WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 7.523
Volume 6, Issue 8, 1354-1360. Research Article ISSN 2277– 7105

RP-HPLC METHOD DEVELOPMENT AND VALIDATION FOR PREGABALIN AND CELECOXIB IN BULK AND DOSAGE FORMS

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Article Received on
30 May 2017,

Revised on 20 June 2017,
Accepted on 10 July 2017

DOI: 10.20959/wjpr20178-8990

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ABSTRACT

A simple, accurate, precise and highly selective reverse phase high performance liquid chromatographic (RP-HPLC) method was developed and validated for Pregabalin and Celecoxib. Chromatographic separation was achieved isocratically by using waters allaiance 2695 separation module, Hypersil BDS(150 mm x 4.6 mm, 5µ) at temperature 30°c. Flow rate selected was 1ml/min. Both changes were identified with 238 nm. Mobile phase employed was potassium di hydrogen orthophosphate buffer of pH 6.5 and acetonitrile in the ratio of (70:30) which resulted best resolution and sensitivity. Developed method was validated in terms of linearity, range(37.5 µg/ml-281.25 µg/ml, for Pregabalin, 100 µg/ml -750 µg/ml Celecoxib), precession (correlation coefficient is less than 0.999), robustness, accuracy (recovery of Pregabalin and Celecoxib were 100.3% and 100.13% respectively). The validation of proposed method was verified by recovery studies and can be applicable in routine pharmaceutical analysis.

KEYWORDS: RP-HPLC, Pregabalin, Celecoxib and potassium di hydrogen orthophosphate buffer.

INTRODUCTION

Pregabalin(PRE) and Celcoxib (CEL) are used for the treatment of diseases like neuropathic pain. Pregabalin is chemically (*S*)-3-(aminomethyl)-5-methylhexanoic acid, act as Anticonvulsant, Analgesic, Celecoxib is chemically 4-[5-(4-methylphenyl)-3-(trifluoromethyl)-1H-pyrazol-1-yl]benzene-1- sulphonamide belonging to category

cyclooxigenage inhibitors. It is needed to develop a method without any draw back because no methods are reported for Pregabalin and Celecoxib.

MATERIALSAND METHOD

Chromatographic separation was carried by using WATERS Aliance 2695 model with empower2 software, Weighing Balance model no ER200A, Sonicator with SE60US and pH Meter AD102U model was used . Pregabalin and Celecoxib standards are obtained as gift from AUROBINDO labs, Hyderabad, the tablet dosage forms as Lyrica 75 (Pregabalin) and TAPAL 50 (Tapentdol). All the chemicals and reagents user were HPLC grade or analytical reagent grade purchased from qualigens, Merck (CHEMICALS), Mumbai, India.

CHROMATOGRAPHIC CONDITIONS

The separation of drugs were performed by using potassium dihydrogen orthophosphate buffer of pH 6.5: Aceto nitrile (70:30) as mobile phase and Hypersil BDS was used as stationary phase. As a result of this the Peak resolution between two peaks was very good, retention time also good and no impuritiy peaks are developed at 238nm.

PREPARATION OF STANDARD SOLUTION

Accurately Weighed and transferred 75mg of Pregablin and 200mg of celecoxib working Standards into two 10 ml clean dry volumetric flasks, add 7ml of diluent, sonicated for 5 minutes and make up to the final volume with diluents.(standard stock).

CALIBRATION CUVRE

Calibration curve was developed to determine the linearity. The plots were developed concentrations verses area of the peak, it must obey Beer's- Lambert's law. The linearity was determined by using serial dilution of 37.5 µg/ml of Pregabalin and 100 µg/ml of celecoxib, (93.75 µg/ml of Pregabalin and 250 µg/ml of celecoxib, 131.25µg/ml of Pregabalin and 350µg/ml of celecoxib, 187.5 µg/ml of Pregabalin and 500 µg/ml of celecoxib, 225 µg/ml of Pregabalin and 600 µg/ml of celecoxib and 281.25 µg/ml of Pregabalin and 750 µg/ml of celecoxib.

ANALYSIS OF FORMULATION

20 tablets were taken individually and triturated to fine powder and take equivalent to 75mg of Pregablin and 200mg of Celecoxib in to a 10 ml clean dry volumetric flasks, add 7ml of diluent, sonicated for 5 minutes and make up to the final volume with diluent.

RESULTS AND DISCUSSION

In the method the both drugs are eluted with a run time of 10min using potassium di hydrogen orthophosphate buffer of pH 6.5 and acetonitrile in the ratio of 70:30 as mobile phase, the retention times for Pregabalin and Celecoxib were 2.527 min and 6.516 min respectively at the flow rate of 1ml/min. the wavelength selected for determination was 238nm for both drugs. The tailing factor, resolution, theoretical plate count were reported in table.

The linearity of PRE and CEL were determined by calibration curves and linearity table the range of 37.5 µg/ml-281.25 µg/ml and 100 µg/ml- 750 µg/ml respectively, the regression coefficient for PRE and CEL were 0.999 and 0.999 respectively. Precision was measured for intraday and checked for repeatability and %RSD for repeatability 0.08 and 0.4 for PRE and CEL respectively. The RSD was found to be within the limit and the results were tabulated.

The accuracy was determined by studying the percentage recovery. the recovery studies are performed for 505, 100% and 150% solution of PRE and CEL mixture. The results are tabulated below.

The robustness was determined by changing the parameters like temperature and flow rate and the %RSD was calculated by taking the results for 3 injections. The results were mentioned in the table.

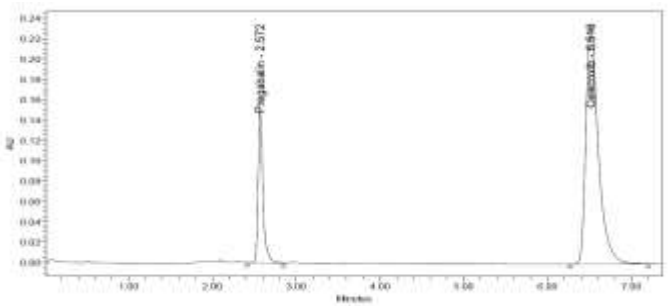


Table: 1 SYSTEM SUITABILITY PARAMETERS

PARAMETER	PRE	CEL
RETENTION TIME	2.572	6.516
TAILING FACTOR	1.44	1.56
THEORETICAL PLATE COUNT	10751	10224

TABLE 2. RECOVERY STUDIES

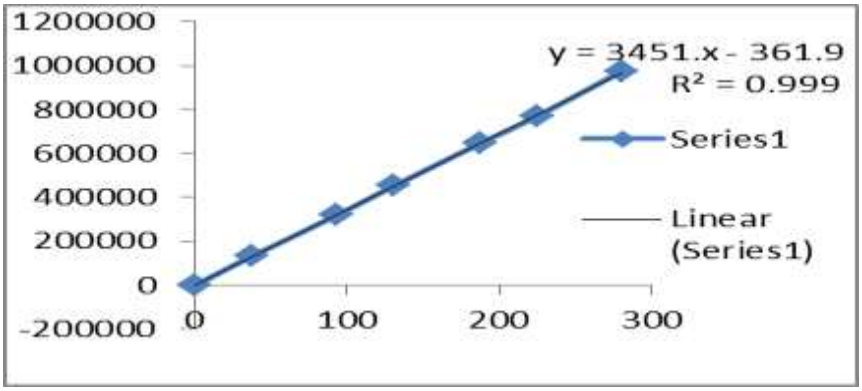
DRUG	AMOUNT ADDED		AMOUNT FOUND		% RECOVERY	
	PRE	CEL	PRE	CEL	PRE	CEL
	375	250	378.46	248.9408	100.9%	99.5%
50%	375	250	377.70	247.34	100.92%	98.9%
	375	250	378.47	248.14	100.92%	99.25%
	750	500	752.36	496.60	100.31%	99.3%
100%	750	500	750.96	496.30	100.12%	99.2%
	750	500	751.57	497.01	100.21%	99.3%
	1125	750	1130.20	756.41	100.46%	100.85%
150%	1125	750	1128.91	756.44	100.34%	100.85%
	1125	750	1130.13	756.44	100.45%	100.85%

TABLE 3 INTRADEY PRECISION

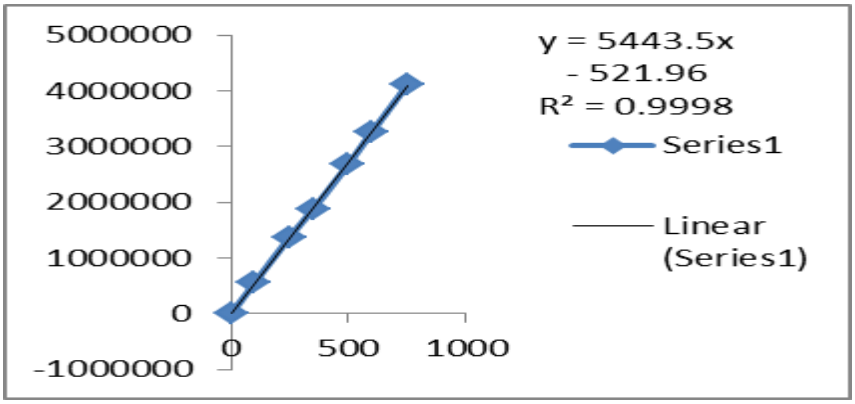
Injection	**Area of Pregabalin**	**Area of Celecoxib**
Injection-1	296551	6148603
Injection-2	299707	6153129
Injection-3	295025	6154783
Injection-4	293010	6127838
Injection-5	298788	6122394
Injection-6	297910	6148809
Average	296831.8	6142593
Standard Deviation	2498.221	13856.92
%RSD	**0.8**	**0.2**

TABLE 4 LINEARITY

Level	**Cocentration (ug/ml) pregabalin**	**Cocentration (ug/ml) celecoxib**	**Response Pregabalin**	**Response celecoxib**
I	37.5	125	132397	555046
II	93.75	250	320251	1378399
III	131.25	375	453398	1883364
IV	187.5	500	645610	2688038
V	225	625	769320	3266534
VI	281.25	750	976614	4105967



Linearity Pregabalin



Linearity Celecoxib

CONCLUSION

The proposed HPLC method was found to be simple, specific, precise, accurate, rapid and economical for simultaneous estimation of Pregabalin and celecoxib in bulk and pharmaceutical dosage form.

The method is completely validated and satisfactory results were obtained for all parameters tested.

ACKNOWLEDGEMENT

This work was partially supported by Sri Balaji College of pharmacy, Pulla Reddy Institute of Pharmacy and Gland Institute Of Pharmaceutical Sciences, Narsapur.

REFERNCES

1. Gurudeep chatwal and sham anand, Instrumental methods of chemical analysis Analysis. Himalaya publishers, 7th edition, 1992; Pg. no 2.624-2.639.
2. HPLC for Pharmaceutical Scientists by Yuri Kazakevich 1st edition 2007; 3-6.

3. Vogel's Text book of quantitative chemical analysis. Published by Dorling Kindersley pvt.ltd. 6th edition, 2008; 289-304.
4. Lloyd R. Snyder et al., practical HPLC method development. John wiley & son's publishers, 2nd edition, 1997; 350-400.
5. Kannapan N et al.., Analytical RP-HPLC Method for Development and Validation of Pregabalin and Methylcobalamine in Combined Capsule Formulation: Journal of Applied Chemical Research, ISSN: 2008-3815, 13, 85-89 (2010).
6. Sarvesh Kumar Mishra et al.., Stability Indicating Rp-Hplc Method For Determination Of Pregabalin Using Ich Guidelines: International Journal Of Natural Product Science 2012; Spl Issue 1: 130.
7. G. B. Kasawar et al.., Development and Validation of HPLC Method for the Determination of Pregabalin in Capsules: Indian J Pharm Sci. 2010 Jul-Aug; 72(4): 517–519.
8. Ashu M et al.., Development and validation of rapid HPLC method for determination of Pregabalin in bulk drug and capsule dosage forms: Der Pharma Chemica, 2011; 3(1): 482-489.
9. Naresh Chandra Reddy M et al., RP-HPLC Determination of Related substances of Pregabalin in bulk and pharmaceutical dosage form: International Journal of Chemical and Pharmaceutical Sciences ISSN: 0976-9390, 2012; June., 3(2): 40-46.
10. P.Nalla Kumar et al., RP – HPLC method development and validation for the simultaneous estimation of methyl cobalamine and pregabalin in capsules.
11. Krishnaveni Get al., A Novel RP-HPLC method for the Quantification of Celecoxib in Formulations: Research Journal of Pharmaceutical, Biological and Chemical Sciences, January – March 2012; 3(1): 340-346.
12. KG. Jadhav et al.., A Validated RP-HPLC Method for the Determination of Celecoxib in Bulk and Pharmaceutical Dosage Form: International Journal of Research in Pharmaceutical and Biomedical Sciences ISSN: 2229-3701 Jul – Sep2012; 3(3): 1312-1316.
13. Pramod M. Dhabu et al., A Stability-Indicating HPLC Method to Determine Celecoxib in Capsule Formulations: informa 2002; 28(7): 815-821.
14. Jayasagar G Validated HPLC method for the determination of celecoxib in human serum and its application in a clinical pharmacokinetic study: Pharmazie. 2002 Sep; 57(9): 619-21.

15. Patel Prakash et al., Development And Validation Of Rp-Hplc Method For The Simultaneous Estimation Of Diacerein And Celecoxib In Pharmaceutical Dosage Form: Inventi Rapid: Pharm Analysis & Quality Assurance Vol. 2013 publication date: 2013/6/4.
16. Rao RN et al., Development and validation of a reversed-phase liquid chromatographic method for separation and simultaneous determination of COX-2 inhibitors in pharmaceuticals and its application to biological fluids: Biomed Chromatogr. 2005 Jun; 19(5): 362-8.
17. www.clinical trails.gov