ISSN-1996-918X

Pak. J. Anal. Environ. Chem. Vol. 14, No. 2 (2013) 38 – 43

Determination of Tapentadol Hydrochloride in Tablets by Three New Validated Spectrophotometric Methods

Deepti Jain[1] and Pawan Kumar Basniwal*[1,2]

[1]*School of Pharmaceutical Sciences, Rajiv Gandhi Technological University, Bhopal – 462 033, Madhya Pradesh, India*

[2]*LBS College of Pharmacy, Jaipur – 302 004, Rajasthan, India*

Received 25 June 2013, Revised 17 December 2013, Accepted 23 December 2013

Abstract

Three UV spectrophotometric methods viz. linear regression equation (LRE), standard absorptivity (SA) and first order derivative (FOD) method were developed and validated according to ICH guideline. The methods were linear in the range of 10-50 µg/mL and validated methods were successfully applied for determination of tapentadol hydrochloride content in tablet dosage forms in the range of 100.42-100.64%, 100.31-100.54% and 100.10-100.59%, respectively. The standard deviation for all the methods was found to be less than one unit (0.45-0.83). The validated spectrophotometric methods may be successfully applied for assay, dissolution studies, bio-equivalence studies as well as routine analysis in pharmaceutical industries.

***Keywords*:** Tapentadol; Spectrophotometric methods; Tablets

Introduction

Tapentadol (Fig. 1), chemically, 3-[(1R, 2R)-3-(dimethylamino)-1-ethyl-2-methylpropyl] phenol hydrochloride which is used a novel analgesic agent. The analgesia effect of drug is result of two mechanisms of action vis. agonistic activity at µ opioid receptor and inhibition of norepinephrine reuptake. Both immediate release and extended release formulations of tapentadol are available in market. It provides analgesia in both states of pain (acute and chronic) similar to morphine but improved gastrointestinal tolerability (nausea, vomiting, and constipation) [1-2].

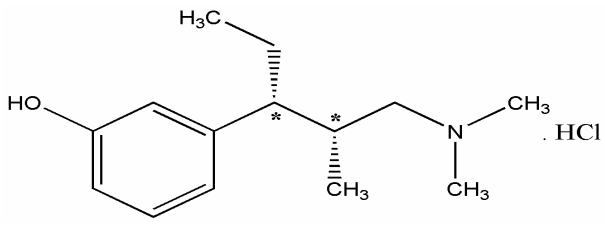

Figure 1. **Structure of tapentadol hydrochloride**

Although tapentadol was determined in different biological matrix viz. in canine plasma by HPLC with spectrofluorimetric detection [3] along with metabolite in urine [4] and in urine and oral fluid [5] by liquid chromatography-tandem mass spectrometry. The drug was also assayed in a pharmacokinetics study [6], in combined dosage form with paracetamol [7], in laboratory samples by chromatographic [8], in tablets by RP-HPLC [9] and spectrophotometric methods [8, 10].

The aim of present work was to develop spectrophotometric methods and apply for determination of drug content in dosage form. The spectrophotometric methods are better applied for routine analysis as these are: economic (solvents and instruments costs are key factor), rapid (UV-analysis time is less), simple (not required too much training to operate), maintenance free (not washing and no special care for UV), accurate and precise. Keeping the view of quality assurance, the

*Corresponding Author Email: pawanbasniwal@gmail.com

present research work was aimed to develop and validate spectrophotometric methods for determination of tapentadol in bulk drug as well as in dosage form to ensure the quality and purity of the drug.

Experimental

Instruments and chemicals

Ultraviolet spectrophotometer (Shimadzu 1800 series) with 1 cm matched quartz cells was used for the designed research work. Shimadzu-Ax-200 electronic balance was used for weighing the samples, and Rivera make volumetric glasswares were used. Tapentadol hydrochloride WS (working standard) was procured from Ranbaxy Laboratories Ltd. Gurgaun, New Delhi, as a gift sample. Tapentadol tablets (TRANSDOL 50, Lupin Ltd. Mumbai, India) were purchased from local market. Distilled water was prepared by distillation assembly.

Linear regression equation (LRE) method

Tapentadol hydrochloride (50 mg) was weighed and dissolved in 50 ml of distilled water to prepare stock A (1000 µg/mL). Aliquots of the stock A was diluted to get concentration of 10, 20, 30, 40 and 50 µg/mL. The dilutions were scanned against water as blank in the range of 200-400 nm to get Gaussian spectra (Fig. 2). The absorbance of the dilutions was recorded at 272 nm. The calibration graph was plotted concentration vs. absorbance and regression equation was determined with correlation coefficient.

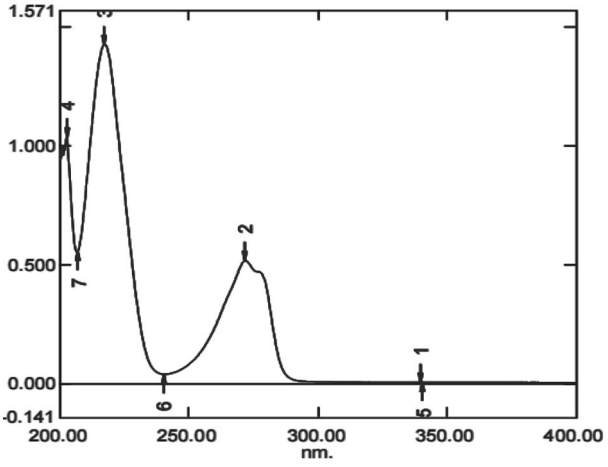

Figure 2. **Gaussian spectrum of tapentadol HCl in water**

Standard absorptivity (SA) method

Five dilutions of drug (10, 20, 30, 40, and 50 µg/mL) were prepared in triplicates and the absorbances were observed at 272 nm against distilled water as blank. From above absorbances, the standard absorptivity A (1%, 1cm) and molar extinction coefficient ε were calculated, which would be used to determine the drug content of dosage forms.

First order derivative (FOD) method

The first order derivative spectrophotometric method was developed, as interference of one analyte in absorbance of another analyte may be nullified in the derivative mode. Dilutions (10, 20, 30, 40 and 50 µg/mL) of tapentadol hydrochloride were scanned to get Gaussian spectra and these spectra were converted into first order derivative mode (Fig. 3). The absorbance was observed at 221 nm in the derivative mode and linear regression equation was calculated.

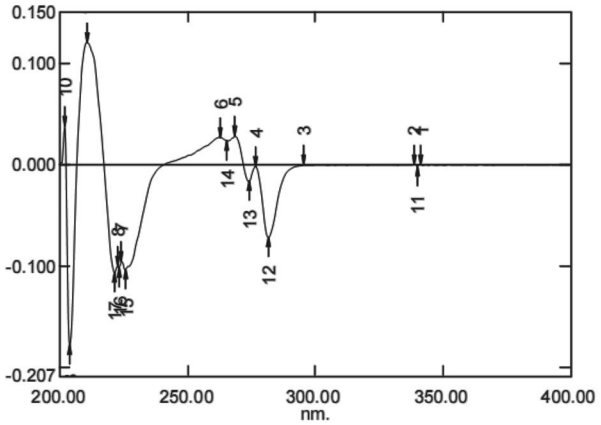

Figure 3. **First order derivative of Gaussian spectrum of tapentadol HCl in water**

Validation of methods

As per ICH guidelines [11] dilutions of the drug in triplicate were used to validate all three methods for linearity, accuracy (by recovery studies, standard addition to pre-analysed samples), repeatability (within day), intermediate precision (days and analyst variation) and robustness (temperature variation: 35°C, 25 °C and 15 °C), and statistical parameters were calculated for them.

Analysis of dosage form

Twenty tapentadol hydrochloride tablets (TRANSDOL 50, Lupin Ltd. Mumbai, India) were finely powdered; a quantity equivalent to 50 mg of tapentadol hydrochloride was dissolved in 100 ml of distilled water and filtered through Whatman filter paper no. 41 to give stock I. Aliquots of stock I were diluted to obtain sample concentrations (20, 30 & 40 µg/mL) in the range of linearity. The absorbance values of these sample dilutions were observed in a multipoint calibration curve of quantitative mode at the selected wavelength to obtain test sample concentration.

Results and Discussion

Linear regression equation (LRE) method

Although tapentadol hydrochloride was soluble in water and methanol but it is freely soluble in distilled water which is cheapest solvent for spectrophotometry; so it was decided to develop and validate spectrophotometric methods for determination of tapentadol hydrochloride using water as solvent. The drug was stable in the aqueous solution as there was no deviation from Gaussian spectrum after one week storage of the stock solutions. The six replicates of all dilutions were processed for the linear regression method and the linear regression equation was found to be $Y = 0.286 x - 0.005$ with correlation coefficient $R^2 = 0.9990$ (Table 1).

Table 1. **Calibration graph of tapentadol hydrochloride in water.**

Conc.	Absorbance at 272 nm					
(µg/mL)	I	II	III	IV	V	VI
10	0.282	0.287	0.286	0.281	0.282	0.279
20	0.572	0.565	0.571	0.574	0.569	0.568
30	0.852	0.853	0.859	0.854	0.857	0.855
40	1.135	1.136	1.134	1.137	1.134	1.132
50	1.436	1.433	1.434	1.432	1.441	1.433
Regression Equation* $Y = 0.286 x - 0.005$; $R^2 = 0.9990$						

* mean of above six replicates

Standard absorptivity (SA) method

Standard absorptivity avoids the need to prepare a standard solution of the reference substance in order to determine its absorptivity. It is an advantage in situations where it is difficult or expensive to obtain a sample of the reference substance [12]. Hence, three replicates of five serial dilutions were used to determine standard absorptivity [A (1%, 1cm)] and molar extinction coefficient (ε), which were found to be 285.90 dl/g/cm and 7370.62 per Mol/cm, respectively (Table 2). Now, we can easily determine the concentration of the any sample without preparing calibration graph with help of the above formula of the standard absorptivity. Thus, it provides single step determination of the tapentadol hydrochloride; measure the absorbance of sample of the drug solution and determine the concentration.

Table 2. **Standard absorptivity A (1%, 1cm) and molar extinction coefficient (ε).**

Conc.	Absorbance at 272 nm			Standard Absorptivity [A (1%, 1cm) = A/bc]		
(µg/mL)	I	II	III	I	II	III
10	0.288	0.291	0.283	288.00	291.00	283.00
20	0.571	0.569	0.574	285.50	284.50	287.00
30	0.855	0.851	0.858	285.00	283.67	286.00
40	1.136	1.141	1.133	284.00	285.25	283.25
50	1.431	1.442	1.439	286.20	288.40	287.80
A (1%, 1 cm)* = 285.90 dl/g/cm; ε ** = 7370.62 per Mol/cm						

* Mean of 15 above standard absorptivities determination
**Molar extinction coefficient ε = A (1%, 1cm) x Molecular weight/10.

First order derivative (FOD) method

Nowadays, FOD technique becomes very useful in spectrophotometry, additional tool which resolve various analytical problems where chances of interference in absorbance by other analyte. It has been applied in number of analytical fields, especially in pharmaceutical, clinical and biochemical analysis. First order derivative of Gaussian spectrum of tapentadol hydrochloride was successfully applied to determine the drug content (Table 3). Zero crossing was found at 242 nm and 272 nm in this mode. When the different

solutions viz. degraded samples and filtered tablet solution were scanned in derivative mode, the zero crossing was not deviated from the 242 nm and 272 nm. No deviation from these values indicates, there was no interference of any excipient in absorbance of Gaussian spectrum of the tapentadol hydrochloride. The six replicates of all dilutions were processed for the linear regression method and the linear regression equation in the derivative mode was found to be Y = 0.080 x + 0.001 with correlation coefficient R^2 = 0.9990.

Table 3. **Calibration graph of tapentadol hydrochloride in water for derivative method.**

Conc. (µg/mL)	Absorbance* at 221 nm in first order derivative mode					
	I	***II***	***III***	***IV***	***V***	***VI***
10	0.021	0.022	0.020	0.021	0.022	0.021
20	0.041	0.042	0.042	0.041	0.043	0.043
30	0.060	0.061	0.062	0.060	0.063	0.061
40	0.080	0.081	0.082	0.080	0.083	0.082
50	0.101	0.104	0.101	0.102	0.103	0.104
Regression Equation Y = 0.080 x + 0.001; R^2 = 0.9990						

* All absorbance are in negative value

Validation of methods

As per ICH guidelines, all the methods were validated within limits to assure the reliability of the methods. The linearity for the all the methods (LRE, SA and FOD method) were determined as 99.49%, 99.92% and 100.58% respectively, which are acceptable as standard deviation was within limit. Accuracy was determined by recovery method and found to be 100.23%, 101.02% and 100.41% respectively, with less than one unit of standard deviation. Thus, all three methods are linear with accurate result within acceptable standard deviation results.

The repeatability and intermediate precision were studied to assure the precision of the methods. The repeatability of the methods was around the 100% with less than one unit of standard deviation for all the methods. The intermediate precision was studies for variation in day of analysis, analyst-to-analyst and instrument-to-instrument. The results for all variations were within 99.56-101.13% limit for all methods. All the methods were also validated for temperature variation of analysis to study the robustness (Table 4) and the variation results were in between 99.67-101.11% with less the unit standard deviation. Results of the validation parameters have been proved that all methods may be equally applicable.

Table 4. **Results of validation parameters for all three methods.**

Validation parameter	% Found (mean)* ± SD		
	LRE method	**SA method**	**FOD method**
Linearity	99.49 ± 0.087	99.92 ± 0.096	100.58 ± 0.097
Accuracy	100.23 ± 0.093	101.02 ± 0.072	100.41 ± 0.027
Precision	100.27 ± 0.094	100.03 ± 0.059	100.17 ± 0.098
I. Repeatability	99.56 ± 0.089	101.11 ± 0.085	100.08 ± 0.052
II. Intermediate precision			
a. Days	100.84 ± 0.059	100.19 ± 0.079	99.98 ± 0.038
b. Analysts	101.08 ± 0.083	99.97 ± 0.090	101.13 ± 0.023
c. Instruments	101.11 ± 0.098	99.99 ± 0.074	99.96 ± 0.057
Robustness			
a. 35°C	100.09 ± 0.089	101.08 ± 0.098	100.03 ± 0.076
b. 25°C	99.94 ± 0.027	100.64 ± 0.037	99.67 ± 0.041
c. 15°C	101.11 ± 0.077	99.74 ± 0.082	100.37 ± 0.088

* mean of six dilutions in three replicates, SD = standard deviation

Table 5. **Analysis of tapentadol hydrochloride in tablets.**

Batch ↓	Determined % of drug content by validated methods								
	LRE method			SA method			FOD method		
Conc. (µg/mL) →	20	30	40	20	30	40	20	30	40
I	100.56	101.05	99.89	101.03	100.98	100.86	99.79	101.01	99.25
II	100.34	101.19	100.98	100.32	99.98	100.43	101.05	99.72	101.01
III	100.01	99.91	100.05	99.96	100.23	99.84	99.78	100.06	99.61
IV	99.95	100.84	99.88	100.18	100.84	101.01	101.04	99.38	101.09
V	101.07	99.86	100.71	100.98	99.85	100.02	101.02	101.14	99.35
VI	100.91	100.98	101.02	100.77	99.97	101.01	100.84	99.27	100.44
Mean	100.47	100.64	100.42	100.54	100.31	100.53	100.59	100.10	100.13
SD	0.46	0.59	0.54	0.45	0.48	0.51	0.63	0.81	0.83

Analysis of dosage form

As already discussed that tapentadol hydrochloride was freely water soluble; the powder of the tablet was sonicated with distilled water to extract the drug content from the tablet. All the methods were applied to determine the drug content at the three levels (20, 30 and 40 µg/mL) as per above described methodology. The drug was assay by all the validated methods in the range of 100.42-100.64%, 100.31-100.54% and 100.10-100.59%, respectively (Table 5). The standard deviation was found to be less than one unit (0.45-0.83).

Conclusion

Hence, three spectrophotometric methods were developed and validated viz. linear regression equation (LRE), standard absorptivity (SA) and first order derivative (FOD) and successfully applied to determine tapentadol hydrochloride in dosage forms. These methods were proven to be highly cost effective as compare to previous spectrophotometric methods because the solvent was distilled water. All methods have been proved equally applicable for the determination of the drug content in tablet dosage from. Therefore, validated methods can be applied to determine tapentadol hydrochloride in bulk drugs, different dosage forms, dissolution studies, bioequivalence studies, degradation studies and in routine pharmaceutical industries.

Acknowledgement

One of the authors, Pawan Kumar Basniwal, earnestly indebted to Science and Engineering Research Board (SERB), DST, New Delhi, India, for the financial support for this research work under Fast Track Scheme for Young Scientists. Authors are highly thankful to Head, School of Pharmaceutical Sciences, RGPV, Bhopal and Principal, LBS College of Pharmacy, Jaipur for providing the experimental facilities for this research work.

References

1. Pre-review report on tapentadol, Expert committee on drug dependence, Thirty-fifth meeting hammamet, Tunisia, *WHO*, (2012).
2. D. Jain and P. K. Basniwal, *Bulletin Faculty Pharm, Cairo University* 51, (2013) 283.
3. M. Giorgi, A. Meizlerb and P. C. Mills, *J. Pharm. Biomed. Ana.* 67-68 (2012) 148.
4. A. B. James, Collins A. A., A. C. Scot, R. Sumankalai and C. B. Ronald, *J. Ana. Toxico.* 34, (2010) 450.
5. C. Coulter, M. Taruc, J. Tuyay and C. Moore, *J. Ana. Toxico*. 34 (2010) 458.

6. M. Giorgi, A. Meizler and P. C. Mills, *The Veterinary J.* 2012 (in-press). http://dx.doi.org/10.1016/j.tvjl.2012.05.019.
7. V. G. Khokhar and A. Agola, *Internat. J. Pharm. Tech. Res.* 5, (2013) 414.
8. O. D. Sherikar and P. J. Mehta, *J. Chem. Pharm. Res.* 4, (2012) 4134.
9. D. Jain and P. K. Basniwal, *J. Ana. Sci. Tech.* 4, (2013) 9.
10. S. B. Bysani, K. P. Kancharla, N. Kalakonda and N. Ramakrishna, *Der Pharmacia Lettre* 5, (2013) 377.
11. ICH "Text on Validation of Analytical Procedures", International conference on harmonization of technical requirements for registration of pharmaceutical for human use, Geneva, (2000).
12. P. K. Basniwal, V. Kumar, P. K. Shrivastava and D. Jain, *Tropical J. Pharm. Res*. 9, (2010) 499.