International Journal of ChemTech Research
CODEN(USA): IJCRGG ISSN : 0974-4290
Vol.5, No.5, pp 2210-2215, July-Sept 2013

Development And Validation Of Spectrophotometric Method For Estimation Of Eperisone Hydrochloride In Bulk And Tablet Dosage Form By Using Area Under Curve Method.

Maske P.B[1]. and Nagras M.A.[1]*

**[1]Department of Pharmaceutical Chemistry,
STES'S Sinhgad College of Pharmacy, Vadgaon (Bk.), Pune-411041, India.**

***Corres.author: madhurinagras@yahoo.com,
*Contact No:-09822517546**

Abstract: A simple, accurate and precise UV spectrophotometric method has been developed for quantitative estimation of eperisone hydrochloride (EPE) in tablet and bulk dosage form by using area under curve method. The method involved measurement of area under curve in the wavelength range of 250 to 270 nm for EPE. Beer's law was obeyed in the concentration range of 2 to 20 µg/ml. The regression equation was y =0.149 x+0.006 with value of R^2 as 0.999. The method showed good linearity, accuracy and reproducibility. Accuracy was determined using standard addition method and mean % recovery was found to be 100%. Percent relative standard deviation values for intra-day and inter-day precision were found to be 0.12 and 0.18 respectively. The limit of detection and limit of quantitation were found to be 0.0286 and 0.095 respectively. Assay of EPE in tablet formulation was performed and percent purity of tablet was found to be 99.44%.
Key Words: Eperisone hydrochloride, Area under curve, UV spectrophotometry.

Introduction

Eperisone hydrochloride (EPE) is chemically (4'-ethyl-2-methyl-3-piperidino) propiophenone hydrochloride. (Fig.1), a piperidine derivative, centrally acting muscle relaxant which is used in the treatment of different pathological conditions like acute and chronic muscle spasm, electroconvulsive therapy, neurological conditions, orthopedic manipulation, myelopathy, encephalomyelitis, spondylosis, spondylarthrosis, cervical and lumbar syndrome, arthrosis of the large joints obliterating arthrosclerosis of the extremity vessels, diabetical angthromboangitis obliterans and Reynaud's syndrome[1]. EPE is official in Japanese pharmacopoeia[2].

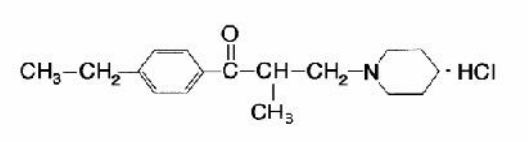

Fig.1. Chemical structure of EPE

Literature survey revealed that EPE is estimated by HPLC[3,4] and simple absorbance UV spectrophotometric method as single component[5,] as well as in combination with other drugs[1,6,7]. To our knowledge AUC method of UV spectrophotometry is not available for estimation of EPE in single component formulation. Hence, an attempt has been made to develop new UV method for its estimation in pharmaceutical formulations using double distilled water as a solvent system, with good accuracy, simplicity, precision and economy.

Materials and methods

Solubility studies

EPE is freely soluble in water, methanol, acetic acid and soluble in ethanol. Hence double distilled water was selected as solvent system.

Instrumentation

The instrument used was Shimadzu double beam UV/Vis spectrophotometer model V-1800. (UV Probe 2.32 software). Weighing was done on electronic single pan weighing balance (Make: Shimadzu Model: AX 200).

Materials

EPE was kindly gifted by Sharon Biomedicine, India Ltd.(Mumbai, India) and was used without any further purification. Tablet (MYSONE™) was purchased from local market, containing EPE 50 mg per tablet.

Preparation of standard stock solution

Standard stock solutions of EPE was prepared by dissolving 10 mg of drug in 10ml of double distilled water to get standard stock solution of 1000 µg/ml.

Preparation of calibration curve of EPE

The standard stock solution (1000 µg/ml) of EPE was further diluted to obtain the final concentration 2-20 µg/ml. This solution was scanned in the spectrum mode from 200.0 nm to 400.0 nm. The maximum absorbance of drug was observed at 260 nm (Fig.2). The calibration curve (Fig.3) was obtained by plotting concentration versus area. The amount of EPE was computed from the calibration curve.

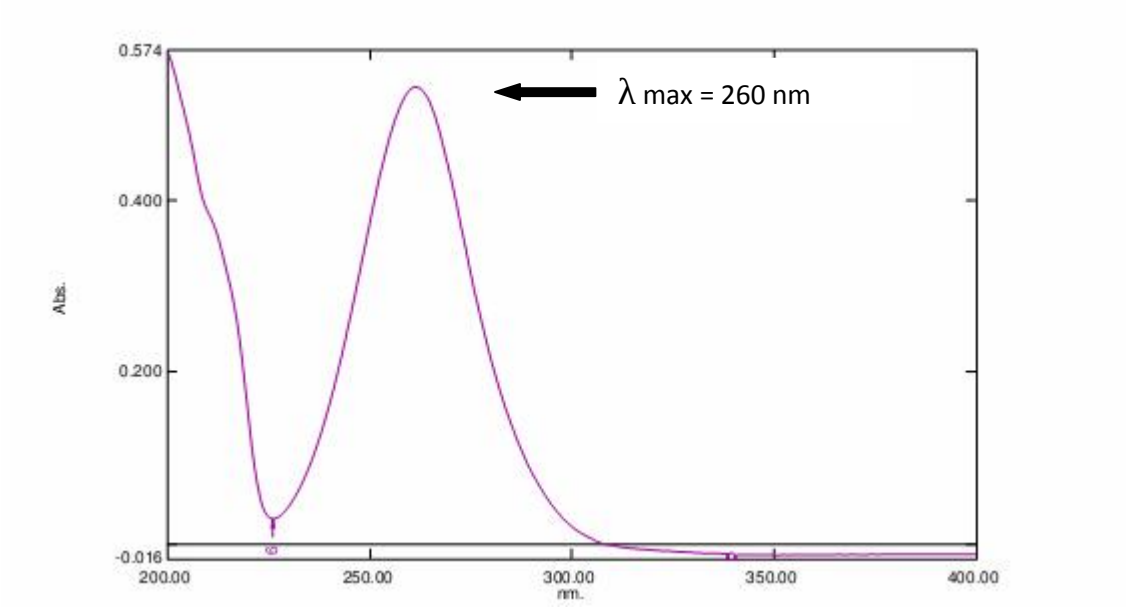

Fig.2. Absorption spectra of EPE.

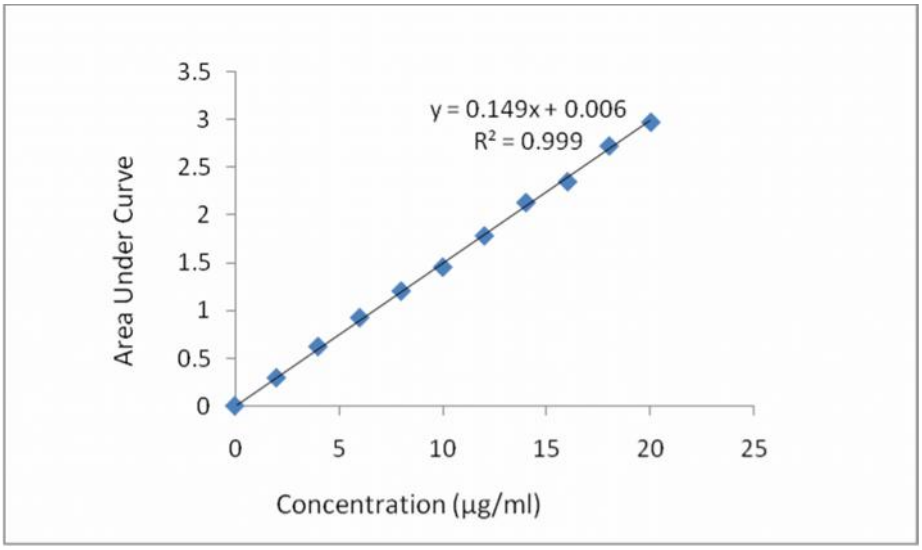

Fig.3. Calibration curve of EPE in double distilled water.

Experimental method

Area Under Curve (AUC) method involves the calculation of integrated value of absorbance with respect to the wavelength between two selected wavelengths, 250 - 270 nm for EPE. The area under curve between selected wavelength range was calculated by inbuilt software. (Fig.4)

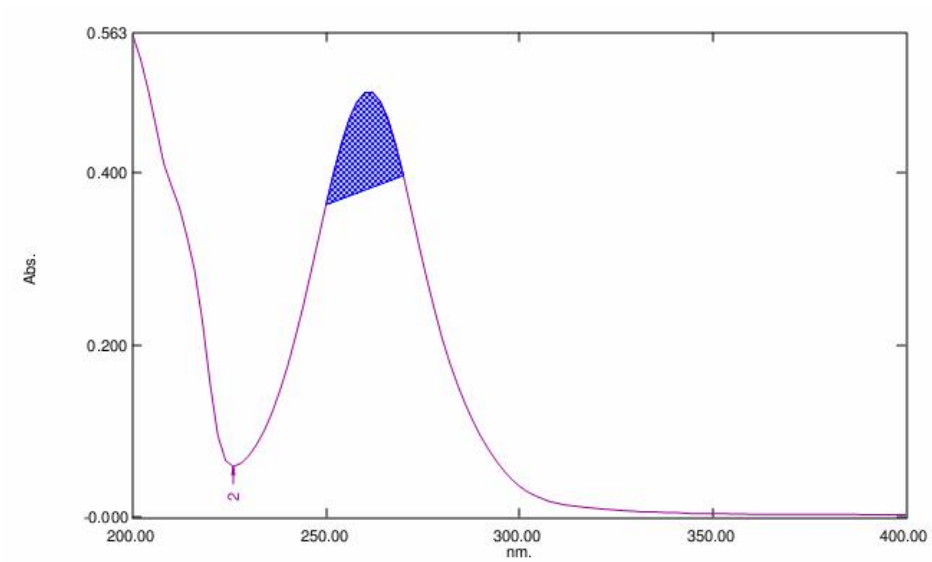

Fig.4. UV spectra of the EPE showing Area Under Curve.

Estimation of EPE from pharmaceutical formulation by area under curve method

The contents of twenty tablets were accurately weighed and crushed into fine powder. A quantity of powder equivalent to 50 mg of EPE was transferred to 100 ml volumetric flask containing 60 ml double distilled water, shaken manually for 20 min and the volume was made up to the mark with double distilled water and filtered through Whatmann filter paper (no.41). The solution was further diluted with double distilled water to give the concentration within the range of Beer's Law. Area under curve of this solution was measured in the range of 250 -270 nm and concentrations of the drug in the tablet formulation was calculated using area under curve method. (Table no. 2)

Results

Method validation

Validation of proposed method was done as per ICH guidelines[8] by means of the following parameters.

Linearity

As per ICH guidelines the linearity of an analytical procedure is its ability (within a given range) to obtain test results which are directly proportional to the concentration (amount) of analyte in the sample.[6] An appropriate volume of EPE in the range of 1-10 ml was transferred into series of separate 10 ml volumetric flasks and volume was made up to mark with double distilled water to get concentrations in the range of 2-20 µg/ml .

The linearity was evaluated by analyzing different concentrations of standard solution of EPE. The Beer's law was obeyed in the concentration range of 2-20 µg/ ml with regression coefficient of 0.999**.** (Table no 1.)

Precision

Intra and inter-day precision was performed by measuring the absorbance of standard solution at three different times during the single day and on three subsequent days respectively. The percent relative standard deviation (%RSD) was calculated. (Table no 1.)

Limit of Detection and Limit of Quantitation

LOD and LOQ were calculated by the data obtained from the linearity studies. The slope of the linearity plot was determined. For each of the six replicate determinations, y intercept was calculated and the standard deviation of the y intercept was computed. From these values, the parameters Limit of Detection (LOD) and Limit of Quantitation (LOQ) were determined by using formulae LOD= 3.3 * S.D. of y intercept/slope and LOQ= 10 * S.D of y intercept/slope. (Table no 1.)

Accuracy (Recovery studies)

Accuracy of the method was studied by recovery studies. The recovery studies were performed by applying the method to drug sample to which known amount of EPE corresponding to 80%, 100% and 120% of the label claim was added (standard addition method). The recovery was performed at three levels of the tablet and results were expressed as % RSD. (Table no 3.)

Table No. 1. Summary of validation parameters.

Parameters		**Observations**
λ max (nm)		260 nm
Linearity range (µg/ml)		2-20 µg/ml
Correlation coefficient (r^2)		0.999
Regression equation (y = mx+c)	Slope	0.149
	Intercept	0.006
Precision	Intraday (% RSD)	0.128
	Interday (% RSD)	0.182
LOD		0.028
LOQ		0.095

Table No. 2. Results of analysis of marketed formulation

Parameters	**Observations**
Label Claim (mg)	50
AUC of sample	1.449
AUC of standard	1.453
Drug content (%) ± SD	99.44 ± 0.24
% RSD	0.2495

Table 3. Results of recovery studies

	Sr. No.	AUC	Amount added	Amount recovered	% recovered	mean % recovery
80 %	1	1.203	8	8	100	
	2	1.205	8	7.98	99.83	
	3	1.208	8	8.01	100.16	100
100 %	1	1.453	10	10	100	
	2	1.451	10	9.98	99.86	
	3	1.455	10	10.01	100.13	100
120 %	1	1.782	12	12	100	
	2	1.785	12	12.02	100.16	100.38
	3	1.788	12	12.04	101	

Discussion

The standard solution of EPE when scanned in the UV range, using double distilled water, showed λ max at 260 nm hence 250-270 nm was selected as range for further studies. Linear relationships between drug concentrations were obtained over the range of 2-20 µg/ml. The regression equation was y =0.149 x+0.006 with value of R^2 as 0.999. Assay of EPE tablets was successfully performed and percentage purity of tablet was found to be 99.44%. No interference from tablet excipients was found. % RSD values for the intraday and inter-day precision were found to be 0.128% and 0.182 respectively. LOD and LOQ values were found to be 0.028 and 0.095 respectively. Results of the accuracy study indicated good recovery of the drug.

Conclusion

The results of our study indicate that the proposed UV spectroscopic method is simple, economic, precise and accurate. The developed UV spectroscopic method was found suitable for determination of EPE in bulk drug and in marketed solid dosage formulation without any interference from the excipients. It can therefore be concluded that use of this method can save much time and money and it can be used in laboratories with accuracy.

Acknowledgement

Authors are thankful to Sharon Bio-Medicine Ltd, Mumbai for providing gift sample of eperisone hydrochloride. We acknowledge infrastructure and facilities provided by the Sinhgad Technical Education Society and Sinhgad College of Pharmacy, Vadgaon (Bk), Pune.

References

1. Suthar J., Patel P. and Shah N., Development and validation of derivative spectrophotometric method for simultaneous estimation of eperisone hydrochloride and diclofenac sodium in combined capsule dosage form, Novus Int. J. Chem., 2012, 1.
2. Japanese Pharmacopoeia, 15th Edition, Shibuya, Tokyo (Japan): Society of Japanese Pharmacopoeia, 2006, 618.
3. Ding L., Wei X., Zhang S., Sheng J. and Zhang Y., Rapid and sensitive liquid chromatography–electrospray ionization-mass spectrometry method for the determination of eperisone in human plasma: method and clinical applications, J. Chrom. Sci., 2004, 42.
4. Ding L., Wang X., Yang Z. and Chen Y., The use of HPLC/MS, GC/MS, NMR, UV and IR to identify a degradation product of eperisone hydrochloride in the tablets, J. Pharm. Biomed. Anal., 2008, 46, 282–287.

5. Bhati S., Padaliya H., Prajapati H., Munjapara A. and Pancholi S., Development and validation of UV spectrophotometric methods for estimation of eperisone hydrochloride in bulk and pharmaceutical formulations by calibration curve method, Inventi rapid: Pharm Analysis & Quality Assurance, 2013.
6. Patel P. U, Patel S. K. and Patel U. J, Spectrophotometric method for simultaneous estimation of eperisone hydrochloride and diclofenac sodium in synthetic mixture, Int. Res. J. Pharm., 2012, 3.
7. Khanage S. G., Popat B. M. and Jadhav S. S., Development and validation of UV-visible spectrophotometric method for simultaneous determination of eperisone and paracetamol in solid dosage form, Adv. Pharm Bulletin., 2013.
8. International conference on harmonization (ICH), Q2A: Text on validation of analytical procedures: definitions and terminology, US FDA federal register,. 1995, 60.

*****