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Original Article

Analytical Method Development and Validation of Rivastigmine in its Pure and Pharmaceutical Dosage form Using UPLC

Aarthi Kommu and Raja Sundararajan*

Department of Pharmaceutical Quality Assurance, GITAM School of Pharmacy, GITAM (Deemed to be University), Visakhapatnam, Andhra Pradesh, INDIA.

ABSTRACT

Objectives: For the quantification of rivastigmine in both its pure form and pharmaceutical formulation, an ultra-performance liquid chromatography method with high speed and sensitivity was created and validated**. Materials and Methods:** Acquity UPLC BEH C_8 (100 mm x 2.1 mm and 1.7 µm) employed to resolve the analysis, and the mobile phase included ammonium phosphate buffer and acetonitrile in a 65:35% v/v ratio. The column temperature was 30°C. The volume of sample injected was 10 µL. The flow rate was 0.5 mL/min, and UV detection was used to detect the analyte at 254 nm. **Results:** As there was no interferences observed by blank and placebo at the retention time of rivastigmine. According to the results of the degradation investigation, considerable degradation was seen under the conditions of alkali and oxidative stress (peroxide). This led to the conclusion that rivastigmine was susceptible to oxidation and alkali. A study using six replicate injections was carried out to obtain system precision. The predicted % RSD from the rivastigmine peak locations was determined to be 0.2%. The correlation coefficient was determined to be 0.9996, and the suggested UPLC method was linear over the range of 12.5 to 75 g/mL. The accuracy studies were displayed as a % recovery for rivastigmine levels between 50% to 150%. The results obtained were determined to be within the limits, and the maximum percentage of recovery revealed was in between 98 and 102%. As a result, the accuracy of the technique was established. The procedure remains unaffected by variations in column oven temperature and wavelength. All the validation parameters satisfy the ICH Q2 specification acceptance limits, and the technique was validated in accordance with ICH rules. **Conclusion:** According to ICH criteria, the developed technique was verified for several parameters including accuracy, precision, linearity, specificity, system compatibility, solution stability, and robustness. The results obtained met the requirements for acceptance. It was determined, then, that the proposed UPLC technology was easy to use, precise, and accurate, and that it can be successfully used for the routine analysis of rivastigmine in bulk and pharmaceutical dosage forms.

Keywords: Rivastigmine, UPLC, Method Development, Forced degradation, Validation.

Correspondence:
Prof. Raja Sundararajan
Department of Pharmaceutical Quality Assurance, GITAM School of Pharmacy, GITAM (Deemed to be University), Visakhapatnam-530 045, Andhra Pradesh, INDIA.
Email: sraja61@gmail.com

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INRODUCTION

Rivastigmine was chemically named as 3-[(1S)-1- (dimethyl amino) ethyl] phenyl N-ethyl-N-methylcarbamate. The molecular formula was $C_{14}H_{22}N_2O_2$ with molecular weight: 250.33 g/mol. In patients with Alzheimer's and Parkinson's disease, rivastigmine, a parasympathomimetic or cholinergic drug, was used to prevent and cure neurodegenerative disease, particularly dementia.[1] Rivastigmine was a carbamate derivative, unlike donepezil and tacrine, that shares no structural similarities with physostigmine. The mechanism of rivastigmine was still unknown, however it was hypothesized that it binds to and inactivates cholinesterase (such as acetylcholinesterase and butyrylcholinesterase), blocking the hydrolysis of acetylcholine and increasing the amount of acetylcholine at cholinergic synapses.[2] Rivastigmine's anticholinesterase activity was more selective for brain acetylcholinesterase than for those in peripheral tissues.[3] Figure 1 represents the rivastigmine chemical structure.

According to the literature survey, HPLC,[4,5] UV[6-8] and LC-MS/MS[9-11] methods were reported for quantification of rivastigmine. The purpose of this work was to develop a novel, specific, and accurate UPLC technique for the determination of rivastigmine both in its pure form and in capsule dosage form in compliance with ICH standards.

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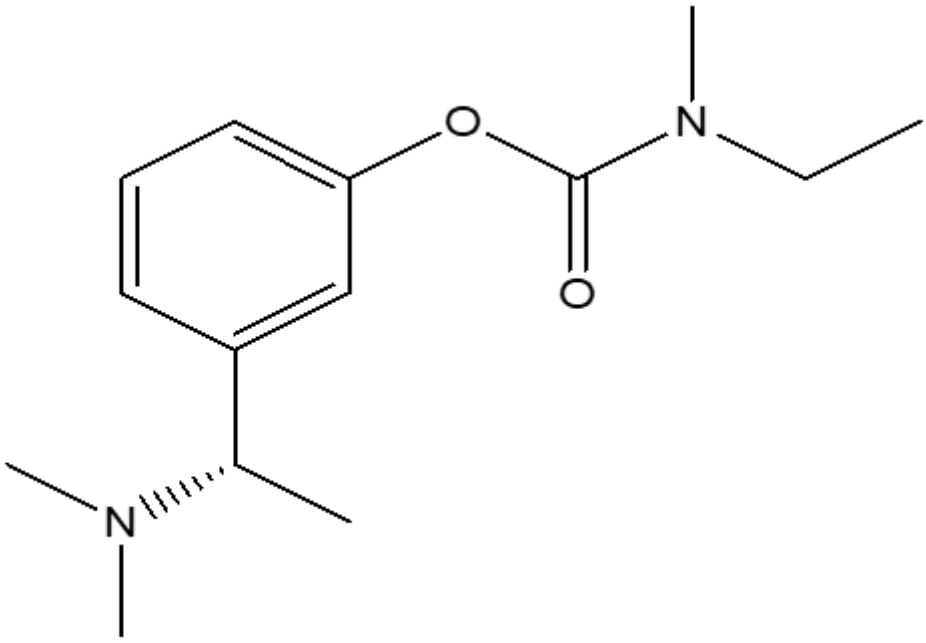


Figure 1: Structure of rivastigmine.

MATERIALS AND METHODS

Chemicals and reagents

Chandra Labs (Hyderabad) provided a free sample of rivastigmine. The chemicals used in the experiment included methanol and potassium di hydrogen orthophosphate (Merck Life Science Private Limited). Acetonitrile (Avantor Performance Materials India Limited), and disodium hydrogen phosphate (Thermo Fisher Scientific India Pvt. Ltd.) were also used in the experiment. Distilled water was prepared in the laboratory.

Instruments and Equipment

Shimadzu 1200 UPLC column system (Open Lab EZ Chrome software) equipped with quaternary pumps, and a photodiode array detector was used. A pH meter (Thermo scientific) was used to determine the pH levels. Analytical balance (Mettler Toledo) was used for all analytical measurements.

Method of Analysis

Preparation of ammonium buffer (pH 3.5)

3.0 g of ammonium phosphate was accurately weighed and added to 1 L of water. The solution was dissolved by sonication. The pH was modified by diluted orthophosphoric acid and filtered via 0.45 µm thickness of membrane filter paper.[12]

Preparation of mobile phase

The mobile phase was made by combining 350 mL of acetonitrile and 650 mL of buffer. The gas was eliminated by sonicating the solution. A 0.45 µm membrane filter paper was used to filter mobile phase.

Preparation of stock solutions

50 mg of rivastigmine was precisely weighed and transferred into 50 mL volumetric flask. The volume was made using mobile phase.

Preparation of working standard solutions

From the above stock solution, 100 µg/mL of rivastigmine was prepared by transferred 5 mL of the stock solution into 50 mL of a volumetric flask. The volume was made up with mobile phase respectively.

Preparation of sample solution

The weight of the 20 capsules was recorded, and the powder was collected in a plastic bag.

The empty capsule shells were weighed, and the average weight of the full powder was calculated by the formula below.

20 capsules powder weight = (20 capsules weight with filled powder - weight of the capsule shells without filled powder)

The average weight was determined. Rivastigmine powder corresponding to 10 mg of the drug was precisely measured and transferred into a 100 mL volumetric flask. To the volumetric flask, 70 mL of mobile phase was added and sonicated for 20 min to get dissolved.

For 10 min, the sample solution was centrifuged at 5000 rpm and the volume was adjusted with the mobile phase.

Preparation of placebo solution

Placebo powder was weighed equivalent to 10 mg of rivastigmine and transferred into 100 mL of volumetric flask. 70 mL of mobile phase was added and sonicated for 20 min to get dissolved.

For 10 min, the sample solution was centrifuged at 5000 rpm and the volume was adjusted with the mobile phase.

Instrumentation

The method development and validation of UPLC was carried out on an Acquity UPLC BEH C_8 (100 mm x 2.1 mm and 1.7 µm), with mobile phase was composed of ammonium phosphate buffer and acetonitrile in the ratio of (65:35% v/v), and flow rate of 0.5 mL/min. The column temperature was 30°C. The volume of sample injected was 10 µL. From the UV spectrum of rivastigmine, 254 nm was chosen as a wavelength. The eluted compounds were monitored at 254 nm. The chromatographic conditions were illustrated in Table 1 and the optimized chromatogram was represented in Figure 2.

RESULTS

Method expansion and optimization of chromatographic conditions

UV-spectroscopic analysis of rivastigmine drug substance showed UV absorbance (λ_{max}) at 254 nm correspondingly.

A simple and robust UPLC technique was developed for the determination of rivastigmine in its pure and in capsules dosage form, dissimilar mobile phases and columns were employed to achieve a good peak shape.

The technique expansion (first trial) was started with Acquity BEH C_{18} (100 mm×2.1mm and 1.7 µm) with the mobile phase composition of ammonium acetate pH 4.0 buffer: methanol in the proportion of 70:30 v/v. It was examined that peak shape was not good. The column stationary phase was not appropriate for the component. The second trial was performed by change in the column Acquity BEH phenyl (100 mm×.2.1 mm and 1.7 µm). The rivastigmine was eluted at void volume and the peak shape was not good. The third trial was performed by change in the mobile phase, ammonium phosphate buffer pH 6.0: methanol in the proportion of 70:30 v/v. The rivastigmine was injected, peak shape was good,

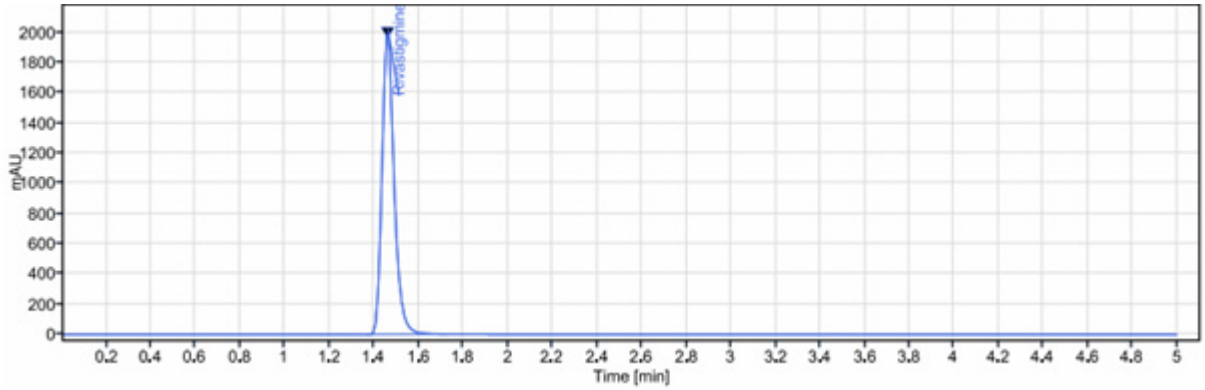


Figure 2: Optimized chromatogram of rivastigmine.

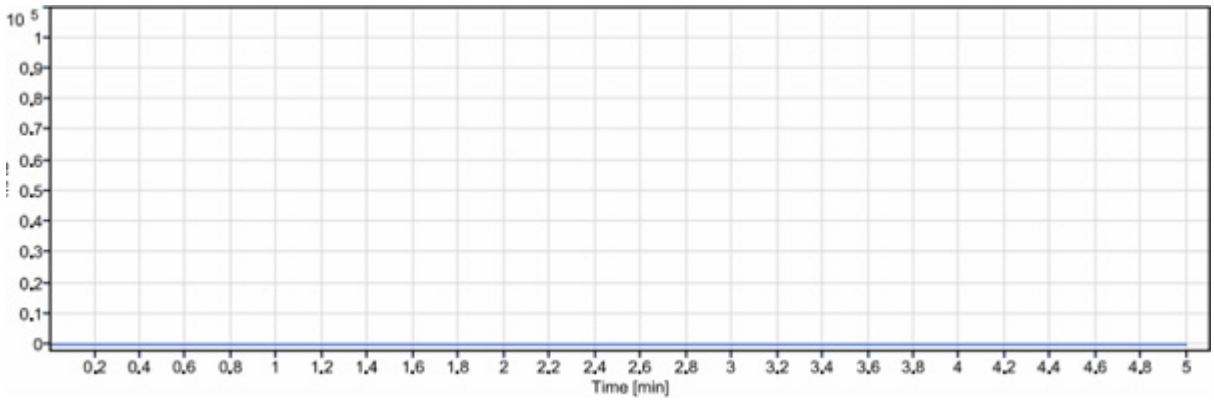


Figure 3: Chromatogram of blank.

Table 1: Optimized chromatographic conditions.

Parameter	Chromatographic conditions
Stationary phase	Acquity UPLC BEH C_8 (100 mm×.2.1 mm and 1.7 µm)
Mobile phase	Ammonium phosphate buffer pH 3.5: acetonitrile 65:35% (v/v)
Injection volume	5 µL
Total run time	10 min
Detector	Photodiode array detector
Elution	Isocratic mode
Flow rate	0.5 mL/min
λ_{max}	254 nm

Table 2: System suitability parameters.

Parameter	Observation
Retention time	1.460 min
Theoretical plates	3632.447
Tailing factor	1.3

Table 3: Specificity data of rivastigmine.

Sl. No.	Solution details	Area of rivastigmine
1	Standard	7524.21
3	Blank	Not detected
4	Placebo	Not detected
5	Test solution	7504.21

Table 4: System precision data of rivastigmine.

Name of the Standard	Area of rivastigmine
Standard-01	7544.39
Standard-02	7539.38
Standard-03	7506.33
Standard-04	7505.25
Standard-05	7524.67
Standard-06	7525.37
Average	7524.01
% RSD	0.2

but eluted at 2.0 min with tailing more than 1.8. The final trial was performed by the column Acquity BEH C_8 (100 mm×.2.1mm and 1.7 µm), the mobile phase composed of ammonium phosphate buffer pH 3.5: acetonitrile in the proportion of 65:35 v/v, eluted at 1.5min and peak shape was good and the efficiency was more than 2000 for rivastigmine. Hence this method was optimized. Figure represents the optimized chromatogram of rivastigmine.

System Suitability Parameters

The system's suitability was assessed by six replicate injections of the drug standard solution (100 µg/mL). Parameters like tailing factor, plate count and column efficiency were noted. Table 2 displayed the data on system suitability parameters.

Specificity

A drug peak response and a placebo response were examined for interference in the obtained chromatograms.[13] There was no interference in the placebo or blank samples. Table 3 showed the specificity data and the chromatograms were represented in Figures 3 and 4.

System Precision

Chromatogram data for system precision revealed that % RSD was found to be 0.2. The precision data was shown in the Table 4.

Method precision

Chromatogram data for method precision revealed that the % RSD was found to be 0.8. The precision data for each method was shown in Table 5.

Intermediate precision results

Chromatogram data for intermediate precision revealed that % RSD was found to be 0.5. Table 6 displayed the intermediate precision data.

Ruggedness

The ruggedness chromatogram data revealed that the % RSD was 0.42. Table 7 displayed data on ruggedness.

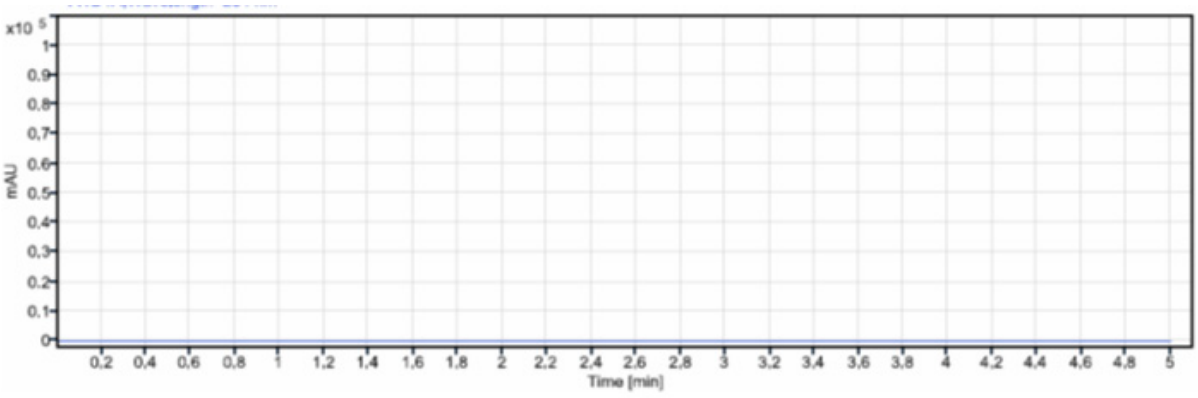


Figure 4: Chromatogram of placebo.

Table 5: Method precision data of rivastigmine.

Sl. No.	Solution details	% Assay of rivastigmine
1	Test solution preparation 1	99.6
2	Test solution preparation 2	99.9
3	Test solution preparation 3	101.1
4	Test solution preparation 4	100.4
5	Test solution preparation 5	99.9
6	Test solution preparation 6	101.8
Average		100.5
Std deviation		0.81
% RSD		0.80

Table 6: Intermediate precision data of rivastigmine.

Sl. No.	Solution details	% Assay of rivastigmine
1	Test solution preparation 1	99.9
2	Test solution preparation 2	98.5
3	Test solution preparation 3	99.3
4	Test solution preparation 4	99.6
5	Test solution preparation 5	99.8
6	Test solution preparation 6	99.7
Average		99.5
% RSD		0.5

Linearity

Linearity was obtained using the suggested UPLC technique at concentrations that range from 50 to 150 µg/mL. The Linearity equation of rivastigmine was found to be y = 107.24x-3163.6, with a correlation coefficient of 0.9996. The linearity data was shown in Table 8 and the calibration data was shown graphically in Figure 5.

Table 7: Ruggedness data of rivastigmine.

Sl. No.	Solution details	% Assay of rivastigmine
Analyst-01	Test solution preparation 1	99.6
	Test solution preparation 2	99.9
	Test solution preparation 3	101.1
	Test solution preparation 4	100.4
	Test solution preparation 5	99.9
	Test solution preparation 6	101.8
Analyst-02	Test solution preparation 1	99.9
	Test solution preparation 2	98.5
	Test solution preparation 3	99.3
	Test solution preparation 4	99.6
	Test solution preparation 5	99.8
	Test solution preparation 6	99.7
Average		99.9
Std deviation		0.47
% RSD		0.42

Accuracy and Recovery

Recovery studies helped to determine the accuracy of the method. The reference standards for the drugs were added to the formulation (pre-analyzed sample) at levels of 50%, 100%, and 150%. The percentage recovery and % mean recovery was computed for the drug that were shown in Table 9.

Sensitivity

LOD and LOQ of rivastigmine were found to be 1.1 µg/mL and 3.4 µg/mL, respectively.

Robustness

Robustness was carried out with 10 µg/mL of rivastigmine and the % RSD was found to be in the range of 0.2-0.9. The results were shown in Table 10.

Forced Degradation studies

Rivastigmine was degraded by acid (0.6%), alkali (1.2%), oxidation (5.8%), photolytic degradation (1.0%), and thermal degradation (0.5%). The results showed that rivastigmine was more resistant to all the forced degradation conditions tested. The outcomes were shown in Table 11. The chromatograms for acid degradation, basic degradation, oxidation, thermal degradation, and photolytic degradation were enumerated in Figures 6-10. The summary of all the validation parameters was displayed in Table 12.

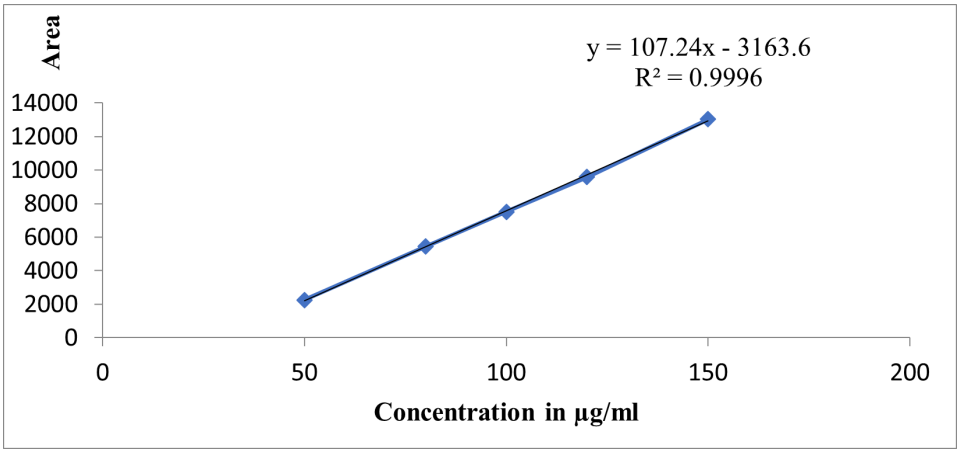


Figure 5: Linearity graph.

Table 8: **Linearity data of rivastigmine.**

Sl. No	Concentration (µg/mL)	Name of the solution	Area of rivastigmine
1	50	Linearity solution, Level-1	2245.62
2	80	Linearity solution, Level-2	5429.26
3	100	Linearity solution, Level-3 (100%)	7517.5
4	120	Linearity solution, Level-4 (120%)	9585.91
5	150	Linearity solution, Level-5 (150%)	13022.47
Slope			107.24
Intercept			3136.3
Correlation coefficient			0.9996

Table 9: **Accuracy and recovery data of rivastigmine.**

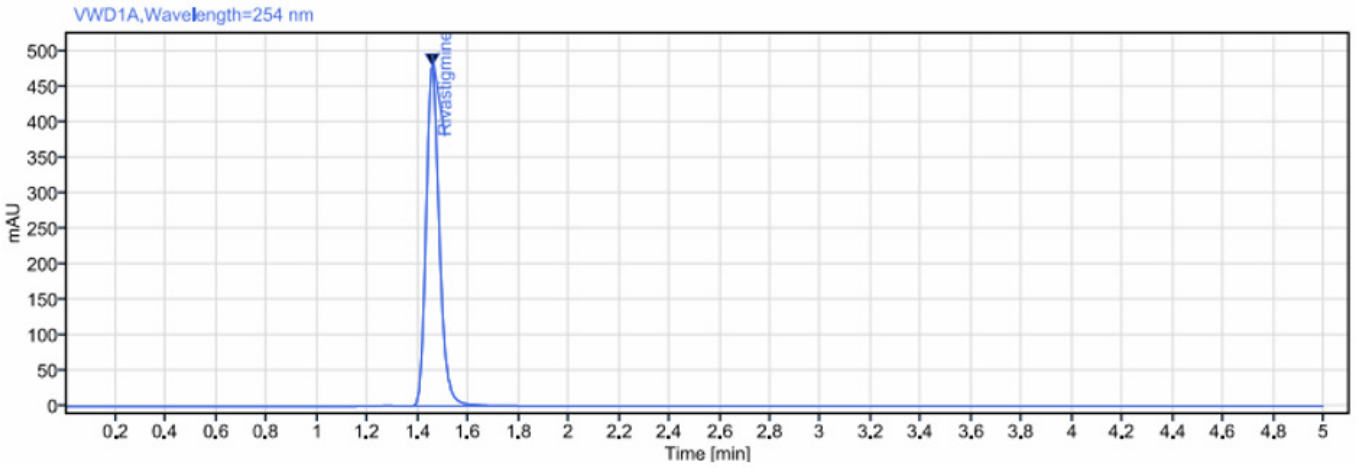
Name of the solution	% Recovery of rivastigmine
Recovery-50%-01	98.1
Recovery-50%-02	100.4
Recovery-50%-03	100.5
Recovery-100%-01	100.9
Recovery-100%-02	101.0
Recovery-100%-03	99.7
Recovery-150%-01	101.0
Recovery-150%-02	101.0
Recovery-150%-03	100.9
Average	100.7
Std deviation	0.96
% RSD	1.0

Table 10: **Robustness data of rivastigmine.**

Name of the parameter	% RSD	Theoretical plates	Tailing factor
Low column oven temperature(35°C)	0.2	3524	1.34
Low column oven temperature(45°C)	0.9	3632	1.36
Lower wavelength (249 nm)	0.8	3637	1.37
Higher wavelength (269 nm)	0.7	3634	1.38

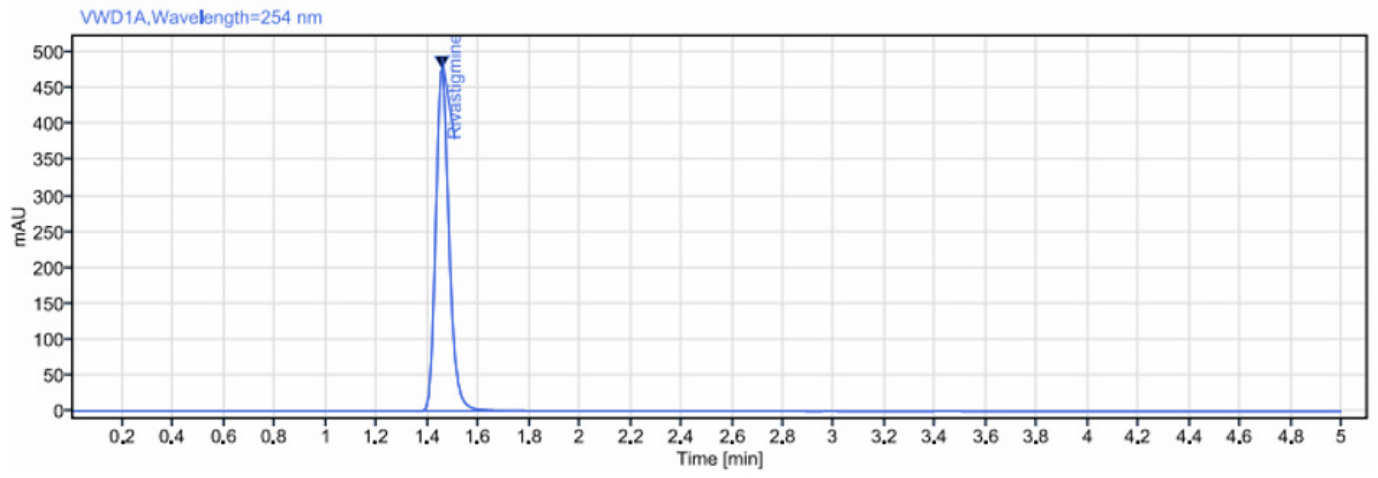
Table 11: Forced degradation studies data of rivastigmine.

Sl. No.	Degradation condition	% Drug undegraded	% Drug degraded	Retention time	Peak area
1	Acid	99.3	0.6	1.456	1763.26
2	Alkali	98.7	1.2	1.456	1744.33
3	Oxidation (peroxide)	94.1	5.8	1.453	11369.99
4	Photo stability	98.9	1.0	1.457	8398.80
5	Thermal	99.4	0.5	1.456	1744.35



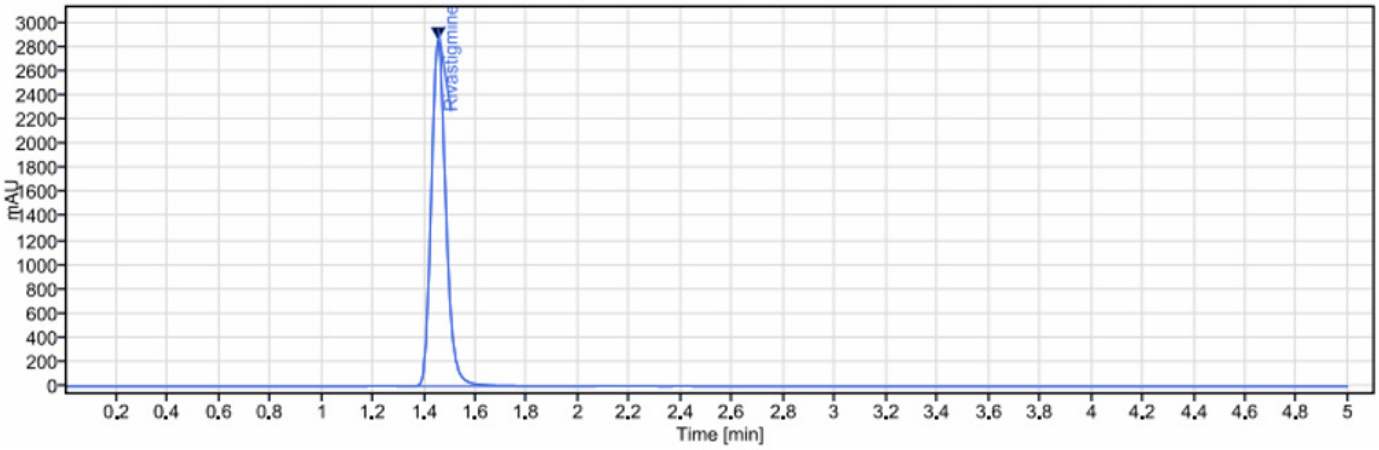
Compound Name	Peak Retention Time	Area	Height	Area%	Peak Tail Factor	Peak Plates Per Meter USP
Rivastigmine	1.456	1763.26	479.63	100.00	1.19926	3864.51511

Figure 6: Chromatogram of acidic degradation of rivastigmine.



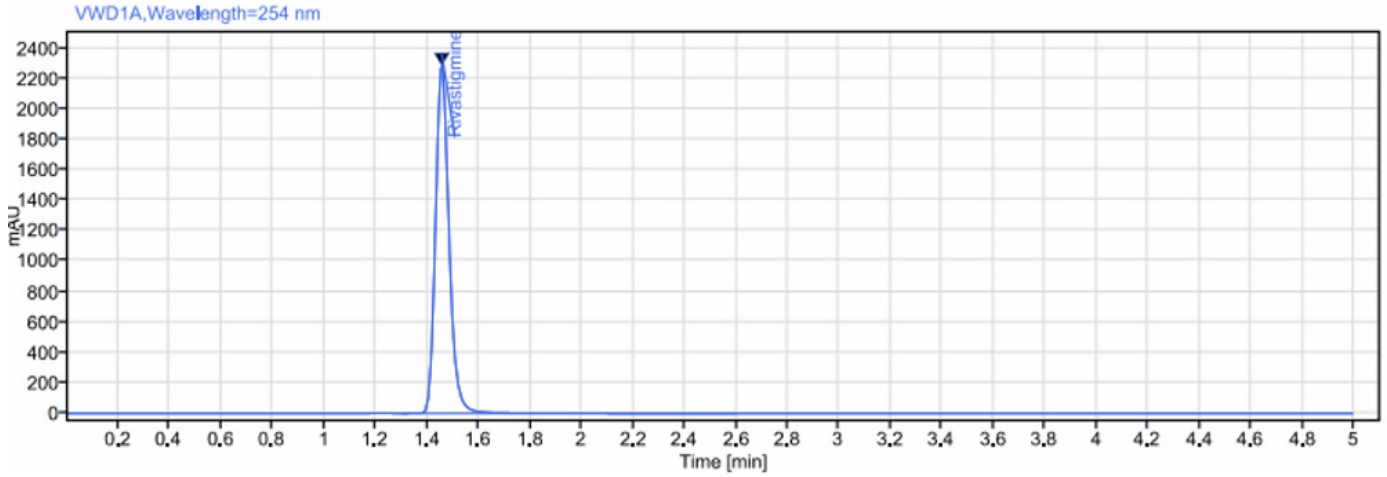
Compound Name	Peak Retention Time	Area	Height	Area%	Peak Tail Factor	Peak Plates Per Meter USP
Rivastigmine	1.456	1744.33	476.83	100.00	1.19786	3872.15711

Figure 7: Chromatogram of basic degradation of rivastigmine.



Compound Name	Peak Retention Time	Area	Height	Area%	Peak Tail Factor	Peak Plates Per Meter USP
Rivastigmine	1.453	11369.99	2853.09	100.00	1.21515	3229.50153

Figure 8: Chromatogram of peroxide degradation of rivastigmine.



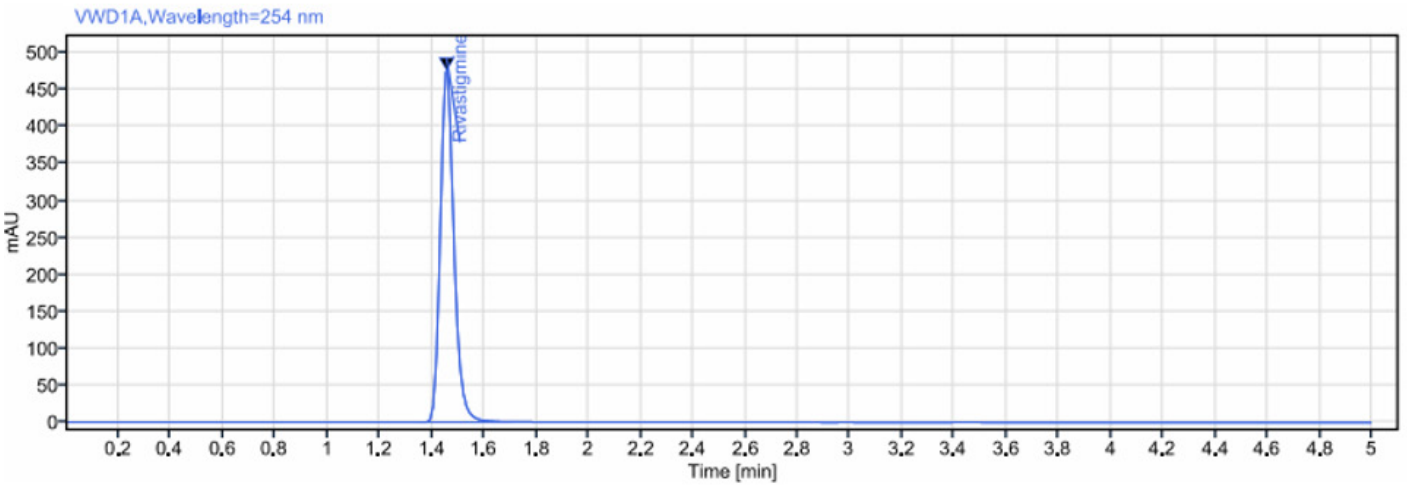
Compound Name	Peak Retention Time	Area	Height	Area%	Peak Tail Factor	Peak Plates Per Meter USP
Rivastigmine	1.457	8398.80	2280.61	100.00	1.19586	3811.75154

Figure 9: Chromatogram of photolytic degradation of rivastigmine.

DISCUSSION

A system suitability test is necessary as a component of the method development, used to ensure that the suitability for rivastigmine analysis. Prior to analyzing samples on each day, a proper procedure has been developed to evaluate the potential of UPLC instrument to conduct techniques that yield results of acceptable accuracy and precision.[14] The system suitability system results revealed that the theoretical plates were >2000 and the tailing factor was < 2.

Specificity was evaluated by using a blank and placebo solution. At the retention time of the standard rivastigmine sample, no interference observed in the placebo or blank samples.[14] The chromatogram data for system and method precision found to be



Compound Name	Peak Retention Time	Area	Height	Area%	Peak Tail Factor	Peak Plates Per Meter USP
Rivastigmine	1.456	1744.35	475.71	100.00	1.19786	3872.15712

Figure 10: Chromatogram of thermal degradation of rivastigmine.

Table 12: Summary.

Parameters		Values
Linearity (µg/mL)		50-150 (µg/mL)
Regression coefficient		0.9996
Slope (m)		107.24x
Intercept (c)		3163.6
Regression equation (y= mx +c)		y = 107.24x+3163.6
Specificity		Specific
System precision (% RSD)		0.2
Method precision (% RSD)		0.8
Intermediate precision (% RSD)		0.5
Ruggedness		0.42
LOD		1.1 µg/mL
LOQ		3.4 µg/mL
Robustness	Low column oven temperature (35°C)	0.2
	High column oven temperature (45°C)	0.9
	Lower wavelength (249 nm)	0.8
	Higher wavelength (269 nm)	0.7

within the specified limit (% RSD NMT 2.0%). As a result, it was shown that the method was found to be precise.

The chromatogram data for intermediate precision found to be within the specified limit (% RSD NMT 2.0%). As a result, the procedure was determined to be precise.[15] The ruggedness chromatogram data was found to be within the specified limit (% RSD NMT 2.0%). As a result, it shown that the method was determined to be rugged.

Dissimilar drug standard solutions were made to evaluate the linearity by diluting the drug stock solutions with diluents in

different concentrations of rivastigmine ranging from 12.5 to 75 µg/mL. By using linear regression analysis, the calibration curve of linearity plot was evaluated.[15] By the standard addition method, three levels of accuracy samples (50%, 100%, and 150%) were created. The percentage recovery was ranged from 98% to 102%. The recovery results demonstrated that it can be used for quality control of capsule dosage forms. The technique was established to be accurate.[16] The lowest limits for detection and quantification were established by means of the subsequent equations based on the slope of calibration and its standard deviation responses using different concentrations of the standard stock solution.

$$\text{Limit of detection} = 3.3 \times \text{SD of the response} / \text{slope of the calibration curve.}$$

$$\text{Limit of quantification} = 10 \times \text{SD of the response} / \text{slope of the calibration curve.}^{16}$$

The signal-to-noise (s/n) ratio is taken into consideration to calculate limit of detection and quantification, with limit of detection defined as approximately s/n ~3 and limit of quantification defined as the lowest validated concentrations with (%) RSD and (%) error ≤ 20% and the results were found to be within the specified limits.[16] The estimation of robustness by various chromatographic conditions, that include temperature and wavelength changes. Samples were injected into UPLC system and the % RSD was determined and the results were found to be within the limits.

Degradation studies revealed that the significant degradation was scrutinized in alkali and oxidation stress circumstances. Hence it can be concluded that rivastigmine was sensitive to alkali and oxidation.[17]

CONCLUSION

This newly created technique for measurement of rivastigmine was shown to be easy to understand, reliable, precise, and high resolution. This method will be successfully used for routine analysis at research institutions, quality control departments in intended industries, and approved testing laboratories studies because of the method's shorter retention time, which also made it more acceptable and cost-effective.

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CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

1. Abhilash KD, George TG. Rivastigmine for Alzheimer's disease. Expert Rev Neurother. 2014;5(5):563-80.
2. Shah BM, Misra M, Shishoo CJ, Padh H. Nose to brain microemulsion-based drug delivery system of rivastigmine: formulation and *ex vivo* characterization. Drug Deliv. 2015;22(7):918-30. doi: 10.3109/10717544.2013.878857, PMID 24467601.
3. Salatin S, Barar J, Barzegar-Jalali M, Adibkia K, Kiafar F, Jelvehgari M. Development of a nanoprecipitation method for the entrapment of a very water-soluble drug into Eudragit RL nanoparticles. Res Pharm Sci. 2017;12(1):1-14. doi: 10.4103/1735-5362.199041, PMID 28255308.
4. Alexandar S, Rohini D, Ashok T, *et al*. A validated RP-HPLC method for estimation of rivastigmine in pharmaceutical formulations. Pharm Lett. 2011;3(3):421-6.
5. Avijit C, Vasanta KP, Suddha D, *et al*. RP-HPLC method for the estimation of rivastigmine in bulk and in dosage forms. J Pharm Res. 2011;4(4):1007-9.
6. Fathima MZ, Sundhararajan R, Roosewelt C. A validated method for estimation of rivastigmine pure and its pharmaceutical formulations by UV Spectroscopy. Int J Sci Health Res. 2022;7(2):372-8.
7. Kulkarni AS, Chandrashekhar VB, Amol NJ. Development of UV spectrophotometric method for estimation of rivastigmine in pharmaceutical dosage form. Int J Pharm Res Scholars. 2018;6(4):59-64.
8. Bhavyasri K. Development and validation of UV- visible spectrophotometer method for estimation of rivastigmine in human plasma. Int J Adv Res. 2019;7(5):954-9. doi: 10.21474/IJAR01/9119.
9. Jignesh B, Subbaiah G, Sandeep K, *et al*. A rapid and sensitive Liquid Chromatography–tandem Mass Spectrometry (LC–MS/MS) method for the estimation of rivastigmine in human plasma. J Chromatogr B. 2007;852(1-2):115-21.
10. Pommier F, Frigola R. Quantitative determination of rivastigmine and its major metabolite in human plasma by liquid chromatography with atmospheric pressure chemical ionization tandem mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci. 2003;784(2):301-13. doi: 10.1016/s1570-0232(02)00816-4, PMID 12505778.
11. Thomas S, Shandilya S, Bharati A, Paul SK, Agarwal A, Mathela CS. Identification, characterization, and quantification of new impurities by LC–ESI/MS/MS and LC–UV methods in rivastigmine tartrate active pharmaceutical ingredient. J Pharm Biomed Anal. 2012;57:39-51. doi: 10.1016/j.jpba.2011.08.014, PMID 21880452.
12. Indian pharmacopoeia. Vol. 1. Government of India, Ministry of Health. Pharmacopoeia of India: (the Indian pharmacopoeia). Delhi: Manager of Publications; 2014. Buffer solutions; p. 757-8.
13. Muriel N, Nicolas A, Maria D, *et al*. Simultaneous Determination of antidementia drugs in human plasma: procedure transfer from HPLC–MS to UPLC–MS/MS. J Pharm Biomed Anal. 2012;64(4):6-25.
14. Deshpande GR, Roy AK, Rao NS, Rao BM, Rudraprasad Reddy J. Rapid screening of volatile ion-pair reagents using UHPLC and robust analytical method development using DoE for an acetyl cholinesterase inhibitor: galantamine hbr. Chromatographia. 2011;73(7-8):639-48. doi: 10.1007/s10337-011-1970-1.
15. Raju TS, Kalyanaraman L, Reddy VV, Swamy PY. Development and validation of an UPLC method for the rapid separation of positional isomers and potential impurities of rivastigmine hydrogen tartrate in drug substance and drug product. J Liq Chromatogr Relat Technol. 2012;35(7):896-911. doi: 10.1080/10826076.2011.613143.
16. Noetzli M, Ansermot N, Dobrinas M, Eap CB. Simultaneous determination of antidementia drugs in human plasma: procedure transfer from HPLC–MS to UPLC–MS/MS. J Pharm Biomed Anal. 2012;64-65:16-25. doi: 10.1016/j.jpba.2012.02.008, PMID 22410501.
17. ICH Expert Working Group. Validation of Analaytical Procedures: Text and Methodology Q2(R1), Version 4. 2005.

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