

## VALIDATED SPECTROPHOMETRIC METHOD FOR THE ESTIMATION OF PREGABALIN IN MARKETED FORMULATION

Rita Bamnela\*<sup>1</sup>, Sarita Nigam<sup>1</sup>, Varsha Chauhan<sup>1</sup>, Prabhat Jain<sup>2</sup>

1. Department of Chemistry, Sarojini Naidu Government Girls P.G. (Autonomous) College, Bhopal (M.P.)
2. Scan Research Laboratories, Bhopal (M.P.)

[ritabamnela@gmail.com](mailto:ritabamnela@gmail.com)

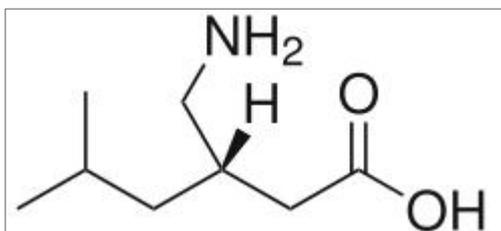
### Abstract

In present work a simple method was developed for the estimation of pregabalin in marketed formulation by spectrophotometer. Spectrophotometric methods are described for determination of pregebalin in bulk and pharmaceutical dosage forms using methyl orange as chromogenic agents. The methods were satisfactory applied for the determination of drugs in both bulk and pharmaceutical dosage forms.

**Keywords:** Pregabalin, Methyl orange, Spectrophotometric method.

### INTRODUCTION

Modern medicines for human use are required to comply with specific standards and regulation set forth by the concerned authorities. The efficacy and safety of medicinal products can only be assured by analytical monitoring of its quality. Pharmaceutical analysis is an art and science of determining the concentration of drug constituents present in marketed formulation.<sup>1</sup>Pregabalin (PRG; (3S)-3-(aminomethyl)-5-methylhexanoic acid; figure 1a) is a widely used Anticonvulsant and Analgesic for Epilepsy.<sup>2</sup>



**Figure 1: Chemical structure of pregabalin**

In this study pregabalin have been determined spectrophotometrically by using methyl orange to determine and estimate the identity, strength, quality and purity of drug.

### Materials and methods

#### Chemicals and reagents

All chemicals of analytical grade were used. Methyl orange dye solution: 2% dye solution was prepared by dissolving 2g of Methyl orange in 100ml of distilled water.

#### Apparatus

The present work was carried out on UV visible spectrophotometer. The absorption spectra of reference and test solution were carried out in a 1 cm quartz cell over the range of 200-800nm.

### **Preparation of calibration curve**

The absorption maxima of pregabalin were determined by running the spectrum of drug solution in double beam ultraviolet spectrophotometer. Accurately weighed 10 mg of Pregabalin separately and dissolved in 10 ml of 0.1N HCL in 10 ml of volumetric flask and prepared suitable dilution to make different concentration of standard with concentration range of 10-50 $\mu$ g/ml. 2 ml of standard drug solution and add 1 ml of methyl orange dye solution and 3 ml of chloroform, pipette out the coloured layer and analyzed for drug content by UV spectrophotometer at a  $\lambda_{max}$  of 416.0 nm using of 0.1 N HCl as blank.

### **Assay of tablet formulation**

Twenty tablets were taken and average weight of tablet was determined. The tablets were crushed in a mortar and the powder equivalent to 100mg of drug was transferred to 100ml standard flask.

The powder was dissolved in 50 ml of 0.1 N HCl and made up to volume with of 0.1 N HCL. The sample was mixed thoroughly and filtered through a 0.45 $\mu$  membrane filter. The filtered solution was diluted suitably and add 1 ml of methyl orange dye solution and 3 ml of chloroform pipette out the coloured layer and analyzed for drug content by UV spectrophotometer at a  $\lambda_{max}$  of 416.0 nm using of 0.1 N HCl as blank.

### **Validation of developed method**

The developed methods were validated according to international conference on harmonization guidelines.<sup>3</sup>

### **Linearity**

Linearity of both drugs was established by response ratios of drugs. Response ratio of drug calculated by dividing the absorbance with respective concentration. Then a graph was plotted between concentration and response ratio.

### **Accuracy**

The accuracy of the proposed methods was assessed by recovery studies at three different levels i.e. 80%, 100%, 120%. The recovery studies were carried out by adding known amount of standard solution of pregabalin to pre analysed tablet solutions. The resulting solutions were then reanalyzed by proposed methods. Whole analysis procedure was repeated to find out the recovery of the added drug sample. This recovery analysis was repeated at 3 replicate of 5 concentrations levels.

### **Precision**

Precision of the methods was studied at three level as at repeatability, intermediate precision (Day to Day and analyst to analyst) and reproducibility. Repeatability was performed by analyzing same concentration of drugs for five times. Day to Day was performed by analyzing 5 different concentration of the drug for three days in a week.

### **Results and discussion**

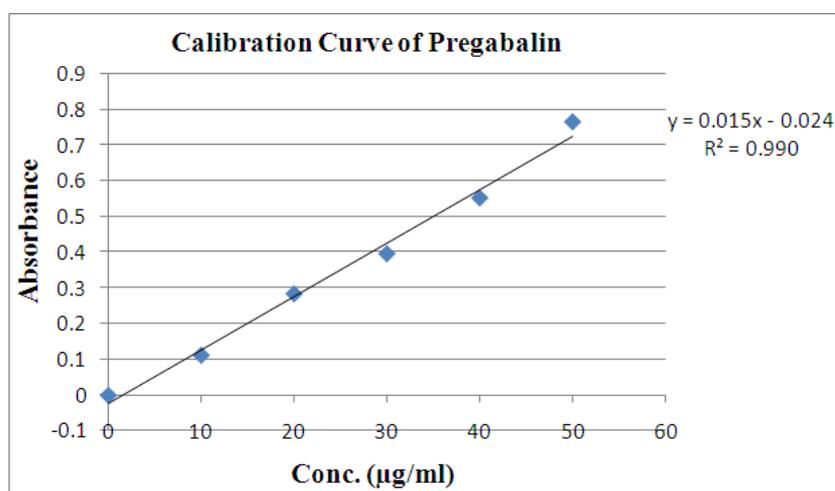
The proposed spectrophotometric methods are indirect and based on the determination of the pregabalin in marketed tablet formulation using methyl orange as chromogenic agents.

**Table 1: Readings for calibration curve of pregabalin**

Replicate	10	20	30	40	50
1	0.121	0.258	0.388	0.535	0.756
2	0.110	0.282	0.394	0.551	0.736
3	0.114	0.268	0.396	0.551	0.766
Mean	0.112	0.284	0.396	0.552	0.765
S.D.	0.002	0.002	0.002	0.001	0.002
% RSD	1.360	0.536	0.500	0.208	0.260

**Table 2: Statistical data for linearity**

S. No.	Parameter	Remark
1	Linearity range	10-50 $\mu\text{g/ml}$
2	Regression equation	$0.015x+0.023$
3	Correlation coefficient	0.990

**Figure 2: Calibration curve of pregabalin at 416nm****Table 3: Results of recovery studies on marketed formulations**

Recovery level %	% Recovery (Mean $\pm$ SD)*
80	99.12 $\pm$ 0.146
100	99.22 $\pm$ 0.282
120	99.12 $\pm$ 0.144

\*Average of five determination

**Table 4: Results of validation (% R.S.D.)**

	<b>Parameters</b>	<b>Results</b>
Precision (% R.S.D.)	Repeatability	0.046
	Day to Day	0.052
	Analyst to Analyst	0.048
	Reproducibility	0.088

\*Average of five determination

Calibration curves have correlation coefficients (r) 0.990 indicating good linearity. The accuracy of the methods were determined by investigating the recovery of drugs at concentration levels covering the specified range (five replicates of each concentration).

Some Pharmaceutical formulations containing stated drugs have been successfully analyzed by the proposed methods. Results obtained were compared to those obtained by applying reported reference methods<sup>4-7</sup> which in turn indicate that there is no significant difference between proposed methods and reference ones relative to accuracy and precision.

### **Conclusion**

The proposed spectrophotometric methods were accurate, precise and reliable for the Measurement of pregabalin in dosage form. The developed spectrophotometric method was validated for estimation of pregabalin using linearity, range, accuracy and precision. The RSD for all parameters was found to be less than one, which indicates the validity of method and assay results obtained by this method are in fair agreement. The developed method can be used for routine quantitative estimation of pregabalin in pharmaceutical preparation. The present methods are superior to the reference method with respect to both sensitivity and selectivity. The methods can be successfully applied for the analysis of marketed tablets and capsules.

### **References**

1. Mehdhem J, Denney RC, Barnes JD and Thomsa MJK. Chemical analysis. Vogel's Text Book of quantitative Chemical Analysis. Pearson Education Limited; Delhi, Sixth Ed. 2002; 1-11.
2. Saravanan.J, Shajan A, Joshi NH, Varatharajan R and Valliappan K. RP- HPLC method for the estimation of methylcobalamin in bulk and capsule dosage form. International journal of chemical and pharmaceutical sciences. 2010; 1: 13-16.
3. Guidance for Industry: Q2B of Analytical Procedures; Methodology: International Conference on Harmonization (ICH).Nov. 1996.
4. Onal A and Sagirli O. Spectrophotometric, spectrofluorimetric methods for the determination of pregabalin in bulk and pharmaceutical preparation. Spectrochim Acta A. 2009; 1:68-71.
5. Vermeij TA and Edelbroek PM. Simultaneous high-performance liquid chromatographic analysis of pregabalin, gabapentin and vigabatrin in human serum by precolumn derivatization with o-phthalaldehyde and fluorescence detection. J Chromatogr B Analyt Technol Biomed Life Sci. 2004; 810:297-303.

6. Berry D and Millington C. Analysis of pregabalin at therapeutic concentrations in human plasma/serum by reversed-phase HPLC. *Ther Drug Monit.* 2005;27:451-6.
7. Michael JL, Timothy RH, Brian T, Stephen RP. Synthesis and characterization of pregabalin lactose conjugate degradation products. *J Pharm Biomed Anal.* 2002; 28:917-24.