Journal of Applied Pharmaceutical Science Vol. 3 (01), pp. 095-098, January, 2013
Available online at http://www.japsonline.com
DOI: 10.7324/JAPS.2013.30118
ISSN 2231-3354

Analytical method development and validation of sitagliptine phosphate monohydrate in pure and tablet dosage form by derivative spectroscopy

G. Jeyabalan[1] and Narendra Nyola*[1, 2]
[1]Department of Pharmaceutical Analysis , Alwar Pharmacy College, Alwar , Rajasthan, India.
[2]Department of Pharmaceutical Sciences ,Shridhar University , Pilani ,Rajasthan, India.

ARTICLE INFO

Article history:
Received on: 15/10/2012
Revised on: 06/12/2012
Accepted on: 08/01/2013
Available online: 28/01/2013

Key words:
Spectrophotometric method, *Sitagliptine, Derivative spectroscopy*.

ABSTRACT

In this study zero, first and second order derivative spectrophotometric method were developed for the estimation of sitagliptine. In zero order spectrophotometery, absorbance value was measured at 267nm. In first derivative spectrophotometry amplitudes were measured at 213nm. In second derivative spectrophotometry amplitudes were measured at 276nm. Calibration curves were linear between the concentration range of 20-60µg/ml, 20-60µg/ml and 40-80µg/ml respectively. The % RSD value is less than 2% and the recovery were near 100% for all methods. This method has been validated for linearity, accuracy and precision and found to be rapid, precise, accurate and economical and can be applied for routine estimation of sitagliptine in solid dosage form. The validation of method was carried out utilizing ICH-guidelines.

INTRODUCTION

Sitagliptin phosphate monohydrate (SPM) chemically, (3*R*)-3-amino-1-[3-(trifluoromethyl)-5,6-dihydro[1,2,4]triazolo [4, 3- *a*] pyrazin -7 (8*H*) - yl] - 4 -(2,4,5 -trifluorophenyl) butan-1 one phosphate hydrate (Fig. 1) is oral hypoglycemic drug of the dipeptidyl peptidase-4(DPP-4) inhibitor class. DPP-4 inhibitors represent a new therapeutic approach to the treatment of type 2 diabetes that functions to stimulate glucose-dependent insulin release and reduce glucagons levels. This is done through inhibition of the inactivation of in cretins, particularly glucagon-like peptide- 1 (GLP-1) and gastric inhibitory polypeptide (GIP), thereby improving glycemic control (Herman *et al*., 2006, Dubal *et al*.,2012, Bala SC *et al.,* 2010) . Several analytical methods based on UV (Parag *et al*., 2011, Khan *et al* 2011), RP-HPLC (Ramzia *et al.,* 2011, Ravi *et al.,* 2011), LC-MS/MS (Zeng W *et al*.,2010 ,Wei Z *et al*.,2008, Nirogi *et al*.,2008)was reported for the determination of sitagliptin phosphate in plasma and urine of humans, rats and dogs.

* *Corresponding Author*
Email: - narennyola2@gmail.com

Selection of detection wavelength

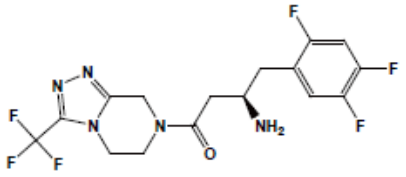

Fig 1 Structure of Sitagliptine

EXPERIMENTAL

Apparatus

A Shimadzu model 1800 double beam UV-Visible spectrophotometer with spectral width of 1 nm, wavelength accuracy of ± 0.1 nm and a pair of 10 mm matched quartz cell was used to measure absorbance of all the solutions. Spectra were automatically obtained by UV-Probe system software (Ver.2.34).

Reagents and Materials

All chemicals and reagents were used of AR grade. Authentic of SPM was obtained as gift samples from MSD Pharmaceutical private Ltd. Maharashtra, India.

Solutions of drug were scanned over the range of 200-400 nm. It was observed that the drug showed considerable absorbance at 267nm, 213nm and 276nm for zero, first and second order were selected as the wavelength for detection. The spectra of SPM show in fig. 2, 3 and 4.

Preparation of standard stock solutions

SPM was weighed (100 mg) and dissolved in 100 ml of methanol and make up the volume up to the mark with methanol, so the final concentration of solution containing 1000 µg/ml.

Preparation of working solutions

Aliquot from the stock solutions of SPM was appropriately diluted with methanol to obtain working standard.

Method Validation

The developed method was validated for its linearity, accuracy, precision and specificity.

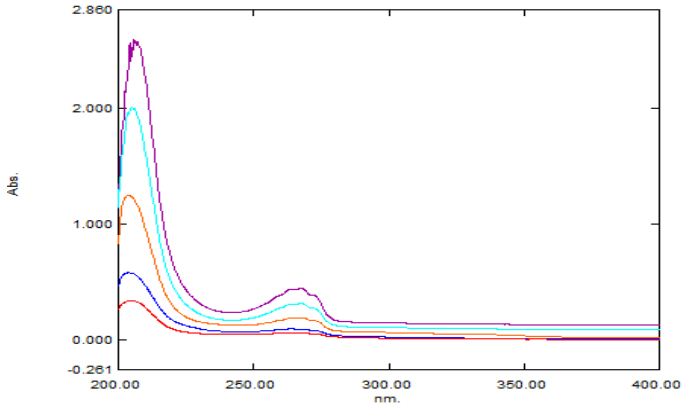

Fig 2 Zero order spectra of Sitagliptine.

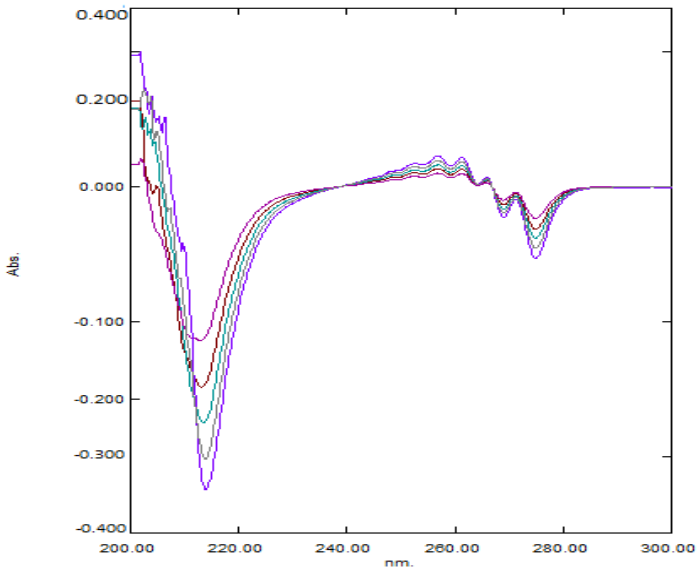

Fig 3 First order spectra of Sitagliptine.

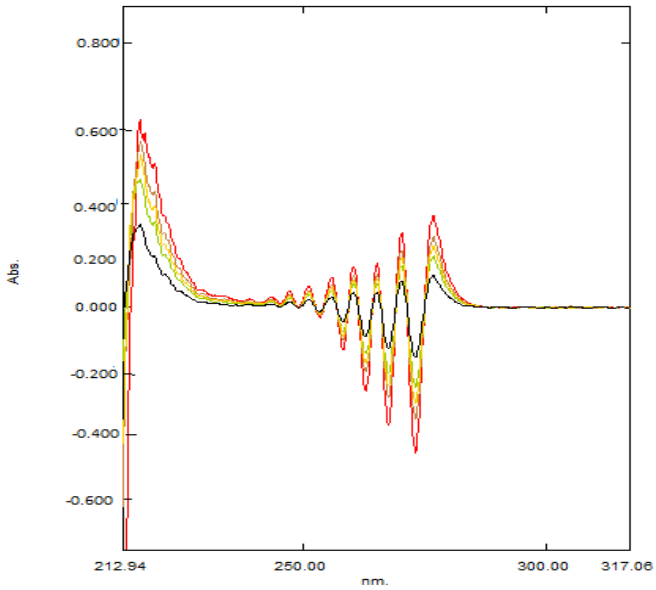

Fig 4 Second order spectra of Sitagliptine.

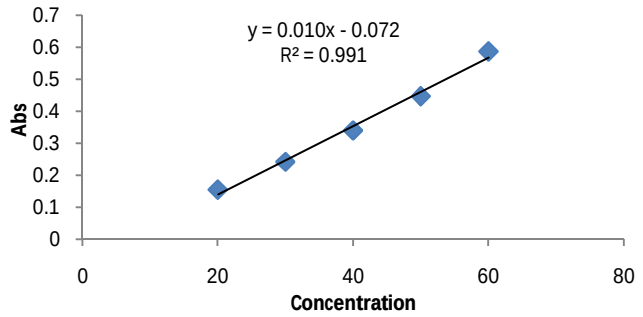

Fig 5 Zero order calibration curve.

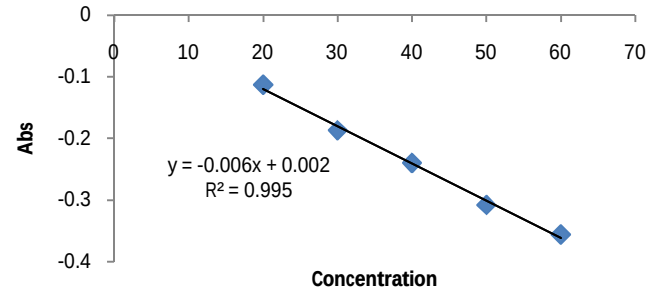

Fig 6 First order calibration curve.

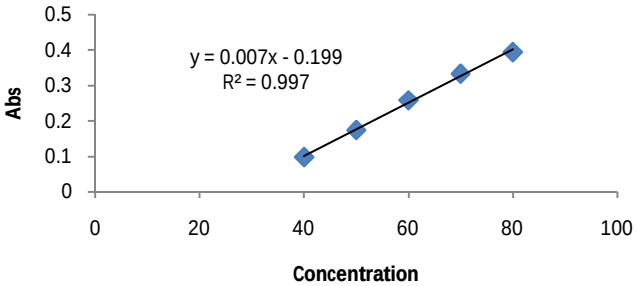

Fig 7 Second order calibration curve.

Linearity

The linearity of measurement was evaluated by analyzing different concentrations of the standard solution of SPM. The results are shown in table 1.

Table 1 System suitability parameters.

Parameters	Zero order	First order	Second order
Wavelength Maximum	267	213	276
Linearity range (µg/ml)	20-60	20-60	40-80
Correlation coefficient	0.991	0.995	0.997
LOD (µg/ml)	6.03	4.14	3.43
LOQ (µg/ml)	18.28	12.54	10.40
Sandell's sensitivity (mg/cm^2/0.001 absorbance unit)	0.117	0.167	0.268

LOD and LOQ

The LOD and LOQ were calculated from the equations, LOD =3.3 σ/S and LOQ = 10 σ/S, where σ is the standard deviation of the lowest standard concentration and S is the slope of the standard curve. The results are shown in table 1.

Precision

The precision of the proposed method was determined by analyzing different concentrations at different time intervals on same day (Intra-day precision) and on three different days (Inter-day precision). The results are shown in table 2.

Table 2 Precision.

Parameters	Zero order	First order	Second order
Intraday	0.32	0.55	0.597
Interday	0.74	0.52	0.600

Accuracy

To ascertain the accuracy of the proposed method, recovery studies were carried out by standard addition method. The results are shown in table 3.

Table 3 Recovery study.

Parameters	Zero order			First order			Second order		
Accuracy	80%	100%	120%	80%	100%	120%	80%	100%	120%
Amount present	20	20	20	10	10	10	40	40	40
Amount Added	16	20	24	8	10	12	32	40	48
Amount recovered	35.75	39.7	44.5	17.87	19.98	22.1	72.1	80.12	88.21
% Recovery	99.31	99.25	101.14	99.28	99.9	100.45	100.14	100.15	100.24

*Mean of three determinations in each level

Estimation of Sitagliptine from tablet.

Marketed preparation of SPM selected for the purpose of analysis. Twenty tablets were accurately weighed and powdered quantity equivalent to 100 mg of SPM was transferred in 100 ml volumetric flask and sonicated for 30 min. Then the volume was made upto the mark with methanol and the solution was filtered using Whatman filter paper no. 42 to obtain sample stock solution. 0.5 ml of filtrate was further diluted to 10 ml with same solvent and absorbance of sample was measured against blank. The amount of SPM was calculated from the calibration curve. The results of assay are shown in table 4.

Table 4: Assay of Dosage Form (Januvia).

Parameters	Zero order	First order	Second order
Label Claim (mg/tablet)	50	50	50
Amount Estimated (mg/tablet)*	49.6	50.2	49.75
Percentage Label Claim (%)	99.20	100.4	99.50

***Mean of three reading

Specificity

Interference and non-interference of excipients and binders was confirmed by performing the specificity study. Specificity was performed by spiking placebo with standard drug.

RESULTS AND DISCUSSION

As shown in fig. 2, 3, 4 SPM showed wavelength maxima at 267,213 and 276 nm in methanol. As shown in fig. 5, 6, 7 and table 1, the calibration curves are clearly indicates linearity of developed method. Results of intra-day and inter-day precision were expressed in % RSD and found to be within the allowable limit of ≤ 2%. It clearly indicates that the developed method is precise. The % Recoveries for SPM were found to be satisfied as shown in table 3;clearly indicate that the developed method is accurate. Assay result is in good agreements with the label claim. Hence, the proposed method can be successfully used for its analysis and quality control of marketed solid dosage preparation with good linearity, accuracy and precision.

CONCLUSION

From the above results it can be concluded that, the developed method is simple, rapid, accurate, precise, specific and economical. Hence, this method can be applied for quantitative analysis of Sitagliptine in bulk and pharmaceutical formulation like tablet dosage form.

ACKNOWLEDGMENTS

The authors are thankful to MSD Pharmaceutical private Ltd. Maharastra, India for providing gift sample of SPM. The authors are very thankful to Principal and Management of Alwar Pharmacy College for providing necessary facilities to carry out research work.

REFERENCE

Bala SC., Prameela RA. Development and validation of spectrophotometric method for the determination of DPP-4 Inhibitor, Sitagliptin, in its pharmaceutical preparations. Int. J. Pharm. Pharm. Sci.2010; 2(4): 138-142.

Dubal A., Khatwal R., Kosaraju J., Meda V., Samanta M.. Bioanalytical method development and validation of sitagliptin phosphate by RP-HPLC and its application to pharmacokinetic study. Int. J. Pharm Pharm Sci.2012; 4 (2): 691-694.

Herman G, Bergman A, Liu F, Stevens C, Wang A, Zeng W, Chen L, Snyder K, Hilliard D, Tanen M, Tanaka W, Meehan A, Lasseter K, Dilzer S, Blum R, Wagner J. Pharmacokinetics and pharmacodynamic effects of the oral DPP-4 inhibitor sitagliptin in middle-aged obese subjects. J.Clin.Pharmacol.2006; 46 (8): 876–886.

ICH Harmonised Tripartite Guideline. Text on Validation of Analytical Procedures, International Conference on Harmonization, Geneva, 1994; 1-5.

Khan G., Agrawal Y P, Sabarwal N., Jain A., Gupta A K. Simultaneous estimation of metformin and sitagliptin in tablet dosage form. Asian J. Biochem. Pharma. Res. 2011; 1(2): 352-358.

Nirogi R., Kandikere V., Mudigonda K., Komarneni P., Aleti R., Boggavarapu R. Sensitive liquid chromatography tandem mass spectrometry method for the quantification of sitagliptin, a DPP-4 inhibitor, in human plasma using liquid–liquid extraction. Biomed.Chromatogr.2008; 22(2): 214–222.

Parag P., Imran Md., Vinod B., Yogesh A. Development and validation of stability indicating UV spectrophotometric method for the estimation of sitagliptin phosphate in bulk and tablet dosage form. J. Pharm. Res. 2011; 4(3): 871-873.

Ramzia I. El-Bagary, Ehab F. Elkady, Bassam M. Ayoub. Liquid Chromatographic Methods for the Determination of Vildagliptin in the Presence of its Synthetic Intermediate and the Simultaneous Determination of Pioglitazone Hydrochloride and Metformin Hydrochloride. Int. J. Biomed. Sci.2011; 7(1): 62-69

Ravi P., Sastry BS., Rajendra PY., Appala RN. Simultaneous estimation of metformin HCl and sitagliptin phosphate in tablet dosage forms by RP-HPLC. Res. J. Pharm. Tech. 2011; 4(4): 646-649.

Wei Z., Donald MG., Alison FL., Li Chen, Michael SS, Eric WJ., Amy QW. Determination of sitagliptin in human urine and hemodialysate using turbulent flow online extraction and tandem mass spectrometry. J. Pharm. Biomed.Anal.2008; 46(3): 534-542.

Zeng W., Xu Y., Constanzer M., Woolf EJ. Determination of sitagliptinin human plasma using protein precipitation and tandem mass spectrometry. J.Chromatogr. B. 2010; 878(21):1817-1823.

How to cite this article:

G. Jeyabalan and Narendra Nyola. Analytical method development and validation of sitagliptine phosphate monohydrate in pure and tablet dosage form by derivative spectroscopy. J App Pharm Sci. 2013; 3 (01): 095-098.