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Research article

Spectrophotometric quantification of Granisetron in bulk drug and pharmaceutical formulations employing multivariate calibration technique

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ABSTRACT

The study goal is to establish a sensitive, accurate, simple and validated Ultra violet (UV) spectrophotometric technique by multilinear regression method for the assessment of Granisetron in the bulk drug and pharmaceutical formulations. The multivariate calibration technique relies on linear regression equations and employs correlation between absorbance and concentration at five different wavelengths. The λmax of Granisetron was at 302 nm. The results were treated statistically. A linear plot was achieved in the concentration range of 10 - 30 µmol/L and regression co-efficient was obtained to be 0.999. The percentage recovery was obtained in the range of 99.48 - 101.81 % w/w.

Keywords: Granisetron, Antiemetic, UV spectrophotometer, Multivariate calibration, ICH guidelines

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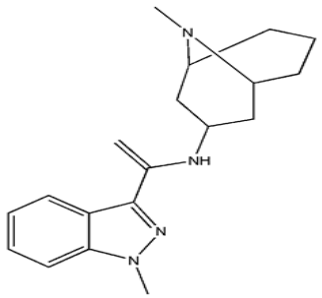
INTRODUCTION

Granisetron hydrochloride is an antiemetic widely used in the patients undertaking chemotherapy and radiotherapy. Excitation of 5-Hydroxy tryptamine3 receptors triggers nausea and vomiting. Granisetron suppresses nausea and vomiting by blocking the action of serotonin at 5HT 3 receptors (1). Granisetron (Figure 1), 1-Methyl-N-[(1R, 3r, 5S)-9-methyl-9-azabicyclo [3.3.1] non-3-yl]-1H-indazole-3-carboxamide hydrochloride is a indazole derivative. Chemical Formula is C18H24N4O and molecular weight is 348.9 g/mol (2). Granisetron has its receptors both centrally and in GIT. The drug Granisetron is official in USP and BP (3,4). The literature surveys revealed various analytical methods like UV and Visible spectrophotometry (5-12), HPLC (13-15), HPTLC (16) and LC-MS/MS [17, 18] for the estimation of the drug in formulations and in human plasma.

Figure1: Chemical structure of Granisetron



The suggested technique is based totally on the direct evaluation of Granisetron with high accuracy and precision. This technique is simple, with less cost and it can be used to bulk drug and in dosage forms. Multivariate standardization method shows the shift of single species analysis from a single defendant variable to 'm' defendant variables. E.g. sensors or wavelengths that can be concurrently comprised within the calibration model (19-25).

Under the optimized circumstances, the applied analytical technique gives great resolving power, rapidity, sensitivity and low price for quantitative and regular analysis of an investing admixture.

In this case, if the absorbance of an analyte(X) i.e. Granisetron, is scanned at 5 wavelengths set (λ = 298, 300, 302, 304 and 306 nm), the subsequent formula can be applied for any preferred wavelength.

$A\lambda 298 = a \times Cx + k1$ ..... (1)

$A\lambda 300 = b \times Cx + k2$ ..... (2)

$A\lambda 302 = c \times Cx + k3$ ..... (3)

$A\lambda 304 = d \times Cx + k4$ ..... (4)

$A\lambda 306 = e \times Cx + k5$ ..... (5)

Where, Aλ represents the absorbance of a, b, c, d, e are the slopes of the linear regression functions of the analyte; k1, k2, k3, k4, k5 are the intercepts of the linear regression at the five preferred

wavelengths and Cx is the concentration of the analyte. The above five equation systems (1-5) can be concised as

AT = a x Cx+ b x Cx+ c x Cx+ d x Cx+ e x Cx+ KT……. (6)

The above equation can be further simplified to

AT = Cx (a+b+c+d+e) + KT…… (7)

Where AT and KT act as the amount of the absorbance acquired and the amount of intercepts of regression equations at five wavelength individually. The concentration of the analyte X in a solution can be computed by the formula.

$$C_x = \frac{A_T - K_T}{(a + b + c + d + e)}$$

MATERIALS AND METHODS

Chemicals and solvents

Distilled Water

Analytical grade Granisetron was obtained ex gratia from Ideal analytical and Research Institute production, Pondicherry. The marketed tablet formulation used was Kytril, Roche Pharmaceuticals, India, (Label claim –2 milligram Granisetron), procured from the local market.

Instrumentation

LAB INDIA 3092 UV-Visible double beam spectrophotometer

Ultra Sonicator

Analytical balance

Method development

Solvent selection

Granisetron was freely soluble in distilled H_2O. Hence it was used as a solvent to dissolve the standard drug and sample.

Preparation of the standard stock solution

Standard stock solution of Granisetron was prepared by liquefying 100 mg of reference drug in 50 mL of distilled water and then finally made up to the mark in a 100 mL standard flask with distilled water. Different concentrations 10 - 30 µmol/L of solution was prepared from this standard stock solution.

Determination of λmax

The stock solution is further diluted appropriately with distilled H2O to acquire a concentration of 20 µmol/L. This was scanned from 200 - 400 nm. The λmax was obtained as 302 nm (Figure 2). A linear curve was obtained when the absorbance was plotted against the concentration (Table 1). The solutions were scanned across the range surrounding 302 nm i.e., 298, 300, 302, 304, 306 nm to enhance the correlation and to reduce the instrumental error.

Preparation of sample solution

Accurately weigh and powder 20 tablets of Granisetron and weight corresponding to 10 mg was measured and dissolved in 10 mL of distilled water to obtain 1 mg/mL solution. This solution was filtered and used for further analysis.

Method Validation

As per ICH Q2B guidelines the technique was validated for sensitivity, accuracy, precision and linearity [26].

Linearity

The different concentrations over the range of 10 - 30 µmol/L were prepared from the standard stock solution of Granisetron. In order to minimize the instrumental fluctuations and to improve the correlation these solution were scanned in the wavelength surrounding its absorbance maxima. i.e. 298, 300, 302, 304, 306 nm. The absorbances were recorded and the standardization graphs were obtained by plotting the concentration vs absorbance. (Figure 3, Table 1).

Table 1: Multivariate UV calibration obtained at five wavelengths

Conc. (µg/ml)	Absorbance (AU)				
	298 nm	300 nm	302 nm	304 nm	306 nm
10	0.322	0.329	0.331	0.327	0.316
15	0.505	0.516	0.519	0.513	0.495
20	0.646	0.661	0.665	0.657	0.634
25	0.815	0.833	0.842	0.831	0.801
30	0.966	0.987	0.995	0.982	0.948

By calculating the LOD and LOQ using the below formulae, the susceptibility of the technique was determined.

LOD = 3.3 σ/S …… (8)

LOQ = 10 σ /S …… (9)

Whereas, σ is the standard deviation (SD) of the lowest concentration and S is the slope of standard curve.

Precision

To assess the intraday and interday precision, 20 µmol/L was scanned six times in a short interval of time on same day for intraday precision and six different days for interday precision.

Accuracy

The recovery study of the suggested technique was resolved by standard addition method at 80%, 100% and 120 %. The stock solution of sample and the standard was prepared. 1 mL of standard solution was pipetted into a three standard flask, add 2.2, 3, 3.8 mL sample solution and make up to the volume of 10 mL with distilled water. The percentage recovery was calculated after measuring the absorbance with a UV spectrometer.

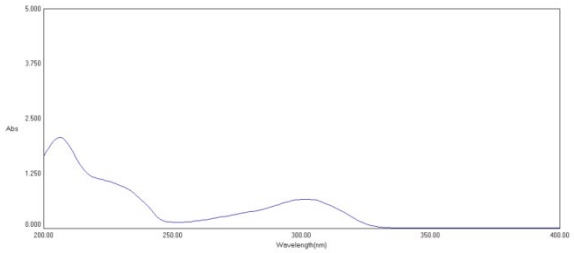
Assay

The absorbance of extracted tablet solution was recorded at 302 nm and the quantity present in the formulation was calculated as in Table4.

RESULTS AND DISCUSSION

The λmax of Granisetron was obtained at 302 nm in distilled H_2O as the solvent.

Figure 2: UV spectrum of Granisetron



The technique was linear in the selected concentration

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range over 10 - 30 µmol/L. The data from the linear regression analysis shows good linear relationship with R^2=0.999 for all the calibration plots. The equation for the linear regression is Y=0.033x + 0.01. For precision, the % relative standard deviation was 0.2013 - 0.8789. The detection limit and quantification limit obtained is 1.118 µmol/L and 3.388 µmol/L respectively. Therefore as per ICH guidelines each of the validation parameters were within the limits.

Linearity

The linearity was recorded at 298, 300, 302, 304 and 306 nm in the concentration range 10 - 30 µmol/mL and it was presented in the Figure 3 and their calibration curves and residual plots were shown in Figures 4 to 8 & 9-13 respectively. At each of the wavelength the fewer values of standard deviation shows the technique was precise and the LOD and LOQ were computed and reported in Table 2.

Figure 3: UV spectrum showing linearity of Granisetron at 302 nm

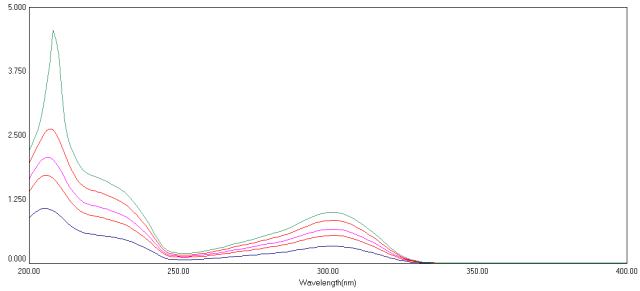


Figure 4: Calibration curve at 298 nm

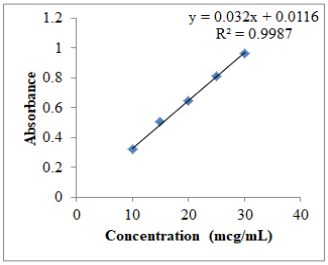


Figure 9: Residual plot at 298 nm

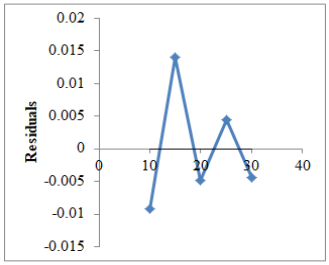


Figure 5: Calibration curve at 300 nm

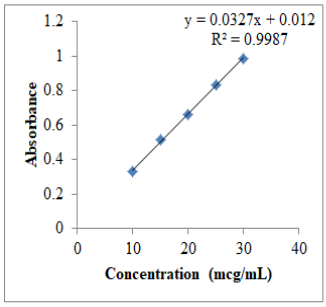


Figure 10: Residual plot at 300 nm

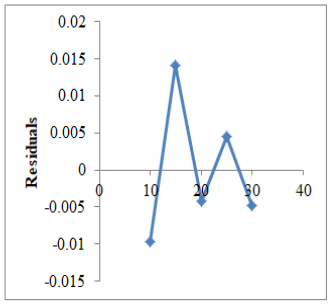


Figure 6: Calibration curve at 302nm

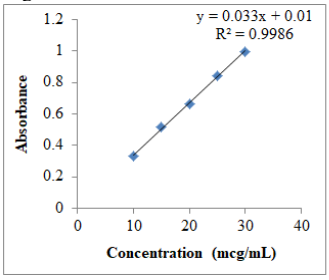


Figure 11: Residual plot at 302 nm

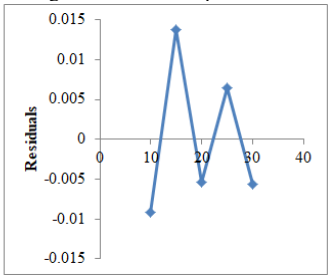


Figure 7: Calibration curve at 304 nm

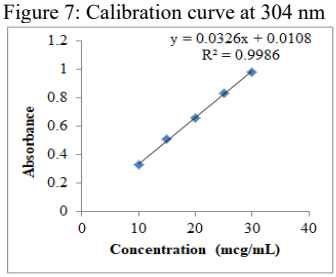


Figure 12: Residual plot at 304 nm

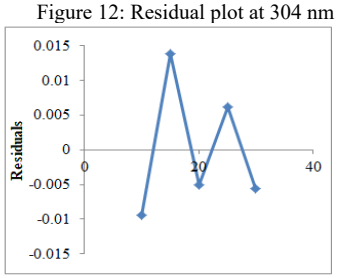


Figure 8: Calibration curve at 306 nm

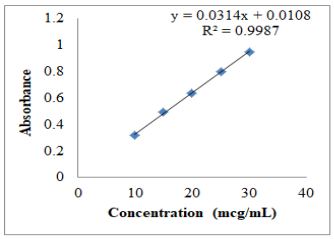


Figure 13: Residual plot at 306 nm

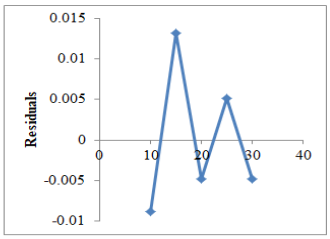


Table 2: Linearity data showing LOD and LOQ at all five wavelengths

Wavelength (nm)	Regression equation	R^2	LOD (µmol/L)	LOQ (µmol/L)	% RSD
298	Y=0.032x+0.0116	0.9987	1.102	3.339	1.6416
300	Y=0.0327x+0.012	0.9987	1.093	3.312	1.6282
302	Y=0.033x+0.01	0.9986	1.118	3.388	1.6679
304	Y=0.0326x+0.0108	0.9986	1.130	3.423	1.6857
306	Y=0.0314x+0.0108	0.9987	1.094	3.314	1.6289

Precision

The lower value of standard deviation shows that the technique was specific and percentage RSD for the inter day and intraday precision obtained were in the range of 0.2013 - 0.8789 and it lies within the limit of value less than 2% at each wavelength. The low value of percentage relative standard deviation reveals the suggested technique was accurate and precise (Figure 14, 15).

Figure 14: UV spectra showing intraday precision

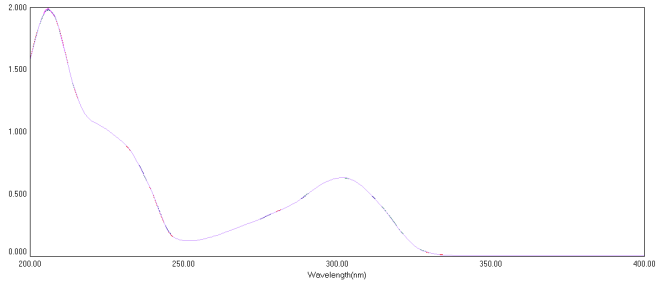
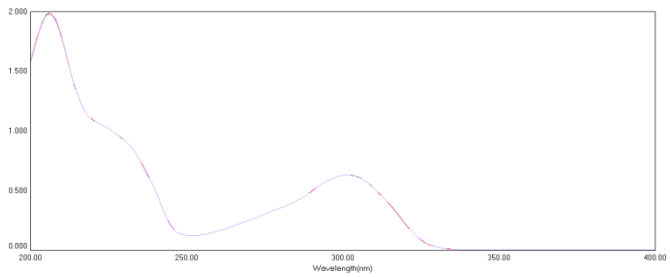


Figure 15: UV spectra showing interday precision



Recovery

As per ICH guidelines the % recovery of the Granisetron was found to be in the range of 99.48 - 101.81 % w/w, it was well within limit of 97 - 103 % w/w (Figure 16, Table 3).

Figure 16: UV Spectrum showing accuracy of Granisetron

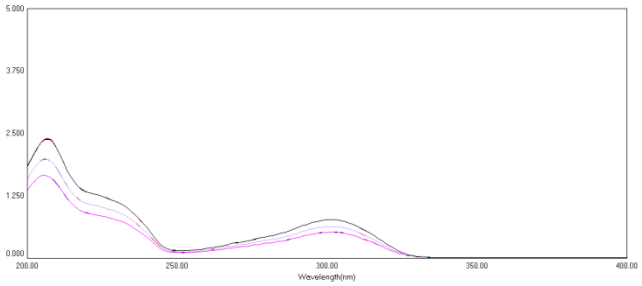


Table 3: Recovery Studies

Wavelength (nm)	Amount present (µg/mL)	Amount added (µg/mL)	Absorbance	Amount recovered (µg/mL)	% Recovery
298	5	11	0.528	15.98	99.89
		15	0.646	19.98	99.92
		19	0.752	23.88	99.48
300	5	11	0.539	16.10	100.61
		15	0.661	20.13	100.63
		19	0.769	23.99	99.96
302	5	11	0.543	16.29	101.81
		15	0.665	20.23	101.16
		19	0.775	24.06	100.24
304	5	11	0.536	16.18	101.15
		15	0.657	20.26	101.30
		19	0.766	23.99	99.96
306	5	11	0.518	16.25	101.59
		15	0.634	20.10	100.48
		19	0.738	24.15	100.63

Assay

The absorbance of the tablet formulation was recorded at 302 nm. The amount and assay percentage was found to be 1.99 mg and 99.33 % w/w with a % RSD value of 1.0478 as in Table 4.

Table 4: Assay of Granisetron

Label claim (mg)	Amount estimated (mg)	% Assay
2	1.98	99
2	2.01	100.5
2	1.97	98.5
Average	NA	99.33
SD	NA	1.0408
% RSD	NA	1.0478

CONCLUSION

The newly progressed technique was evidenced to be accurate, specific, sensitive and reproducible for the assessment of Granisetron in its pharmaceutical formulation by multilinear regression technique. The method was validated using several validation parameters and are obtained to be in the limits according to ICH guidelines. This method is a simple working procedure as comparable to the expensive techniques like HPLC, and therefore, it can be employed for regular analysis of Granisetron in bulk drug and pharmaceutical formulations.

List of symbols/abbreviations

nm = Nanometer

µmol/L = Micromole per litre

ICH = International Conference on Harmonization

UV = Ultraviolet

g/mol = Gram per Mole

GIT = Gastro Intestinal Tract

HPLC = High Performance Liquid Chromatography

HPTLC = High Performance Thin Layer Chromatography

LC/MS = Liquid chromatography-Mass spectrometry

M = Molar

CONFLICTS OF INTEREST

The authors reports on the conflict of interest on the study.

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REFERENCES

1. Rang, Maureen M Dale, 2007. Rang & Dale's Pharmacology, 7th ed., Edinburgh, Churchill Livingstone, pp 198.
2. Sean C Sweet man, 2009. Martindale, The complete Drug Reference, 37th ed, London, Pharmaceutical Press, pp 1884.
3. The United states Pharmacopoeia, 2018. The national formulary, Rockville, MD: The United States Pharmacopeial Convention, Inc, I, 1994-1995.
4. The British Pharmacopoeia, 2007. 6th ed, Stationery office on behalf of the Medicines and Health care Products Regulatory Agency (MHRA), I, 992-994.
5. Mohammed Yunooos, Seenivasan Rajesh, Sandeep V, 2014. Development and validation of a stability indicating UV spectrophotometric method for the estimation of granisetron hydrochloride in bulk and tablet dosage forms. Indo Am J Pharm Res 4, 5337-44.
6. Venkata Rao SV, Ramu G, Malleswara Rao NVN, Rambabu C, 2012. Development of simple and sensitive visible spectrophotometric methods for the determination of Granisetron hydrochloride in pure and pharmaceutical formulations, Pharm tech 4, 1508-12.
7. Ismail Hewala, Mona Bedairb M, Sherif Shousha M, 2010. First derivative spectrophotometric determination of Granisetron hydrochloride in presence of its hydrolytic products and preservative and application to pharmaceutical preparations, Drug Test, Anal.
8. Ahid Zaheer, Sayad Imran, 2011.A simple and validated spectrophotometric determination of Granisetron in tablet dosage form. Inventi Impact: Pharmaceutical Analysis & Quality Assurance 64.
9. Ayengar Kanchana S, Ajithdas Aruna, Niraimathi V, Jerad Suresh A, 2010. Spectrophotometric estimation of Granisetron in bulk and tablet formulation, Int, J Chem, Sci 8, 1169-73.
10. Panzade Prabhkar, Puranik Prashant, Moghal Vipul, Patni Mayur, 2010. Development and validation of UV

spectrophotometric method of Granisetron hydrochloride in bulk and pharmaceutical formulation, Asian J Res Chem 3, 634-6.

11. Balapbalap Aishwarya R, Prasad Deepshikha V, Khidse Anuja S, Jadhav Shailaja B, Joshi SV, Chaudhari Praveen D, 2010. Development and validation of spectrophotometric methods for estimation of Granisetron hydrochloride in pure and its pharmaceutical dosage forms, Asian J Res Chem 3, 928-31.
12. Shirwaikar A, Prabu LS, Kumar DC, Kumar R, 2007. Spectrophotometric determination of Granisetron hydrochloride from bulk and its formulation, *Indian Drugs 44, 606-9.*
13. Venkata Rao SV, Ramu G, Ravi Kumar D, Rambabu C, 2012. A New Isocratic RP-HPLC method development for the assay of Granisetron HCl in API and dosage forms, Rasayan J Chem 5, 229-33.
14. Effat Souri, Zahra Kargar, Shahrooz Saremi, Nazanin Shabani Ravari, Farhad Alvandifara, Massoud Amanlou, 2011. Development and validation of a stability indicating HPLC method for determination of Granisetron, J Chin Chem Soc 58, 443-9.
15. Gopalakrishnan S, Jeyashree B, Thenmozhi M, Chenthilnathan A, 2011. Development and validation of new RP-HPLC method for the estimation of Granisetron hydrochloride in bulk and pharmaceutical dosage form, J Chem Pharm, Res 3, 470-6.
16. Lakshmana Babu S, Selvamani S, Latha S, 2010. HPTLC method for quantitative determination of Granisetron Hydrochloride in bulk drug and in tablets, Lat Am J Pharm, 29, 1455-8.
17. Yang Zhaoa, Hui-Jun Chena, Steve Caritisc, Raman Venkataramanan, 2016. Development and validation of a sensitive liquid chromatographic–tandem mass spectrometric method for the simultaneous analysis of Granisetron and 7-hydroxy Granisetron in human plasma and urine samples: application in a clinical pharmacokinetic study in pregnant subject, Biomed Chromatogr 30, 149-54.
18. Yunyun Jiang, Mei Lin, Guorong Fan, Yi Chen, Zhen Li, Weiquan Zhao, Yutian Wu, Jinhong Hu, 2006. Rapid determination of Granisetron in human plasma by liquid chromatography coupled to tandem mass spectrometry and its application to bioequivalence study, J Pharm Biomed Anal 42, 464-73.
19. Arayne MS, Najma Sultana, Zuberi MH, Siddiqui FA, 71 Spectrophotometric quantification of Metformin in bulk drug and pharmaceutical formulations using Multivariate technique, Indian J Pharm, Sci 71, 331-5.
20. Kokilambigai KS, Seetharaman R, Kavitha J, Sowndaravel P, Lakshmi KS, 2017. Multivariate Calibration technique for the spectrophotometric quantification of Zaleplon in bulk and pharmaceutical formulations, J Pharm, Sci, Res 9, 824-9.
21. SaiSusmitha A, Kokilambigai KS, Lakshmi KS, 2019. Spectrophotometric quantification of Telmisartan employing multivariate calibration technique in bulk and pharmaceutical formulations, Res J Pharm, Technol, 12, 1799-805.
22. Madhan S, Kavitha J, Lakshmi KS, 2016. Multivariate calibration technique for the spectrophotometric quantification of Ivermectin in Pharmaceutical Formulation, Asian J Pharm Clin, Res 12, 444-51.
23. Durgadevi P, Kokilambigai KS, Lakshmi KS, 2020. First order derivative spectrophotometric method for the quantification of Telmisartan employing Multivariate Calibration Technique, Res J Pharm, Technol 13, 774-80.
24. Naveena Rani D, Kokilambigai KS, Lakshmi KS, 2020. Multivariate calibration technique for the spectrophotometric quantification of Rasagiline in bulk drug and pharmaceutical formulation, Res J Pharm, Technol 13, 843-9.
25. Kanaka Parvathi Kannaih, Abimanyu Sugumaran, 2021. Eco-friendly multivariant green analytical technique for the estimation of Ketoconazole by UV spectroscopy in bulk and cream formulation, Quim. Nova 1-8.
26.

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