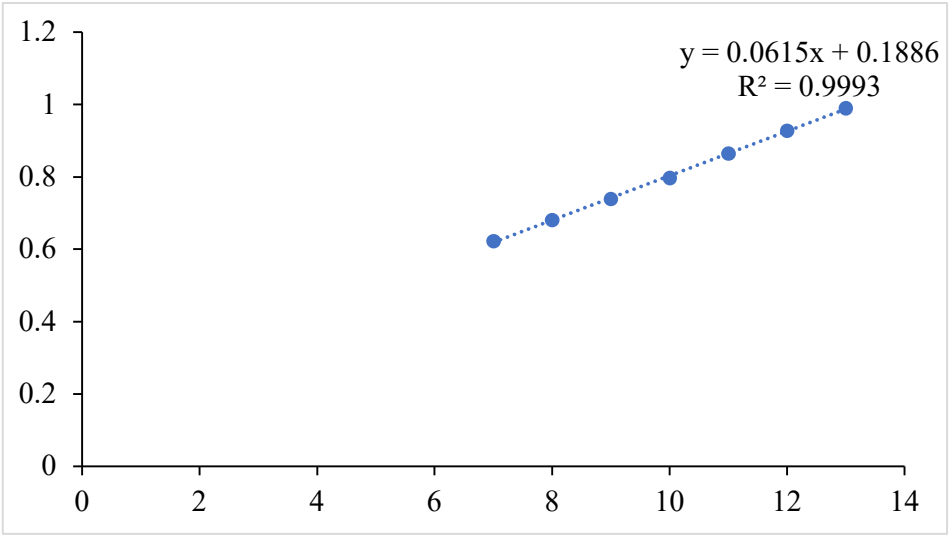

Figure 7: Calibration curve at 228 nm

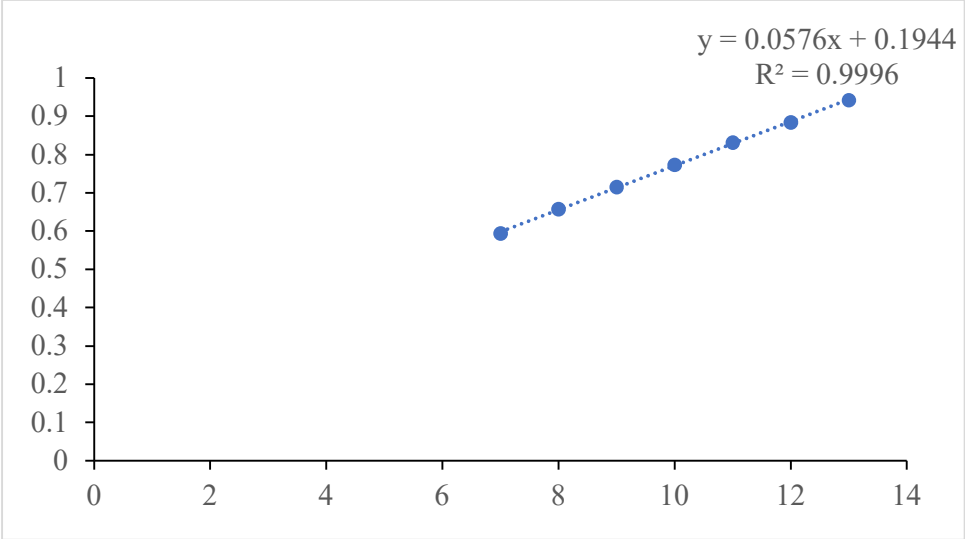

Figure 8: Calibration curve at 230 nm

Table 2: Linearity data with LOD and LOQ at selected five wavelengths

Wavelength (nm)	Regression equation	R^2	LOD ($\mu g\ mL^{-1}$)	LOQ ($\mu g\ mL^{-1}$)	% RSD
222	y = 0.0647x + 0.1466	0.9955	0.5227	1.5840	1.2912
224	y = 0.062x + 0.1875	0.9996	0.1521	0.4611	0.3540
226	y = 0.0618x+0.2019	0.9999	0.0583	0.1768	0.1332
228	y = 0.0615x + 0.1886	0.9993	0.2056	0.6233	0.4769
230	y = 0.0576x + 0.1944	0.9996	0.1604	1.1746	0.3636

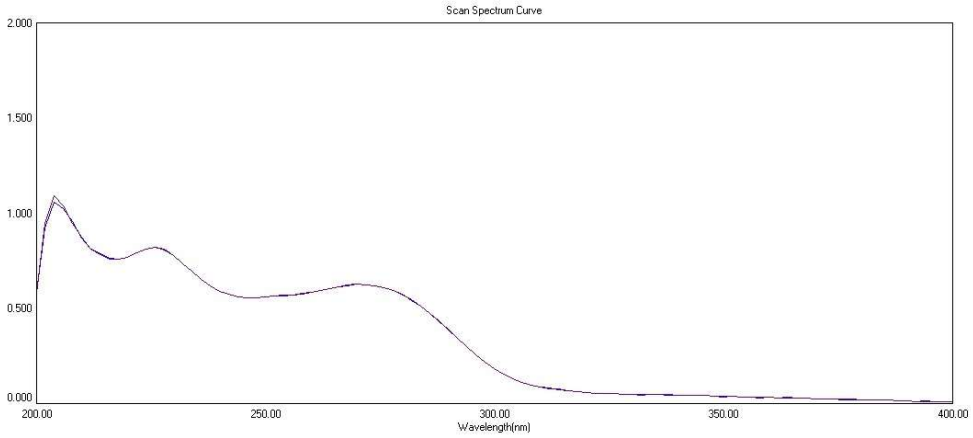

Figure 9: System precision overlay spectra of Olanzapine

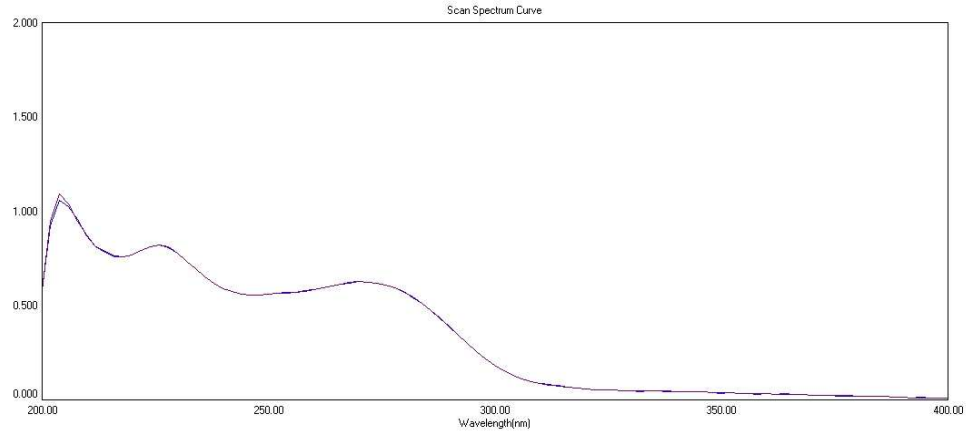

Figure 10: Interday precision overlay spectra of Olanzapine

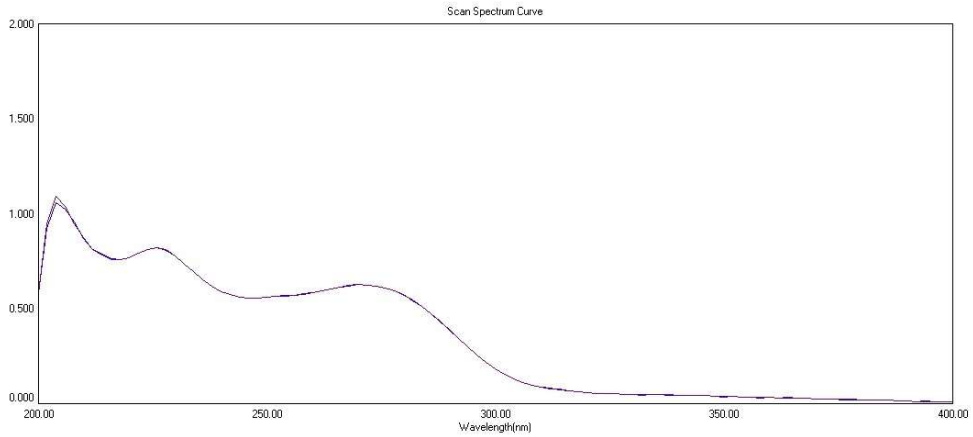

Figure 11: Intraday precision overlay spectra of Olanzapine

Table 3: System precision, Intraday and Interday precision data for the prepared method of Olanzapine

	System Precision	Interday and Intraday Precision		
	Amplitude of Standard (10 µg mL^{-1})	% Recovery of sample equivalent to 10 µg mL^{-1} of sample		
		Day 1	Day 2	Day 3
1	3.969	99.45	98.69	99.58
2	3.972	98.95	98.22	99.38
3	3.985	99.58	99.48	99.57
4	4.002	99.47	99.57	99.58
5	3.981	99.57	98.55	99.66
6	3.965	98.55	98.27	99.84
Mean	3.979	99.26	99.89	99.60
SD	0.014	0.42	0.59	0.15
% RSD	0.34	0.42	0.59	0.15

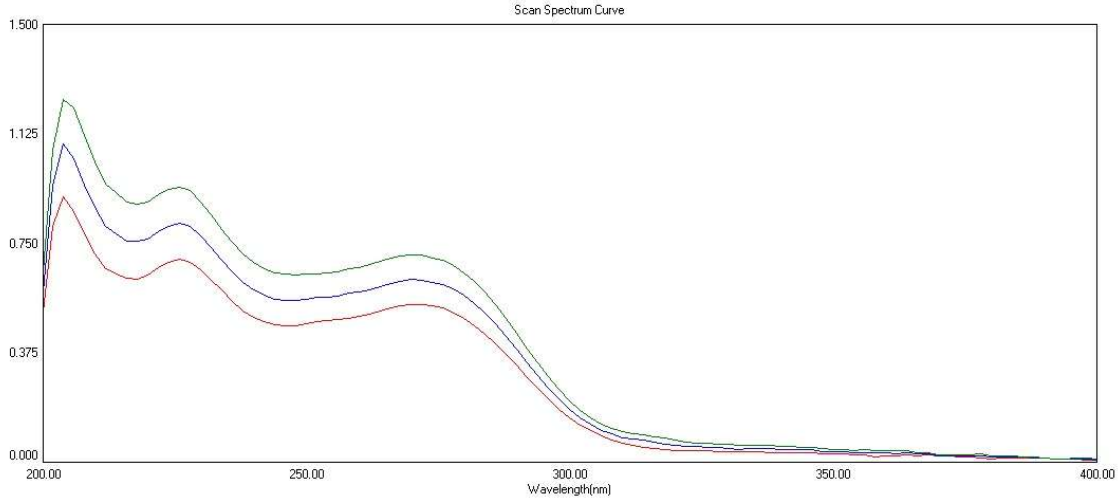

Figure 12: Overlay spectra of accuracy of Olanzapine at 80, 100, 120 % spiking

Table 4: Results of recovery Studies

Wavelength (nm)	Amount present (µg mL^{-1})	Amount added (µg mL^{-1})	Absorbance	Amount recovered (µg mL^{-1})	% Recovery
222nm	4	4	0.669	3.97	99.25
		6	0.787	5.97	99.50
		8	0.913	8.01	100.13
224nm	4	4	0.621	4.02	100.50
		6	0.812	5.95	99.17
		8	0.932	7.98	99.75
226 nm	4	4	0.696	3.98	99.50
		6	0.820	6.03	100.50
		8	0.943	7.95	99.38
228nm	4	4	0.681	3.99	99.75
		6	0.797	6.01	100.17
		8	0.928	7.98	99.75
230nm	4	4	0.657	3.97	99.25
		6	0.773	5.96	99.33
		8	0.884	7.97	99.63

Table 5: Assay results for marketed formulation of oLANZAPINE

Marketed formulation	Label claim (mg)	Mean ± SD (n=3)	% RSD
Batch - 1	10	99.50 ± 0.30	0.17
Batch - 2	10	99.07 ± 0.38	0.22

CONCLUSION

The evaluation of the validation parameters in accordance with ICH Q2 (R1) guidelines helped to validate this newly created method of MVC technique using UV spectrophotometry. The study shows this method is affordable, accurate, and non-destructive, It can be utilized for routine analysis, quality control procedures, and the examination of olanzapine both in bulk and pharmaceutical dose forms in industry. The proposed method for determining Olanzapine in its tablet formulation was shown to be sensitive, accurate, precise, and repeatable. We strongly advise using this method for routine analysis of Olanzapine in pharmaceutical formulations because it is more useful and dependable than the other UV spectrophotometric methods.

ETHICAL STATEMENT

In the study experiments, neither humans nor animals were used as subjects.

ACKNOWLEDGMENTS

For supplying the authors with a variety of reprographic sources for this study, the management of SRM Institute of Science and Technology and SRM College of Pharmacy at SRM Institute of Science and Technology, Kattankulathur, is to be acknowledged.

CONFLICTS OF INTEREST

There are no financial conflicts of interest that might affect this content.

REFERENCES

[1] National Center for Biotechnology Information. PubChem compound summary for CID 135398745, olanzapine; 2023 [cited Sep 3, 2023]. Available from: https://pubchem.ncbi.nlm.nih.gov/compound/Olanzapine.

[2] Firdous S, Aman T, Nisa U. Determination of olanzapine by UV spectrophotometry and nonaqueous titration. J Chem Soc Pak. 2005;27:163-7.

[3] Joseph E, Balwani G, Nagpal V, Reddi S, Saha R. Validated UV spectrophotometric methods for the estimation of olanzapine in bulk, pharmaceutical formulations and preformulation studies *Br*. 2015;6(3):181-190. doi:10.9734/BJPR/2015/16361.

[4] Olajire A. Adegoke, Olusegun E. Thomas, Damilola M. Makanjuola, Oyeronke O. Adewole, Spectrophotometric determination of olanzapine after condensation with p-dimethylaminobenzaldehyde, Journal of Taibah University for Science, Volume 8, Issue 3, 2014, Pages 248-257, ISSN 1658-3655,https://doi.org/10.1016/j.jtusci.2014.03.007.

[5] Salama FM, Attia KAM, Said RAM, El-Olemy A, Abdel-raoof AM.

Spectrophotometric Determination of Olanzapine in the Presence of its Acidic Degradation Product; Application of Kinetic Study [Internet]. Vol. 7, Analytical Chemistry Letters. Informa UK Limited; 2017. p. 663–75. Available from: http://dx.doi.org/10.1080/22297928.2017.1372207.

[6] Rajendraprasad N, Basavaiah K. Determination of olanzapine by spectrophotometry using permanganate Vol. 45, Brazilian Journal of Pharmaceutical Sciences. FapUNIFESP (SciELO); 2009. p. 539–50. Available from: http://dx.doi.org/10.1590/S1984-82502009000300020.

[7] Rani P, Sekaran b, Chandra. Development of HPLC method for the determination of olanzapine in bulk and dosage forms. Int J Pharm Tech Res. 2008;1.

[8] Raggi MA, Casamenti G, Mandrioli R, Izzo G, Kenndler E. Quantitation of olanzapine in tablets by HPLC, CZE, derivative spectrometry and linear voltammetry. J Pharm Biomed Anal. 2000 November;23(6):973-981. doi:10.1016/s0731-7085(00)00382-4, PubMed: 11095298.

[9] Basavaiah K, Rajendraprasad N, Vinay KB. Isocratic High-Performance Liquid Chromatographic Assay of Olanzapine: Method Development and Validation [Internet]. Vol. 2014, ISRN Analytical Chemistry. Hindawi Limited; 2014. p. 1–6. Available from: http://dx.doi.org/10.1155/2014/616941.

[10] Cui D, Li Y, Lian M, Yang F, Meng Q. Development of a simple and stability-indicating RP-HPLC method for determining olanzapine and related impurities generated in the preparative process [Internet]. Vol. 136, The Analyst. Royal Society of Chemistry (RSC); 2011. p. 3149. Available from: http://dx.doi.org/10.1039/C1AN15155J.

[11] Tantawy MA, Hassan NY, Elragehy NA, Abdelkawy M. Simultaneous determination of olanzapine and fluoxetine hydrochloride in capsules by spectrophotometry, TLC-spectrodensitometry and HPLC. Vol. 4, Journal of Advanced Research. Elsevier BV; 2013. p.173–80. Available from: http://dx.doi.org/10.1016/j.jare.2012.05.004.

[12] Rp-HPLC method development, validation and stability studies of olanzapine and samidorphan in combined dosage forms. J Pharm Neg Results. 2022:8691-8713. doi:10.47750/pnr.2022.13.S09.1021.

[13] Reddy BV, Reddy KVNS, Sreeramulu J, Kanumula GV. Simultaneous determination of olanzapine and fluoxetine by HPLC. Chromatographia.

2007;66(1-2):111-114. doi:10.1365/s10337-007-0257-z.

[14] Pathak A, Rajput SJ. Development of a Stability-Indicating HPLC Method for Simultaneous Determination of Olanzapine and Fluoxetine in Combined Dosage Forms [Internet]. Vol. 47, Journal of Chromatographic Science. Oxford University Press (OUP); 2009. p. 605–11. Available from: http://dx.doi.org/10.1093/chromsci/47.7.605

[15] Ahmed R, Bhusari KP, Tajne MR, Khan SA. Development of a stability indicating HPTLC method for the estimation of olanzapine in pharmaceutical dosage forms [Internet]. Vol. 98, Journal of the Indian Chemical Society. Elsevier BV; 2021. p. 100215. Available from: http://dx.doi.org/10.1016/j.jics.2021.100215.

[16] Patel RB, Patel MR, Bhatt KK, Patel BG. Development and validation of an HPTLC method for determination of olanzapine in formulations. J AOAC Int. 2010 May-June;93(3):811-819. doi:10.1093/jaoac/93.3.811, PubMed: 20629381.

[17] Kothari, Charmy & Suhagia, Dr & Shah, N & Patel, Dr Rajendra & Patel, Neha Maheshbhi. (2008). Stability-indicating Simultaneous HPTLC Method for Olanzapine and Fluoxetine in Combined Tablet Dosage Form. Indian journal of pharmaceutical sciences. 70. 251-5. 10.4103/0250-474X.41469.

[18] Ahmad, Reema & Bhusari, Kishore & Tajne, Madhukar & Khan, Shah. (2021). Development of a stability indicating HPTLC method for the estimation of olanzapine in pharmaceutical dosage forms. Journal of the Indian Chemical Society. 98. 100215. 10.1016/j.jics.2021.100215.

[19] Shah CR, Suhagia BN, Shah NJ, Patel DR, Patel NM. Stability-indicating simultaneous HPTLC method for olanzapine and fluoxetine in combined tablet dosage form. Indian J Pharm Sci. 2008;70(2, March-April):251-255. doi:10.4103/0250-474X.41469, P. 251. Gale Academic OneFile.

[20] Patel, S.K., Patel, N.J., Patel, P.U. et al. Validation of a Stability-Indicating HPTLC Method for Analysis of Duloxetine Hydrochloride in Capsule Dosage Form. Separation and Analysis of Duloxetine Hydrochloride and Olanzapine in a Synthetic Mixture. JPC-J Planar Chromat 22, 121–126 (2009). https://doi.org/10.1556/JPC.22.2009.2.8

[21] Patel S, Patel N. Simultaneous RP-HPLC and HPTLC estimation of fluoxetine hydrochloride and olanzapine in tablet dosage forms [Internet]. Vol. 71, Indian Journal of Pharmaceutical Sciences. OMICS Publishing Group;

2009. p. 477. Available from: http://dx.doi.org/10.4103/0250-474X.57306.

[22] Kokilambigai KS, Seetharaman R, Kavitha J, Sowndaravel P, Lakshmi KS. Multivariate calibration technique for the spectrophotometric quantification of zaleplon in bulk and pharmaceutical formulations. J Pharm Sci Res. 2017;9(6):824-9.

[23] ICH. Harmonized tripartite guideline, validation of analytical procedures: text andmethodology. Vol. Q2(R1). Geneva, Switzerland; 2005.