

Research Article

UV SPECTROPHOTOMETRIC METHOD FOR THE DETERMINATION OF RASAGILINE MESYLATE IN BULK AND PHARMACEUTICAL FORMULATIONS

Rama Bukka* and Kalyani Prakasam

Dept. of Pharmaceutics, Nargund College of Pharmacy, II Main Dattatreya Nagar, Bangalore-560085, Karnataka, India.

*Corresponding author's E-mail: ramabukka@gmail.com

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ABSTRACT

A simple and cost effective spectrophotometric method is developed for the determination of Rasagiline Mesylate in bulk form and in pharmaceutical formulations. The drug in aqueous solution showed absorption maximum at 271.6 nm and obeys Beer's Lambert's law in the concentration range 5-150 $\mu\text{g mL}^{-1}$. ($r^2 = 0.9999$) The apparent molar absorptivity was 935.6 $\text{Lmol}^{-1}\text{cm}^{-1}$. The slope and intercept of the equation of the regression line are 0.0054 and 0.005 respectively. The limit of detection and limit of quantification was found to be 0.5 $\mu\text{g mL}^{-1}$ & 5 $\mu\text{g mL}^{-1}$. The proposed method was successfully applied to the determination of Rasagiline in pharmaceutical formulations without any interference from common excipients.

Keywords: Rasagiline Mesylate, UV spectrophotometry, Validation, Pharmaceuticals.

INTRODUCTION

Rasagiline Mesylate is designated chemically as: 1H-Inden-1-amine, 2,3-dihydro-N-2-propynyl-, (1R)-, methanesulfonate¹. The empirical formula of Rasagiline mesylate is $(\text{C}_{12}\text{H}_{13}\text{N})\text{CH}_4\text{SO}_3$ and its molecular weight is 267.34¹. Figure 1 shows the structure of Rasagiline mesylate¹. Literature survey reveals that, few gas chromatography- mass spectrometry² and high performance liquid chromatography³ methods have been reported for the estimation Rasagiline mesylate. A validated LC-MS/MS method for the determination of Rasagiline in plasma is also reported⁴. The aim of this study is to develop a rapid, simple, accurate, selective, sensitive and inexpensive UV spectrophotometric method for the determination of Rasagiline Mesylate as bulk drug and in commercial pharmaceutical formulations as tablet. The developed method is validated as per the validation parameters.⁵⁻⁷

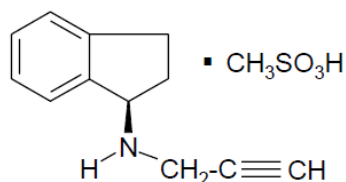


Figure 1: Chemical structure of Rasagiline Mesylate

Rasagiline Mesylate is a new drug used as irreversible monoamine oxidase inhibitor indicated for the treatment of idiopathic Parkinson's disease⁸. Rasagiline Mesylate is not an official drug in any of the pharmacopeia.

MATERIALS AND METHODS

The spectrophotometric measurements were carried out using a Shimadzu model 1800 UV VIS spectrophotometer (190-1100 nm). UV spectra of standard and sample

solutions were recorded in 1cm quartz cells at the wavelength ranges of 190-400 nm.

Rasagiline Mesylate was kindly provided by Apotex Research Private Ltd (Bangalore, India) and it was used without further purification. Rasalect tablets (containing Rasagiline Mesylate equivalent to 1 mg of Rasagiline per tablet) were procured from Sun Pharmaceutical IND LTD, Silvasa. Double distilled water was used for the preparation of solutions.

Standard solutions: Standard stock solution of Rasagiline (1000 $\mu\text{g mL}^{-1}$) was prepared in distilled water. (1.56mg of Rasagiline Mesylate is equivalent to 1 mg of Rasagiline) Working standard solutions were daily prepared by diluting stock solutions at the concentrations of 5-150 $\mu\text{g mL}^{-1}$ in distilled water.

Procedure for formulations

Twenty tablets of Rasalect were accurately weighed and finely powdered. A portion of the powder equivalent to the average weight of one tablet was transferred into a 25 mL volumetric flask and 20 mL of DW was added. The contents of the flask were sonicated for 15 min and diluted to volume with DW. The solution was filtered using whatman filter paper. Then the absorbance of the filtered solutions was measured at 271.6nm. The amount of Rasagiline per tablet was calculated using the calibration curve.

RESULTS AND DISCUSSION

Rasagiline Mesylate is freely soluble in water and ethanol. So, aqueous solution was selected for estimation and the solution was scanned in the wavelength range 190-400 nm. The spectrum showed a well-defined peak and the drug showed maximum absorbance at wavelength of 271.6nm.



Validation

Validation is one of the most important steps in method development for analytical determinations. The main validation parameters such as linearity and range, accuracy, precision, ruggedness, sensitivity and specificity were evaluated in the developed method.

Linearity and range

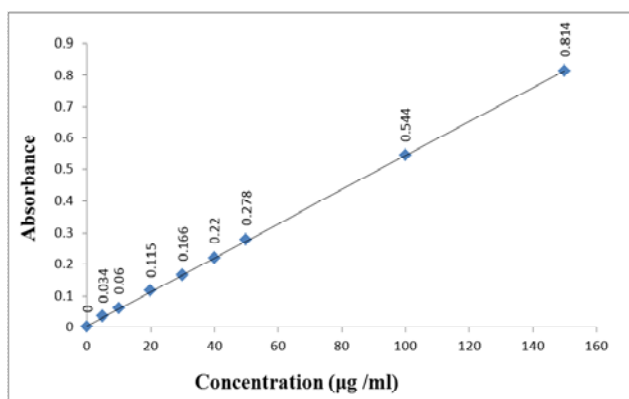


Figure 2: Calibration curve of Rasagiline Mesylate.

The calibration graph of the absorbance versus concentration was found to be linear over the range of 5-150 $\mu\text{g mL}^{-1}$. The calibration graph Figure 2 was constructed after analysis of 8 different concentrations with each concentration was measured five times. Each point of the calibration graph corresponded to the mean value obtained from 5 independent measurements.

The regression equation was $y = 0.0054x + 0.005$ where y is the absorbance and x is the concentration in $\mu\text{g mL}^{-1}$ ($r^2 = 0.9999$)

Table 1. Analytical characteristics of Rasagiline Mesylate by proposed UV spectrophotometric method (n=5).

Parameter	UV spectrophotometric method
λ_{max} nm	271.6
Beer's law limit $\mu\text{g mL}^{-1}$	5-150
Molar absorptivity $\text{Lmol}^{-1}\text{cm}^{-1}$	935.6
Regression equation	$y = 0.0054x + 0.005^*$
Correlation coefficient (r^2)	0.9999
Linearity range $\mu\text{g mL}^{-1}$	5-150
Limit of detection (LOD) $\mu\text{g mL}^{-1}$	0.5
Limit of quantification (LOQ) $\mu\text{g mL}^{-1}$	5
Number of data points	8

* $y = bx + a$ where 'x' is the concentration in $\mu\text{g mL}^{-1}$, 'y' is absorbance, 'b' is the slope, 'a' is the y- intercept

Accuracy

The accuracy of the method was assessed by spiking a drug solution of known concentrations and absorbance (30 $\mu\text{g/ml}$) with known amounts of the drug (10 μg , 15 and 20) and checked for the increase in absorbance. Solutions of the tablet formulations are also spiked with known amount of the drug. Samples were checked for the absorbance. In both the cases recovery studies were replicated three times. The accuracy was expressed in terms of percent recovery and calculated by multiplying the ratio of measured drug concentration to the expected drug concentration with 100 so has to give the percent recovery.

The mean percentage recovery of Rasagiline from drug solutions and from tablet solutions were found to be 99.1 ± 0.99 to 100.9 ± 1.801 indicating complete recovery of Rasagiline. (Table 2 and Table 3)

Table 2: Recovery of Rasagiline mesylate from standard solutions

Amount of drug added (μg)	Mean% recovered (\pm s.d.) (n=3)
10	99.10 ± 0.99
15	100.9 ± 1.801
20	100.45 ± 0.780

Table 3: Recovery of Rasagiline Mesylate from tablets sample

Amount of drug added (μg)	Mean% recovered (\pm s.d.) (n=3)
10	100 ± 1.70
20	100.90 ± 0.780

Precision

The precision of a method is defined as the closeness of agreement between independent test results obtained under optimum conditions. Three different concentrations of Rasagiline Mesylate in the linear range (30, 40 and 50 $\mu\text{g mL}^{-1}$) were analyzed in 5 independent series in the same day (intra-day precision) and 3 consecutive days (inter-day precision). The precision of the analysis was determined by calculating the Coefficient of Variation (C V). The C V values of intra-day and inter-day studies varied from 0.199 to 0.457 showed that the precision of the method was satisfactory (Table 4).

Table 4: Precision of the proposed method

Conc. of Rasagiline ($\mu\text{g/ml}$)	Conc. of Rasagiline ($\mu\text{g/ml}$) found on			
	Intraday (mean \pm s.d.) (n=5)	C V	Interday (mean \pm s.d.) (n=3)	C V
30	29.88 ± 0.09	0.314	29.88 ± 0.105	0.351
40	39.85 ± 0.10	0.249	39.76 ± 0.182	0.457
50	50.04 ± 0.10	0.199	49.95 ± 0.145	0.290



Sensitivity

The limit of quantification (LOQ) is the lowest concentration of Rasagiline Mesylate on the calibration curve that can be quantified with acceptable precision and accuracy. The LOQ was found as $5 \mu\text{g mL}^{-1}$ (CV = 1.31) (n=5) for proposed method. The limit of detection (LOD) was found to be $0.5 \mu\text{g mL}^{-1}$ (CV = 1.55) (n=5). The results indicate that the proposed method is sensitive to detect and quantify $0.5 \mu\text{g mL}^{-1}$ and $5 \mu\text{g mL}^{-1}$ respectively.

Specificity

The spectra obtained from tablet solution were identical with that obtained from standard solution containing an equivalent concentration of Rasagiline indicating that the wavelength of maximum absorbance of Rasagiline Mesylate did not change. It was concluded that the excipients did not interfere with quantification of Rasagiline Mesylate in this method and the proposed method could be considered specific as there was no spectral interaction in the analysis of Rasagiline Mesylate in pharmaceutical formulations by the proposed method. Thus the proposed method can be used for the determination of Rasagiline Mesylate in the presence of excipients.

Ruggedness

The ruggedness of the proposed method was evaluated by applying the developed procedures to assay of $30 \mu\text{g mL}^{-1}$ of Rasagiline Mesylate using the same instrument by two different analysts under the same optimized conditions at different days. The obtained results were found to be reproducible, since there was no significant difference between analysts (Table 5). Thus, the proposed methods could be considered rugged.

Table 5: The ruggedness of proposed method

Analyst 1 found $\mu\text{g mL}^{-1}$	Analyst2 found $\mu\text{g mL}^{-1}$
X:29.91	X :30.05
SD : 0.112	SD :0.15
CV :0.374	CV :0.499

x : Mean, SD : standard deviation., CV: Coefficient of Variation

Table 6: Assay of pharmaceutical formulations containing Rasagiline Mesylate analyzed by proposed method (n=5).

Tablet Solution containing	Amount Found (mg)	% found	C V
Rasagiline Mesylate 1.0mg	1.0272	102.72	0.706
	1.0181	101.81	
	1.0227	102.27	
	1.0136	101.36	
	1.009	100.9	

Analysis of pharmaceutical formulations

The optimized spectrophotometric method was applied to the direct determination of Rasagiline Mesylate in tablet formulation. The average amount present was determined by taking average of five replicate analysis and the amount present were found to be 1.01 mg/tab. Coefficient of variation was found to be 0.706. The results show that the proposed method was successfully applied for the assay of Rasagiline Mesylate in its pharmaceutical formulations (Table 6).

CONCLUSION

In this study a rapid, accurate, precise, sensitive and selective method was developed for the determination of Rasagiline Mesylate in pharmaceutical formulations and validated as per the ICH guidelines. Moreover, this method is simple and inexpensive and it can be employed for the routine quality control of Rasagiline Mesylate in pharmaceutical formulations

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REFERENCES

- Sean C Sweetman, Martindale, The complete Drug Reference, 36th Ed, Pharmaceutical Press, 2009, 815.
- Thebault J, Guillaume M, Levy, Pharmacotherapy, 24, 2004, 1295- 1305.
- Fernandez M, Barcia E , Negro S, Journal of Pharmaceutical and Biomedical Analysis, 49, 2009, 1185-91.
- Chen X, Duan X, Ma J, Deng P, Wang H, Zhong D, Journal of Chromatography B, 873(3), 2008, 203-85.
- The United States Pharmacopoeia, 22th Revision, Rockville: 2007, 1711.
- ICH, Topic Q 2B, Validation of Analytical Procedures: Methodology, Step 4, consensus Guideline, 1996.
- Jeffery GH, Mendham J , Denny RC, Vogel's textbook of quantitative chemical analysis, 5th ed, John Wiley and sons, 1989, 135.
- Jack J. Chen, David M. Swope, Khashayar Dashtipour, Clinical Therapeutics; Volume 29, Issue 9, 2007, 1825-1849.
- Youdin, Gross A , Finberg J.P, Br. J. pharmacol, 132, 2001, 500-506.
- Fazlul Huq, Journal of Pharmacology and Toxicology. 3(2), 2008, 168-172.

