



ISSN : 2347-2251

**Indo-American Journal of
Pharma and Bio Sciences**



www.iajpb.com

iajpb.editor@gmail.com
editor@iajpb.com



A NOVEL VALIDATED RP-HPLC METHOD FOR ESTIMATION OF PROPIOMAZINE IN BULK AND PHARMACEUTICAL DOSAGE FORMS

M. Lakshmi Surekha*, Bharghava Bhushan Rao P¹, R. Sunitha², Padma R³, Jhansi Rani M⁴
Ajay Kumar Ch⁵

ABSTRACT

A simple, rapid, accurate, precise and reproducible RP-HPLC method was developed for the estimation of Propiomazine in liquid dosage forms. The method was carried out using Inertsil ODS 3V(150mm x 4.6 mm), 5µm column in an binary mode with mobile phase comprising gradient mixture of pH 3.0 Potassium Di-Hydrogen phosphate and Acetonitrile. The flow rate was 1.2 ml/min and detection was carried out at 260 nm using a UV detector. The retention time for Propiomazine was found to be at 3.76 min and 9.74 min. The method for Propiomazine showed linearity in the concentration range of 153.7- 461µg/ml ($R^2=1.000$) and for Propiomazine showed linearity in the concentration range of 12.6-37.8µg/ml ($r^2=0.9997$). The recovery studies for Propiomazine also carried out and %RSD for reproducibility was found to be below 2%. The method was simple, sensitive and specific. Hence method can be used for the quantification of Propiomazine in pharmaceutical dosage form.

Key Words: Propiomazine; RP-HPLC; ICH validation; PDA Detector.

Introduction

Propiomazine is a member of the class of phenothiazines that is 10H-phenothiazine substituted by a 2-(dimethylamino)propyl group at nitrogen atom and a propanoyl

group at position 2. It is a member of phenothiazines, an aromatic ketone and a tertiary amino compound. It derives from a hydride of a 10H-phenothiazine.

²³⁴⁵A.M. Reddy Memorial College of Pharmacy, petlurivaripalem, Andhra Pradesh 522601. India.

¹ Professor, V V Institute of Pharmaceutical Sciences, Gudlavaluru, A.P.

*Corresponding author

Dr.M.Lakshmi Surekha

Professor & Head

A.M.Reddy Memorial College of Pharmacy,

Dept .of Pharmaceutical Analysis,

Uppalapadu, Narasaraopet, Guntur (Dt),

Andhra Pradesh, India.

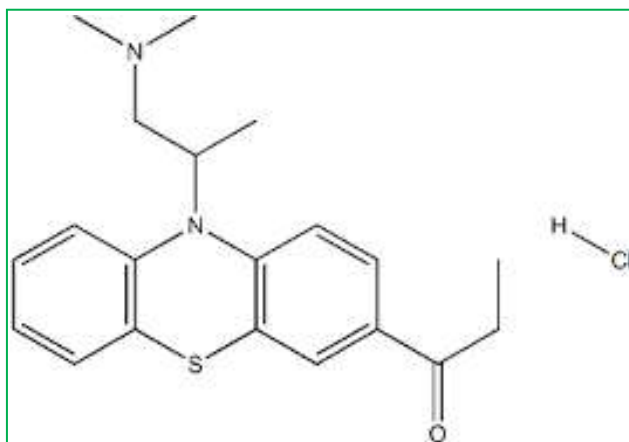


Fig.01. Chemical Structure of Propiomazine.

Propiomazine, an atypical antipsychotic agent, is used to treat both negative and positive symptoms of schizophrenia, acute mania with bipolar disorder, agitation, and psychotic symptoms in dementia. Future uses may include the treatment of obsessive-compulsive disorder and severe behavioral disorders in autism. Structurally and pharmacologically similar to clozapine, propiomazine binds to $\alpha(1)$, dopamine, histamine H₁, muscarinic, and serotonin type 2 (5-HT₂) receptors.

Mechanism of action

Propiomazine acts as an antagonist of dopamine 1, 2, and 4 receptors, serotonin (5-HT) receptor types 2A and 2C, muscarinic receptors 1 through 5, $\alpha(1)$ -receptors, and histamine H₁-receptors. Its main use as a sedative is due to its antihistamine effect.

Method Development and optimization of Propiomazine:

Preparation of solutions:

Diluent: Based up on the solubility of the drug diluent was selected, Methanol

taken in the ratio of 50:50.

Preparation of standard stock solutions:

Accurately weighed 10mg of Propiomazine transferred in 10ml volumetric flask $\frac{3}{4}$ th of diluent was added and sonicated for 10 mins. Flask was made up with diluent and labelled as standard stock solution (1000ug/ml).

Preparation of standard working solution:

One ml of Propiomazine from stock solution was pipette out and taken into a 10ml volumetric flask and make up with diluents (100ug/ml).

Determination of wavelength:

Standard working solution was scanned between 200-400nm in double beam UV-Visible spectrophotometer. Maximum absorbance ($\lambda_{\max}$) for the Propiomazine was determined by UV spectrophotometer and observed that maximum absorbance at 247nm.

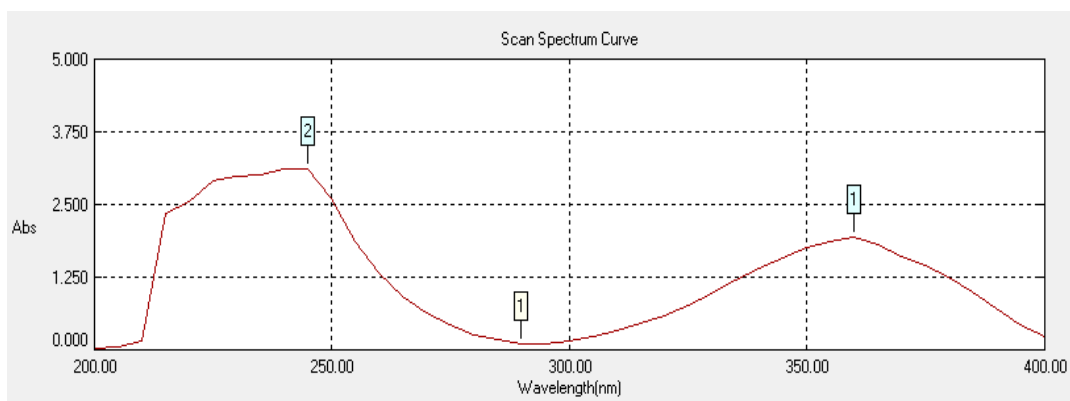


Fig.2 Propiomazine UV-Visible Spectrum

Chromatographic condition:

Stationary Phase	:	INERTSIL ODS 3V (5um 4.6×250nm)
Wavelength	:	247nm
Flow rate	:	1.0ml/min
Temperature	:	30 ⁰ C
Injection volume	:	20ul
Run time	:	5 min
Mobile phase	:	Acetonitrile: water (80:20)
Diluent	:	Methanol :Water

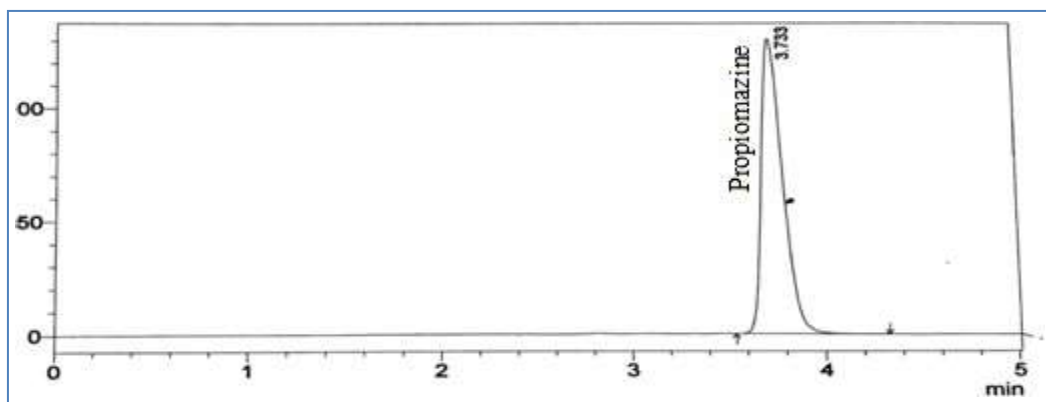


Fig.3 Typical Chromatogram of Propiomazine

RESULTS AND DISSCUSSION

Method validation:

Linearity: The concentration range of 10-50 µg/ml for Propiomazine and all the standard solutions were filtered and injected.

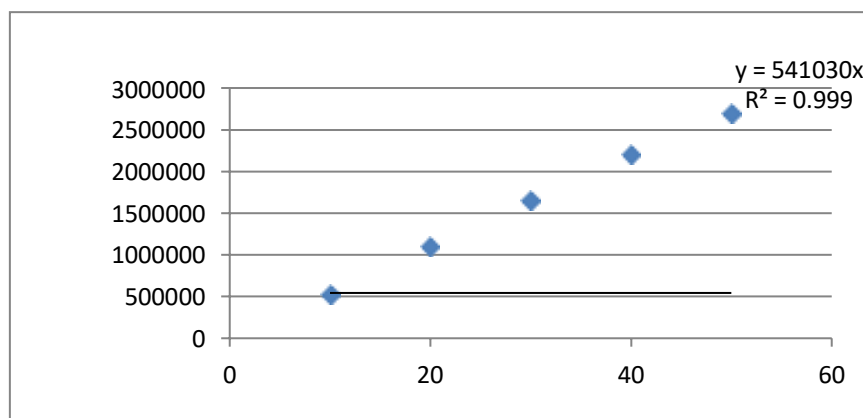


Fig.03 Calibration curve of Propiozamine

Table 1 Linearity data of Propiomazine

Linearity level	Concentration (µg/ml)	Peak Area		Mean
		Set 1	Set 2	
1	10	531665	533356	531088
2	20	1106624	1106424	1106524
3	30	1650303	1652426	1651364

4	40	2201066	2201266	2201166
5	50	2693870	2683944	2688907

Table: 2 Precision data of Propiomazine

S.NO	Concentration (µg/ml)	Amount (µg/ml)	% of Amount	Avg	S.D	%RSD
1	30	30.1	100.3	100.2	0.90	0.89
2		30.1	100.3			
3		30	100			
4		30.1	100.3			
5		30.1	100.3			
6		30	100			

Accuracy

Accuracy was done by recovery studies

into a 10ml volumetric flask, to that 4.5ml from each standard solution was pipette out and make up the mark with diluents.

Accuracy: Three concentrations of 50%, 100% and 150% are prepared and injected.

Preparation of 50% Spiked solution:

3.0ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.5ml from each standard solution was pipette out and make up the mark with diluents.

Preparation of 100% Spiked solution:

3.0ml of sample stock solution was taken into a 10ml volumetric flask, to that 3.0ml from each standard solution was pipette out and make up the mark with diluents.

Preparation of 150% Spiked solution:

3.0ml of sample stock solution was taken

Table 3 Accuracy data

Percentage level	Amount (µg/ml)	Amount added (µg/ml)	Amount found	Amount Recovery	% Recovery	Avg %	S.D	%RSD
50	30	15	44.9	14.9	99.3	100.6	1.1	1.09
			45.2	15.2	101.3			
			45.2	15.2	101.3			
100		30	60.1	30.1	100.3	100.7	0.40	0.41
			60.3	30.3	101			
			60.3	30.3	101			
150		45	74.8	44.8	99.5	99	0.93	0.93
			74.1	44.1	98			
			74.9	44.9	99.7			

Limit of detection & Limit of quantification:

The limit of detection and the limit of quantification were calculated by using the average value of slope and the standard deviation of intercept and the results are listed in the table.4

Table 4.Data of LOD and LOQ

SNO	Parameters	Propiomazine
1	LOD	0.15
2	LOQ	0.48

Robustness:

Robustness was done by changing the column temperature ($\pm 5^{\circ}\text{C}$), flow rate ($\pm 10^{\circ}\text{C}$), changing the wavelength ($\pm 5\text{nm}$), and organic compounds of mobile phase ($\pm 5\%$). All the system suitability parameters must be met as per the method.

1. The tailing factor of Propiomazine maleate should be not more than 2 for variation in flow and wavelength.
2. The % RSD of asymmetry and retention time for fexofendine should be not more than 2% for variation in flow and wavelength. Small deliberate change in the method is made like Flow minus, Flow plus, Wavelength minus, Wavelength plus, Temperature minus, Temperature plus. The % RSD of the above conditions is calculated shown Table 5.

Table 5 Robustness studies of Propiomazine maleate

Parameter	Conditions	Variation	%RSD
Wavelength Variation	245	250	2
		240	0.55
Column oven temperature Variation	30°C	25°C	1.3
		35°C	0.95
Flow rate Variation	1ml/min	0.9ml/min	0.22
		1.1ml/min	0.39

7. SUMMARY AND CONCLUSION

Propiomazine was determined by RP-HPLC method, optimisation of chromatographic parameters was done. Parameters were optimised by altering the mobile phase ratio and flow rate at a wavelength of 247 nm. The trials for optimisation were conducted by using different mobile phases which include Acetonitrile: Water (90:10), ACN: Water (80:20), Methanol: Water (90:10), ACN: Water (75:25). Out of all trials, 75:25 ratio of ACN: Water (80:20) at 0.7ml/min flow rate was selected for this proposed method and it shows good system suitability values which include, tailing factor, retention time, no. of theoretical plates. The calibration was performed by using external calibration method. The stock solution of Propiomazine was prepared and dilution was made by using mobile phase and absorbance was

measured at 247 nm. Serial dilutions 10 to 50 $\mu\text{g/ml}$ of Propiomazine were prepared and injected and the chromatograms are recorded. The calibration curve using peak area Vs concentration was plotted. The correlation coefficient was calculated as 0.999, the system precision was done both intraday and interday and the % RSD is below 2.

The recovery studies were passed out to confirm the accuracy of the method by added standard drug to a previously analysed formulation. The average percentage recovery was appeared as 101%. LOD and LOQ were calculated and were in within limits. Robustness was performed by deliberate changes in the optimised condition such as flow rate, temperature, wave length, and chromatograms were noted. Twenty tablets are accurately weighed; powdered and

equivalent quantity i.e. 50 mg of Propiomazine tablet powder was taken and diluted by using diluent. The percentage of Propiomazine present in the formulation was occurred to be 101%.

Here by concluded that the method showed tremendous sensitivity, reproducibility, accuracy and repeatability, which is proved the low percentage relative standard deviation. The results of recovery studies, determines that there is no interference from the excipients used in formulation. RP-HPLC method can be effectively applied for the routine analysis of Propiomazine pure and tablet formulation in quality control analysis.

References

1. Takeru Higuchi, Elinar Brochmann, Hanffen Hansen. Pharmaceutical analysis. 1sted. New Delhi: CBS Publishers and Distributors Pvt Ltd; 1997.
2. Alexeyev V. Quantitative analysis, 1stEd. New Delhi: CBS Publishers and Distributors Pvt Ltd; 1994. P: 13.
3. Kaur H. Instrumental method of chemical analysis. 8th ed. Meerut: Pragati Prakashan; 2012. p 9-10.
4. Sharma B K. Instrumental methods of chemical analysis. 3rded. New Delhi: Vallabh Prakashan; 2004: p. 7-8
5. Skoog DA, Holler FJ, Crouch. Principles of instrumental Analysis. 6thEd. New Delhi: Cengage learning publishers; 2007: p. 1-39.
6. Hobert Willard H. Instrumental Methods of Analysis. 1st Ed. New Delhi: CBS publisher; 1986: p: 529-563.
7. Braun D. Introduction to Instrumental Analysis. 1st ed. Hyderabad: Pharma press; 2006: 821-889.
8. Supriya Mahajan. Instrumental Methods of Analysis. 1st ed. Mumbai: Prakashanpublishers: 2010: p: 252-256.
9. http://www.waters.com/waters/en_IN/How-does-High-performance-liqui-chromatography-work%3F/nav.htm?cid=10049055 HPLC Instrumentation. Block diagram of HPLC.
10. Stenlake JB, Beckett AH. Practical Pharmaceutical Chemistry. 4thed. New Delhi: CBSpublisher; 199: 494.
11. <http://www.pharmatech.com/pharmatech/artcledetail.jsp?id=691672H> PLC Instrumentation. HPLC detector properties [cited on 21-3-2016].
12. Synder L, Kirkland. Practical HPLC Method development. 2nd ed. New York: A Wiley-Interscience Publication; 1997: p: 1-19.
13. Billet, Ripper, Brown R, Phyllis E. Advances in Chromatography: Selectivity Optimisation in HPLC. International Journal of Phytopharmacology, 1998; 39: p: 264- 265.

14. Wickam D. Practical Guide to HPLC Detection. 1st ed. San Diego: Academic Press; 1993; p: 67.
15. Leblane DA. Rinse sampling for cleaning validation studies. Pharm tech; 1998, 22 (5), 66-74.
16. ICH Harmonized Tripartite Guidelines, Validation of analytical procedures: Text and methodology Q2(R1), November 2005.
17. A guide to good manufacturing practice 1996, 1st edition, chapter 9.
18. K. Kathiresan and Kiran Krishnan. Basics of validation – pharmaceutical perspective, 2005, 1st edition, K.K. Publisher's Chidambaram, 1-122
19. Available from: <http://www.ich.org/products/guidelines/quality/validation-of-analytical-procedures-text-and-methods.html>. ICH Harmonised Tripartite Guideline. Validation of Analytical Procedures: Text and Methodology, Q2 (R1), 2005.
20. Available from: http://www.ich.org/products/guidelines/quality/Q1A_R2/Step4/Q1A_R2_Guideline.pdf. ICH Harmonised Tripartite Guideline. Stability Testing of New Drug Substances and Products Q1A, (R2), 2003.