Journal of Applied Pharmaceutical Research
Volume 13 Issue 1, Year of Publication 2025, Page 112 – 122
DOI: https://doi.org/10.69857/joapr.v13i1.782

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Research Article

JOURNAL OF APPLIED PHARMACEUTICAL RESEARCH | JOAPR
www.japtronline.com ***ISSN: 2348 – 0335***

NOVEL RP-HPLC METHOD DEVELOPMENT AND VALIDATION FOR PRECISE QUANTIFICATION OF PROCHLORPERAZINE MALEATE IN PHARMACEUTICAL DOSAGE FORMS

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Article Information
Received: 17th November 2024
Revised: 15th January 2025
Accepted: 11th February 2025
Published: 28th February 2025

Keywords
Prochlorperazine Maleate, RP-HPLC, Method Validation, Pharmaceutical Dosage Forms, Analytical Method Development, Quality Control

ABSTRACT

Background: Prochlorperazine Maleate, a piperazine phenothiazine derivative, exhibits strong antiemetic and antipsychotic properties. However, existing analytical methods for its quantification in pharmaceutical formulations often face limitations regarding sensitivity, specificity, and accuracy. Many conventional techniques involve extensive sample preparation and prolonged analysis times, making them less feasible for high-throughput quality control. This study developed and validated a novel, precise, and highly sensitive reverse-phase high-performance liquid chromatography (RP-HPLC) method for Prochlorperazine Maleate quantification to overcome these challenges. **Methodology:** An RP-HPLC method was established using an Agilent Zorbax Bonus-RP column (250 × 4.6 mm, 5 µm) with a mobile phase of 0.1% formic acid and acetonitrile (70:30). The detection was performed at 258 nm using a diode array detector. Method validation followed ICH guidelines, assessing linearity, precision, accuracy, robustness, and sensitivity across a 100–150 µg/mL concentration range. **Results and Discussion:** The method displayed strong linearity (R^2 = 0.999). The LOD and LOQ were 1.76 µg/mL and 5.35 µg/mL, respectively. High precision (%RSD < 2%) and recovery rates (99–101%) confirmed accuracy. Robustness was established through consistent retention time and peak symmetry. **Conclusion:** This validated RP-HPLC method is reliable, sensitive, and cost-effective, making it ideal for routine pharmaceutical quality control and future stability studies.

INTRODUCTION

A multitude of new pharmaceuticals are discussed extensively; consequently, regulations and analytical techniques for certain drugs may be absent from the pharmacopoeias. It is thus essential to develop more straightforward and contemporary analytical techniques for pharmaceuticals. Numerous drugs are introduced into the pharmaceutical market annually, which may involve either substantial structural alterations of current medications or entirely novel molecules (Figure 1). Prochlorperazine Maleate is the maleate salt of prochlorperazine, a chemically manufactured piperazine phenothiazine compound exhibiting antiemetic, antipsychotic, antihistaminic, as well as anticholinergic properties. Prochlorperazine binds to and inhibits postsynaptic dopamine D2 receptors in the brain's center of the chemoreceptor trigger

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zone (CTZ), possibly avoiding chemotherapy-induced nausea. Prochlorperazine maleate additionally inhibits anticholinergic and alpha-adrenergic receptors.

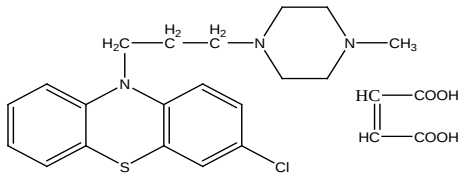

Figure 1: Structure of Prochlorperazine Maleate

The molecular structure highlights functional groups such as the phenothiazine core and piperazine ring, contributing to its solubility, stability, and interaction with chromatographic columns. The phenothiazine group affects polarity and retention, while the piperazine group aids in solubility, influencing the mobile phase composition during RP-HPLC analysis.

Prochlorperazine maleate exerts an antagonistic impact on the α-1 adrenergic receptors, leading to unconsciousness, relaxation of muscles, and hypotension. Prochlorperazine demonstrates antiemetic effects and has been shown to suppress apomorphine-induced vomiting by antagonizing D2 dopamine receptors in the chemoreceptor trigger zone (CTZ) [1]. Prochlorperazine, commonly referred to as Compazine, is a piperazine-phenothiazine derivative and a first-generation antipsychotic medication utilized for the therapy of acute vomiting and nausea, in addition to the short-term management of psychotic disorders, including nonspecific non-psychotic distress and dementia. It primarily functions by inhibiting the chemoreceptor trigger zone and obstructing the dopamine receptors D2 throughout the neural network. It was additionally demonstrated to obstruct histaminergic, cholinergic, and noradrenergic receptors. Prochlorperazine was created in the 1950s and received FDA approval in 1956. Prochlorperazine remains extensively utilized for the management of vomiting and nausea despite the greater promotion of novel antiemetic agents like 5-HT3 antagonists [2]. Prochlorperazine undergoes hepatic biotransformation involving oxidation, hydroxylation, demethylation, sulfoxide formation, and conjugation with glucuronic acid. CYP2D6 mediates the oxidation reaction. Upon oral & buccal administration, N-desmethyl prochlorperazine was detected in the plasma, as well as prochlorperazine sulfoxide, prochlorperazine 7-hydroxide, and prochlorperazine sulfoxide 4'-N-oxide. Prochlorperazine may enter the enterohepatic circulation [3-4]. Prochlorperazine Maleate, a piperazine phenothiazine derivative, is a commonly utilized medicinal compound with significant antiemetic and antipsychotic effects. This compound primarily functions by opposing dopamine D2 receptors located in the chemoreceptor trigger zone (CTZ) of the brain, effectively managing nausea, vomiting, and symptoms of psychotic disorders such as schizophrenia and generalized non-psychotic anxiety. Prochlorperazine Maleate retains clinical significance due to its varied pharmacological effects despite introducing novel antiemetic medicines such as 5-HT3 antagonists.

Prochlorperazine Maleate exhibits moderate aqueous solubility, essential for its bioavailability and formulation stability. The compound demonstrates a solubility of approximately 10 mg/mL in water at 25°C. The pKa values of Prochlorperazine are reported to be 8.2 (for the piperazine ring) and 3.9 (for the phenothiazine core), indicating ionization at physiological pH. These pKa values are critical in optimizing the mobile phase for RP-HPLC analysis, ensuring effective separation and peak symmetry. The Log P (partition coefficient) value is 4.0, reflecting its lipophilic nature, contributing to its high affinity for lipid membranes and enhancing permeability across biological barriers. This physicochemical profile helps determine chromatographic behavior, influencing retention time and separation efficiency in reverse-phase chromatography [5].

Considering its medicinal significance, analytical techniques for detecting Prochlorperazine Maleate in pharmaceutical products are relatively restricted. Current methodologies frequently encounter difficulties concerning sensitivity, accuracy, or specificity. Techniques such as UV-spectroscopy and traditional chromatographic methods may be insufficiently precise for monitoring quality in formulation evaluation, especially in identifying small quantities and differentiating Prochlorperazine Maleate among additives and degradation byproducts. Moreover, existing high-performance liquid chromatography (HPLC) techniques can involve intricate mobile phases or protracted preparation procedures, impeding their utility in high-throughput quality control environments [6].

Existing methodologies for the quantification of prochlorperazine maleate in pharmaceutical formulations often face critical limitations, such as inadequate sensitivity for detecting low concentrations of impurities, non-specificity in distinguishing the analyte from excipients or degradation products, and inefficiencies in high-throughput environments due to lengthy preparation and analysis times. These

shortcomings pose significant challenges in ensuring quality control and regulatory compliance, especially in complex formulations. The lack of robust, stability-indicating methods limits the reliability of existing approaches in routine analysis. Addressing these unmet needs, this study introduces an innovative, effective, and accurate reverse-phase HPLC (RP-HPLC) method for analyzing Prochlorperazine Maleate in its various forms. This method was developed for optimum parameters that improve resolution, decrease analysis time, and uphold high standards of correctness and repeatability. This study aims to establish a reliable, cost-effective method suitable for routine quality control in pharmaceutical laboratories by adhering to the International Conference on Harmonisation (ICH) guidelines for method validation. The method's simplicity and efficacy render it a significant alternative for regulatory and quality assurance processes, especially in developing regions where available analytical tools are essential [7-8].

MATERIALS AND METHOD

Chemicals and reagents

Prochlorperazine Maleate was obtained as a gift sample from Aadhar Life Sciences. RO Water, HPLC-grade Acetonitrile and Formic acid (Merck Specialities Pvt. Ltd., Mumbai, India), and 0.45 µm Millipore syringe filters (Ultipor®N_{66} ®Nylon Membrane) were also from Adhar Life Sciences, India.

Rationale for HPLC Conditions

The selection of HPLC conditions was optimized to achieve rapid, reliable, and reproducible detection of Prochlorperazine Maleate. The Agilent Zorbax Bonus-RP column (250 x 4.6 mm, 5 µm) was chosen for its outstanding durability and exceptional peak uniformity for this substance, ensuring optimal separation effectiveness. The mobile phase was comprised of 0.1% formic acid and acetonitrile in a 70:30 ratio, chosen to optimize orientation, enhance peak morphology, and augment sensitivity by reducing tailing. The specific concentration of 0.1% formic acid was selected because it stabilizes the pH of the mobile phase, ensuring consistent peak symmetry and reproducibility. Previous studies have demonstrated that lower concentrations (e.g., 0.05%) may result in inadequate resolution, while higher concentrations (e.g., 0.2%) can increase column backpressure and affect separation efficiency. This optimal concentration balances effective ion-pair formation and elution strength, yielding symmetrical peaks with minimal tailing and robust sensitivity. These requirements ensure a consistent retention time and improve technique resilience for regular analysis. The selection of HPLC conditions was optimized to achieve rapid, reliable, and reproducible detection of Prochlorperazine Maleate. The Agilent 1260 Infinity II HPLC system (Agilent Technologies, USA) was used for chromatographic analysis and was equipped with a DAD detector (G7115A). The separation was performed on an Agilent Zorbax Bonus-RP column (250 x 4.6 mm, 5 µm), chosen for its durability and peak uniformity.

Reagents and Solutions Preparation

Stock Solution Preparation: A standard stock solution (1,250 µg/mL) was formulated by carefully weighing 12.5 mg of Prochlorperazine Maleate, dispersing it in 5 mL of solvent (0.1% formic acid: acetonitrile in a 50:50 ratio), sonicating for 5 minutes, and subsequently adjusting the amount of solution to 10 mL using solvent.

Working Standard Solution: 1 mL of the original solution was reduced to 10 mL using the solvent to obtain 125 µg/mL.

The preparation of stock and working solutions involved the use of additional instruments, including a pH meter (Eutech pH 700, Thermo Fisher Scientific, India) for pH adjustments and a sonicator (Ultrasonic Cleaner, Model USC1200, Spectra Lab, India) to ensure complete dissolution of Prochlorperazine Maleate. Using these instruments ensured reproducibility and homogeneity during solution preparation, contributing to the accuracy and reliability of the developed RP-HPLC method.

Sample Preparation for Assay: Exactly 1 mL of the experimental solutions was introduced into a 10 mL volumetric flask, diluted to the desired level with a sonicated dilution agent, and calibrated to the final concentration of 125 µg/mL. Every sample underwent filtration through a 0.45 µm nylon filter to remove particulates before injection [9].

Chromatographic Conditions

Column: Agilent Zorbax Bonus-RP (250 x 4.6 mm, 5 µm)
HPLC System: Agilent 1260 Infinity II HPLC (Agilent Technologies, USA)
Detector: Diode Array Detector (DAD) - G7115A
Mobile Phase: 0.1% formic acid: acetonitrile (70:30)
Flow Rate: 1 mL/min
Injection Volume: 10 µL
Detection Wavelength: 258 nm
Run Time: 10 minutes

Data Analysis

Data were processed using Agilent's OpenLAB CDS software, which facilitates accurate peak integration, calibration curve generation, and quantification. The method development and data analysis utilized Agilent OpenLAB CDS software (version 2.8). This version ensures compatibility with advanced chromatographic techniques and offers enhanced features for peak integration accuracy, calibration, and system suitability assessment. Calibration curves were constructed by plotting the peak area against Prochlorperazine Maleate concentration, with a linear range confirmed over the specified concentrations.

Preparation of calibration standards and quality control sample

Calibration standard solutions were prepared through successive dilutions of the standard used for calibration to obtain concentrations between 80 and 120 µg/mL, which ensures correctness across a broad spectrum. Quality control specimens at low, medium, and high levels were employed to validate technique precision and reproducibility.

VALIDATION OF ANALYTICAL METHOD

Specificity and Assay

The blank, operating standard, and medication were introduced and monitored for any solvent peak that might interfere with the primary peak, and the assay was subsequently calculated as shown in Table 1 [10].

Table 1: Grouping of calibration standards & corresponding prochlorperazine maleate concentrations

% Conc.	Prochlorperazine Maleate conc. (µg/ml)
80	100
90	112.5
100	125
110	137.5
120	150

Repeatability and System Suitability

Under the specified experimental conditions, the precision of an analytical parameter refers to the proximity of the test outcome to the real value derived from repeated samplings of a homogeneous sample. Equipment precision was evaluated by generating one sample of solution, from which five injections were performed to evaluate systems suitability [11]. The system suitability parameters for Prochlorperazine Maleate are discussed as below:

- Theoretical plates: 6252 - Retention time: 2.24 min.
- Wavelength: 258 nm - Asymmetry (Tailing Factor): 0.97

Accuracy

The accuracy of an experimental variable is defined as the proximity of the experimental results to the true value. The accuracy of the proposed method was assessed using the conventional addition technique or recovery study through the spiking of the reference stock solution.

a. Samples were prepared at concentrations of 75%, 100%, and 125% for Prochlorperazine Maleate.
b. Percent relative standard deviation (% RSD) was calculated by injecting specimens in duplicate.
c. The percentage recovery was also computed [12].

Intra & Inter-day Precision

Single mixture working standard and drug product samples were prepared and injected twice in a day at different time intervals to evaluate intra-day precision. Same mixture working standard and drug product samples were analysed on second day to evaluate the inter-day precision. % Assay was calculated at each interval and stability of solutions were estimated [13].

LOD and LOQ

The limit of detection (LOD) indicates the minimum concentration of an element in the specimen that can be readily detected, though it may not be evaluated. The limit of quantitation (LOQ) in an analytical parameter indicates the minimum concentration of an analyte in a sample that can be reliably identified and analyzed with appropriate precision and accuracy. These parameters are critical in ensuring trace impurities in pharmaceutical formulations are detected and quantified effectively. The calculated LOD (1.76 µg/mL) and LOQ (5.35 µg/mL) demonstrate the method's sensitivity and make it suitable for monitoring even minute quantities of Prochlorperazine Maleate in complex formulations. Furthermore, the robustness of these values was evaluated under varied environmental conditions, including temperature fluctuations and changes in sample matrices. The results confirmed consistent detection limits across these variations, underscoring the method's reliability for routine quality control applications [14]. By using the following equations, LOD & LOQ were determined;

$LOD = 3.3*\sigma/S$ (σ = Standard deviation)

$LOQ = 10*\sigma/S$ (S= slope of the regression coefficient)

Robustness

The robustness was assessed by varying the column temperature by ± 2˚C. Every specimen was administered and the assay was determined under every circumstance [15].

Table 2: Robustness Evaluation of Prochlorperazine Maleate: Effect of Column Temperature on Retention Time and % Assay

Condition	Increased	Normal	Decreased
Column Oven Temperature	32˚C	30˚C	28˚C
Mobile Phase A Strength	0.08%	0.10%	0.12%

Method Precision

Six specimens of identical proportions were made up and injected to ascertain the method's precision. The selection criterion stipulates that the % RSD must not exceed 2% [16].

Intermediate Precision

To check the intermediate precision, 6 samples of the same concentration were prepared by different analysts and injected. The acceptance criterion is a % RSD of over 2% [17].

RESULTS

Method Development

The suggested HPLC technique is created and modified for various trials regarding the mobile phase selection, wavelength, composition, stationary part of the column, flow rate, and column temperatures. Prochlorperazine Maleate exhibited an absorption maximum at 258 nm. Therefore, that wavelength was selected as the operational frequency for the suggested HPLC technique. To ensure the robustness of the developed method, matrix effect studies were conducted by testing the influence of excipients and degradation products commonly present in pharmaceutical formulations. The retention time and peak symmetry were evaluated for samples spiked with typical formulation excipients such as binders, fillers, and disintegrants. The results demonstrated consistent retention times and peak shapes, with no significant interference from matrix components. These findings indicate that the method is highly selective and capable of accurately quantifying Prochlorperazine Maleate even in the presence of complex matrices.

Method Validation

The optimal mobile phase ratio for Acetonitrile and 0.1% Formic acid is 30:70, with a 1 ml/min flow rate. The samples were separated using a Zorbax Bonus-RP column at 30°C ± 0.5°C. Samples and typical specimens were introduced in a volume of 10 µl using an autosampler. The duration of retention of Prochlorperazine Maleate in the suggested technique was determined to be 2.24.

Specificity and Assay

Figure 2A presents an overlay of chromatograms for blank, standard, and sample injections. The chromatograms demonstrate the absence of interference peaks at the retention time of Prochlorperazine Maleate, supporting the method's specificity. The comparison highlights the method’s ability to distinguish the analyte from excipients and potential degradation products, ensuring accurate quantification in pharmaceutical formulations. The solution was analyzed at 258 nm (Figure 2).

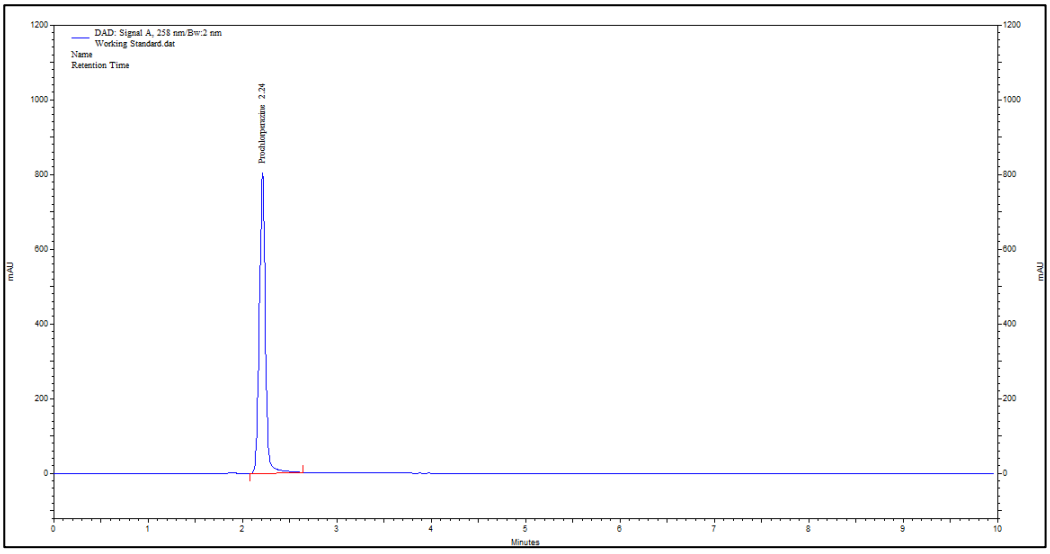

Figure 2: Chromatogram of mixture of Prochlorperazine Maleate

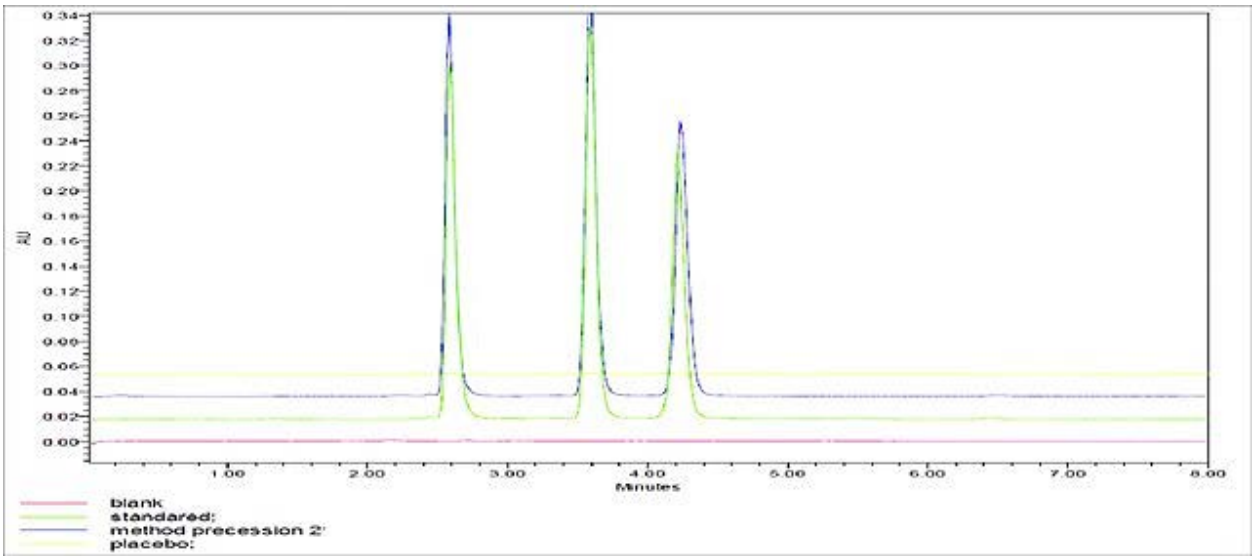

Figure 2A: Overlaid chromatograms of blank, standard, and sample injections. The absence of interference peaks confirms the specificity of the RP-HPLC method for Prochlorperazine Maleate.

Table 3 presents the specificity and assay results, indicating the retention times and calculated assay values for blank and standard solutions.

Table 3: Specificity and Assay Results for Prochlorperazine Maleate

Sample ID	RT	Area	% Assay
Blank	-	-	-
WS	2.24	7451889	-
DP	2.24	7395482	99.24

Linearity

The linearity was evaluated over the 80% to 120% concentration range, revealing an accurate relationship between concentration and peak area, as illustrated in Table 4. The calibration curve (Figure 3) further substantiates the linear relationship, which is crucial for precise quantification.

Table 4: Linearity Assessment of Prochlorperazine Maleate: Peak Area versus Concentration

%Conc.	Conc (µg/ml)	Area
80	100	5910552
90	112.5	6695326
100	125	7451889
110	137.5	8204694
120	150	8984030

Repeatability

The findings (Table 5) indicate reliability throughout six separate experiments at 100% concentration, exhibiting slight variation in retention time, theoretical plates, symmetry, and peak purity values. The %RSD results fall inside permissible parameters, confirming technique dependability.

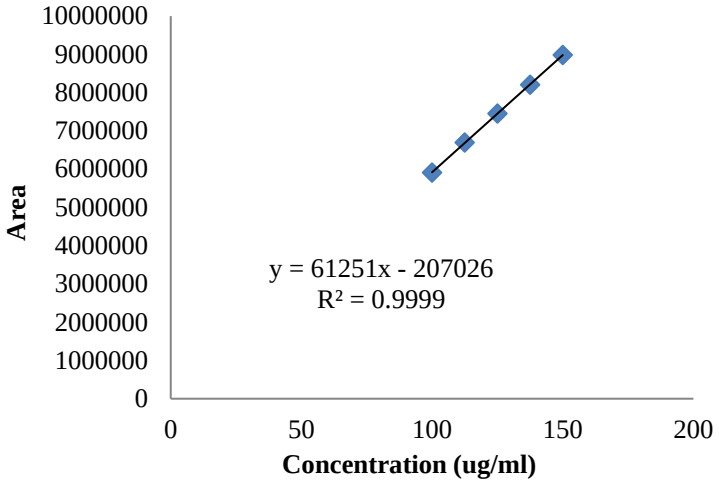

Figure 3: Calibration curve of Prochlorperazine Maleate

Accuracy

Recovery studies at three concentration levels (80%, 100%, and 120%) confirmed accuracy, as shown in Table 6. The method displayed recovery rates close to 100%, with %RSD values well within acceptable limits, ensuring method accuracy.

Recovery Analysis with Degraded/Spiked Samples

To further evaluate the robustness of the proposed method under real-world conditions, recovery studies were conducted using intentionally degraded samples and spiked formulations. Samples were degraded under controlled conditions (e.g., heat

and light exposure) to simulate typical stress scenarios. The method successfully quantified Prochlorperazine Maleate in these degraded formulations with recovery values ranging from 98–102%, demonstrating its applicability in handling variable sample conditions.

Table 5: Repeatability Results for Prochlorperazine Maleate (100% Standard Solution Injections)

Sample ID	Area	Retention Time	Theoretical Plates	Asymmetry	Peak Purity
100% Rep 1	7451889	2.24	6219	1.00	1.00
100% Rep 2	7423587	2.24	6299	0.99	1.00
100% Rep 3	7435864	2.24	6158	0.98	1.00
100% Rep 4	7461175	2.24	6059	1.03	1.00
100% Rep 5	7429547	2.24	6328	1.03	1.00
100% Rep 6	7453511	2.24	6157	1.00	1.00
Average	7442596	2.24			
STDEV	15017.42	0			
% RSD	0.20	0.00			

Table 6: Accuracy and Recovery Data for Prochlorperazine Maleate at 80%, 100%, and 120% Spiked Concentrations

Sample ID	Spiked Conc (ug/ml)	Area	Amount Recovered (ug/ml)	% Recovery	AVG	STDEV	% RSD
80% Rep 1	99.7	5910552	98.97	99.27	99.23	0.226317	0.23
80% Rep 2	99.7	5893595	98.69	98.98			
80% Rep 3	99.7	5920214	99.13	99.43			
100% Rep 1	124.625	7451889	124.78	100.12	99.93	0.19069	0.19
100% Rep 2	124.625	7423587	124.31	99.74			
100% Rep 3	124.625	7435864	124.51	99.91			
120% Rep 1	149.55	8984030	150.44	100.59	100.43	0.174405	0.17
120% Rep 2	149.55	8953215	149.92	100.25			
120% Rep 3	149.55	8972584	150.24	100.46			

Precision

Precision was evaluated through both intraday and interday studies (Table 7). The method consistently achieved low %RSD values across days, underscoring robust precision.

Detection and Quantitation Limits

Tables 8 and 9 outline the limits of detection (LOD) and quantitation (LOQ) for Prochlorperazine Maleate.

Robustness

The robustness of the HPLC method was evaluated by examining the effects of minor variations in method parameters, precisely column oven temperature and mobile phase composition, on the retention time, peak area, and assay results of Prochlorperazine Maleate. The consistency of results under these slight modifications confirms the method's robustness, demonstrating it can reliably withstand minor operational changes.

Temperature Variations

Table 10 illustrates the effect of varying the column oven's temperature at 28°C, 30°C, and 32°C. The retention time and percentage assay exhibited consistency, with negligible variation in peak area and percentage assay, demonstrating that the method is resilient to temperature fluctuations within this range.

Table 7: Intra-day and Inter-day Precision Results for Prochlorperazine Maleate

Intra Day precision			
Day 1	**Sample ID**	**Prochlorperazine Maleate**	
		Area	**%Assay**
Morning	**WS**	7451889	-
	DP	7395482	99.24
Evening	**WS**	7417854	-
	DP	7352217	99.12
Inter Day precision			
Day	**Sample ID**	**Prochlorperazine Maleate**	
		Area	**%Assay**
Day 2	**WS**	7428633	-
	DP	7374502	99.27
% RSD			0.08

Table 8: LOD for Prochlorperazine Maleate

Parameter	**Prochlorperazine Maleate**
LOD (µg/ml)	1.76

Table 9: LOQ for Prochlorperazine Maleate

Parameter	**Prochlorperazine Maleate**
LOQ (µg/ml)	5.35

Table 10: Robustness of Prochlorperazine Maleate Method: Effect of Mobile Phase Composition Variations

Condition	**Sample**	**Prochlorperazine Maleate**		
		RT	**Area**	**%Assay**
28℃	**WS**	2.24	7463251	-
	DP	2.24	7412214	99.32
30℃	**WS**	2.24	7451889	-
	DP	2.24	7395482	99.24
32℃	**WS**	2.24	7450257	-
	DP	2.24	7402469	99.36

Mobile Phase Composition Variations

Table 11 displays the outcomes of varying the concentration of Mobile Phase A (formic acid) at 0.08%, 0.10%, and 0.12%. Notwithstanding these modifications, retention time, area, and % assay remained consistent, demonstrating that the technique can withstand minor variations in the composition of the mobile phase without compromising the reliability or accuracy of the assay.

Table 11: Results of Robustness of Prochlorperazine Maleate-Change in Mobile Phase-A Strength

Condition	**Sample**	**Prochlorperazine Maleate**		
		RT	**Area**	**%Assay**
0.08%	**WS**	2.24	7428974	-
	DP	2.24	7362541	99.11
0.10%	**WS**	2.24	7451889	-
	DP	2.24	7395482	99.24
0.12%	**WS**	2.24	7459874	-
	DP	2.24	7412154	99.36

DISCUSSION

The RP-HPLC method for Prochlorperazine Maleate has been tested following ICH Q2(R1) guidelines, yielding results that indicate high precision, accuracy, and robustness. A comparison of our results with previously reported techniques suggests that this method offers enhanced sensitivity and simplicity.

The technique's %RSD values for precision and accuracy are within an optimal range (<2%), demonstrating repeatability and reliability similar to or superior to current methods for Prochlorperazine Maleate analysis. Initial investigations of Prochlorperazine Maleate served as the basis for developing analytical methods. Stock solutions of the Prochlorperazine Maleate were made in the diluent Acetonitrile: 0.1% and formic acid (50:50) and the mobile phase Acetonitrile: 0.1% formic acid (30:70) since they are readily soluble in water and formic acid. The Prochlorperazine Maleate in API and pharmaceutical formulation RP-HPLC method was created. Acetonitrile: 0.1% with formic acid (30:70) was used as the mobile phase, while a ZOROBAX Bonus-RP Column (250 x 4.6 mm, 5 m) was used as the stationary phase. The mobile phase's flow rate of 1.0 ml/min was kept at 258, and it was eluted at 2.24 minutes.

Precision and Accuracy

The method’s precision was confirmed through intra- and inter-day studies, yielding %RSD values of 0.08% and 0.35%, respectively, demonstrating excellent consistency. Similarly, the method's accuracy, as indicated by % recovery rates ranging from 99-101%, confirms its reliability for routine quality control applications. Compared to previously published HPLC methods for Prochlorperazine Maleate, this approach is highly reproducible, with minimal deviation observed across replicates, emphasizing its suitability for consistent and reliable quantification.

Sensitivity and Practical Benefits

The method's low limits of detection (LOD, 1.76 µg/ml) and quantitation (LOQ, 5.35 µg/ml) signify its high sensitivity, making it particularly advantageous for detecting low concentrations in formulations. Furthermore, the rapid elution time of 2.24 minutes enhances throughput in quality control settings, reducing analysis time without sacrificing accuracy. These aspects, combined with the straightforward sample preparation process (using a diluent of 0.1% formic acid and acetonitrile), position this method as cost-effective and time-efficient for routine analysis of Prochlorperazine Maleate.

Challenges and Mitigation

Future studies should extend this method to stability-indicating investigations under extreme conditions, such as prolonged exposure to light, high temperatures, and variable storage conditions. Such studies would evaluate the method's capability to detect degradation products and assess its reliability in monitoring Prochlorperazine Maleate's stability over time. These additional insights would strengthen the method's application in regulatory environments and pharmaceutical quality assurance.

Specificity and System Suitability

The lack of interfering peaks in the chromatograms for blank and standard samples demonstrated specificity, which confirms the method's selectivity. System suitability parameters, including theoretical plate counts (above 6000) and tailing factors (close to 1), further validate the method's effectiveness for clear, symmetrical peak formation. Figures 1–3 illustrate the chromatograms of blank, standard, and sample injections, displaying clear retention times and peak shapes that underscore the method specificity and suitability for accurate quantification.

Robustness

Statistical analysis was performed to evaluate the method's robustness under variations in column oven temperature and mobile phase composition. The variance in retention time, peak area, and assay percentage for both parameters was negligible. For example, the standard deviation of assay values across temperature variations (28°C, 30°C, and 32°C) was ±0.06%, and for mobile phase composition variations (0.08%, 0.10%, 0.12%), it was ±0.07%. These results confirm the method's reliability and reproducibility under minor operational changes, supporting its suitability for routine analysis.

Table 14: Summary of RP-HPLC Method of Prochlorperazine Maleate

Sr. No.	Parameters	Prochlorperazine Maleate
1.	**Specificity**	The retention time of Prochlorperazine Maleate was 2.24 minutes
2.	**Linearity Range (µg/ml)**	100-150 µg / ml
3.	**Regression Equation (y = mx+c)**	y = 61251x – 20702
4.	**Correlation Coefficient (r^2)**	0.999
5.	**LOD (µg/ml)**	1.76 µg/ml
6.	**LOQ (µg/ml)**	5.35 µg/ml
7.	**% Recovery**	99-101%
8.	**Instrument Precision (%RSD)**	0.20%
9.	**Intra & Inter Day Precision (%RSD)**	0.08%
10.	**Method Precision (%RSD)**	0.48%
11.	**Intermediate Precision (%RSD)**	0.35%
12.	**Matrix Effect Studies**	No significant interference was observed from excipients or degradation products; consistent retention time and peak area were confirmed.

CONCLUSION

The developed RP-HPLC method for Prochlorperazine Maleate is robust, sensitive, and efficient, making it highly suitable for routine quality control (QC) analysis in pharmaceutical formulations. This method demonstrated excellent precision, accuracy, and linearity over the specified concentration range, with low limits of detection (LOD) and quantitation (LOQ) values. The short analysis time and straightforward sample preparation enhance its practicality, offering a cost-effective solution for QC laboratories, especially in regions where rapid

and reliable testing methods are essential. Future work could explore the application of this method to other dosage forms of Prochlorperazine Maleate and related compounds to further validate its versatility. During the validation studies, the developed method was tested on pharmaceutical formulations, including tablets and syrups, to demonstrate its generalizability. The consistent recovery rates and low %RSD values observed across these formulations confirm the method's applicability for routine analysis of diverse dosage forms. These results highlight the method's potential for broad use in pharmaceutical quality control settings. Additionally, this method could be adapted for stability studies and pharmacokinetic research, where rapid and precise quantification of Prochlorperazine Maleate is crucial.

ABBREVIATIONS

LOD - Limit of Detection; **LOQ -** Limit of Quantification; **RP-HPLC -** Reverse-Phase High-Performance Liquid Chromatography; **%RSD -** Percent Relative Standard Deviation; **CTZ -** Chemoreceptor Trigger Zone; **ICH -** International Conference on Harmonization; **QC -** Quality Control; **API -** Active Pharmaceutical Ingredient; **CYP2D6 -** Cytochrome P450 2D6; **UV –** Ultraviolet; **FDA -** Food and Drug Administration; **WS –** Working Standard; **DP –** Drug Product

FINANCIAL ASSISTANCE

NIL

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

Nikhil Shrisunder and Prashant Kumar Dhakad collected data and performed experiments. Ritu Gilhotra conducted the analysis. Nikhil Shrisunder wrote the first draft of the manuscript, and all authors reviewed and revised previous versions. All authors contributed to the study's conception and design and gave final approval.

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