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Analytical Method Development and Validation of Tapentadol Hydrochloride in Tablet Formulation By UV Spectroscopic Method

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ABSTRACT

A simple method for the estimation of Tapentadol Hydrochloride in bulk and pharmaceutical dosage forms has been developed. Water was chosen as solvent system. The λ_{max} was found to be 272nm.The responses were linear in the range of 15-65µg/ml.The regression equation of calibration graph and correlation coefficient were found to be y=0.006+0.008 and0.999 respectively. The %RSD values for both intraday and interday precision were less than 1%. Recovery of the drug from the sample was ranged between 98-102%.The proposed method was validated for Precision,Accuracy,Intraday,Interday Assay,commercial tablets containing 50mg of Tapentadol Hydrochloride was analyzed by the proposed method and the results were well within the claimed limits. Further stability studies of Tapentadol Hydrochloride were carried out under acidic, alkaline, hydrolytic and photolytic conditions as per ICH Guidelines.

KEY WORDS :- Tapentadol Hydrochloride; ICH Guidelines; Validation; UV Spectroscopic Method.

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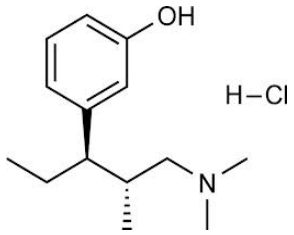
INTRODUCTION

Tapentadol is a novel centrally acting analgesic that was approved for use by the Food and Drug Administration in November 2008. It has structural similarities to tramadol. The drug has a unique mode of action in that it functions as an agonist at the mu -opioid receptor, and as a norepinephrine reuptake inhibitor. This dual mode of action provides analgesia at similar levels of more potent narcotic analgesics such as hydrocodone, oxycodone and morphine, but with a more tolerable side effect profile[18-19]. The chemical name is 3 - [(1*R*, 2R) -3 - (dimethylamino) –l - ethyl-2 -methylpropyl] phenol monohydrochloride. The molecular weight of Tapentadol HC1 is 257.80, and the molecular formula is C14H23NO·HCl. The n-octanol: water partition coefficient log P value is 2.87.



Tapentadol Hydrochloride

The Objective of this Research work is to Develop and Validate[2,3] a simple, precise, accurate and robust method for routine estimation of Tapentadol HCl from tablet dosage form using UV spectroscopic method according to the ICH guidelines[1] and the Literature Review reviles that methods have been reported using HPLC and LCMS methods but no methods has been reported using UV spectroscopic method[4,5]. As UV spectroscopic method is simple and cheap.

MATERIALS AND METHODS

Instruments:

Shimadzu UV-1800, UV-Visible double beam Spectrophotometer with matching pair of 1 cm quartz cuvettes (Shimadzu Corporation, Kyoto, Japan). The spectral bandwidth is 0.5 nm.

Reagents and Chemicals

Pure drug samples of Tapentadol hydrochloride were obtained as gift sample. Combined dose tablet formulation (TYDOL) was procured from market. Sodium hydroxide Hydrochloric acid Hydrogen peroxide Distilled water

(a)**Selection of solvent**

Tapentadol hydrochloride pure form and its market formulation were found to be freely soluble in water and other organic solvents. Hence water was selected as solvent for UV spectroscopic determination.

(b)**Preparation of stock solution**

A standard stock solution of Tapentadol hydrochloride was prepared by dissolving 25mg of drug in 25ml of distilled water. Above stock solution was further diluted with same solvent to get the required concentrations.

(c) **Determination of λmax**

The stock solution of Tapentadol hydrochloride having the concentration 1000µg/ml was further diluted to 25µg/ml with distilled water. The absorbance of resulting solution was scanned in the UV spectrometer ranging from 200-400nm.The plot shows maximum absorbance at 272nm.

Preparation of sample solution

Commercially available tablets of Tapentadol hydrochloride (TYDOL) 50mg were selected for the estimation of total content by the proposed method. An amount equivalent to 0.025g of Tapentadol hydrochloride was weighed accurately and transferred into 25 ml volumetric flask , containing 5 ml of distilled water, mix thoroughly and make up the volume with distilled water. From this solution transfer 1ml in to 10ml volumetric flask and make up it with distilled water, and again transferred 3.5ml in to 10ml volumetric flask and make up it with distilled water to get the final concentration 35µg/ml.and absorbance was measured at272nm.

$$\text{Percentage purity} = \frac{\text{Sample absorbence} * \text{standard dilution} * \text{Average weight} * \text{potency}}{\text{Standard absorbence} * \text{sample dilution} * \text{lable claim} * 100} * 100$$

Spectrophotometric Analysis for Single Component Drug[6]

Single component samples are analyzed by using fallowing methods

(a) Standard absorptivity value method.

(b) Calibration graph method

(c) Single (or) double point standardization.

(a)Standard Absorptivity Value

Directly determine absorptivity of given sample by using standard, ε & a value.

We can represent Beer-Lambert's law as,

$$A = \varepsilon bc$$

Where, A = absorbance

b =path length and ε = Molar absorptivity.

By using above formulae we can easily determine concentration of the sample.The above procedure is adopted by official Pharmacopeia like British Pharmacopoeia for stable substance. e.g Methyl testosterone.

(b) Calibration Graph Method[7]

- In this procedure the absorbance of number of standard solutions are noted.
  The concentration of standard solutions should encompassing the sample concentration.
- Then calibration graph is constructed.
- The concentration of analyte in the sample solution read from the graph as concentration corresponding to the absorbance of solution.

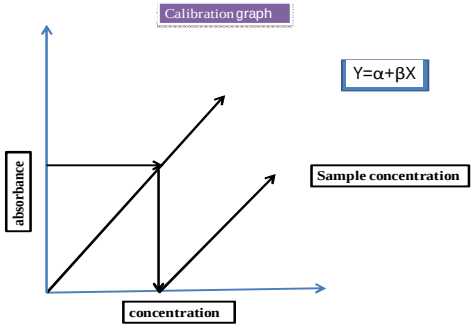


Figure 1: Calibration Graph

- If the absorbance values & concentrations bear a linear relationship, then we obtain the regression line Y=α+βx

$$\alpha = \frac{(\sum y)\,(\sum x^2) - (\sum x)\,(\sum xy)}{N\sum x^2 - (\sum x)^2}$$

$$\beta = \frac{N\sum xy - (\sum x)\,(\sum y)}{N\sum x^2 - (\sum x)^2}$$

Where, y = absorbance,

x = concentration, N = number of pairs of values.

(c)Single (Or) Double Point Standardization.

Single point procedure involves measurement of the absorbance of sample & standard solutions. Sample & standard solutions should prepare in similar manner, concentrations should be close.

Absorbance α concentration

$$C_{test} = \frac{(A_{test} * C_{std})}{A_{std}}$$

Generally it obeys Beer's law:

- Occasionally, it shows linear but non-proportional relation between concentration & absorbance which shows a significant +ve (or) –ve intercept. When there is deviation from Beer's Law then two point standardization is necessary.
- The concentration of one of the standard solution should be greater and the other standard solution should be lower than the sample solution.

$$C_{test} = \frac{(A_{test} - A_{std})(C_{std_1} - C_{std_2}) + C_{std_1}(A_{std_1} - A_{std_2})}{A_{std_1} - A_{std_2}}$$

Where

C_{std} :-Standard Concentration

C_{test} :-Test Concentration

A_{test} :-Test Absorbance

A_{std} :-Standard Absorbance

RESULTS AND DISCUSSION

λmax of Tapentadol hydrochloride

Determination of λmax of Tapentadol hydrochloride

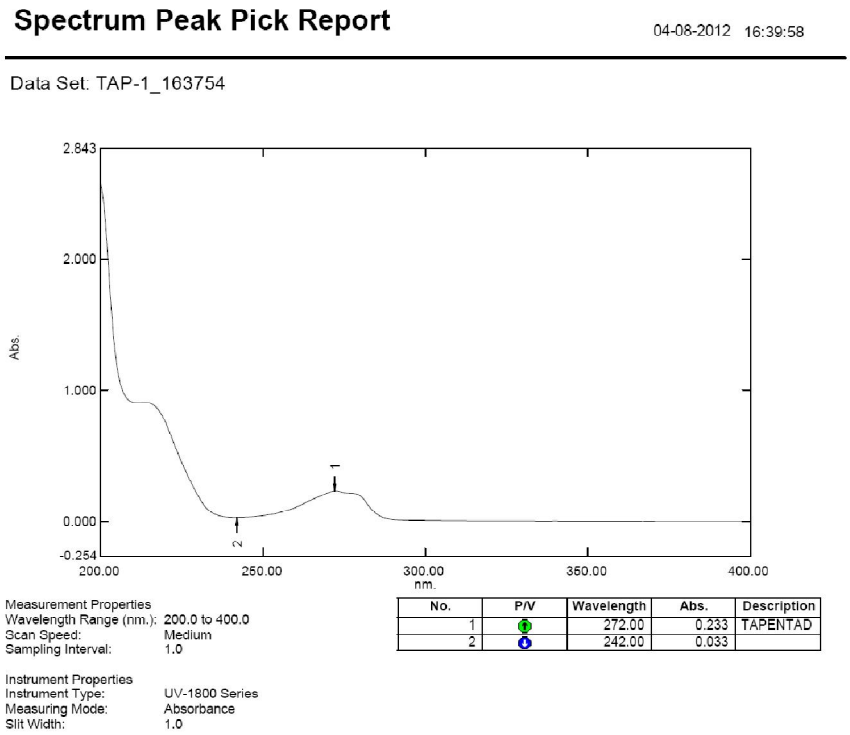


Figure 2: spectrum of Tapentadol (λmax)

The λmax of Tapentadol hydrochloride by using UV spectrophotometer was found to be 272 nm

Validation Method Parameters

1. Specificity

Degradation Studies

(a)Heat

Prepare 100µg/ml concentration solution by diluting 1ml of stock solution in 10 ml in volumetric flask with distilled water then heat it at 80^{o}C for one hour then take 3.5 ml of this solution in 10ml volumetric flask and make up the volume with solvent

(b)UV light

Prepare 100µg/ml concentration solution by diluting 1ml of stock solution in 10 ml in volumetric flask with distilled water then expose it to UV light for one hour then take 3.5 ml of this solution in 10ml volumetric solvent and make up the volume with solvent

(c)Acid

Prepare 100µg/ml concentration solution by diluting 1ml of stock solution in 10 ml in volumetric flask with distilled water then make up it with 5N HCl then heat it at 80^{o}C for one hour, take 3.5ml of this solution in10ml volumetric flask and make up the volume with solvent.

(d)Base

Prepare 100µg/ml concentration solution by diluting 1ml of stock solution in 10 ml in volumetric flask with distilled water then make up it with 5N NaOH then heat it at 80^{o}C for one hour, take 3.5ml of this solution in10ml volumetric flask and make up the volume with solvent.

(e)H_2 O_2

Prepare 100µg/ml concentration solution by diluting 1ml of stock solution in 10 ml in volumetric flask with distilled water then make up it with 5N H_2 O_2 then heat it at 80^{o}C for one hour, take 3.5ml of this solution in10ml volumetric flask and make up the volume with solvent.

Table 1 Specificity table

Parameter	Observation
Light	No interference at absorption maxima of analyte
Temperature	
Acid	
Base	
Peroxide	

The uv spectrums of degraded products showed no interference at the analyte peaks. Hence the developed method was highly specific.

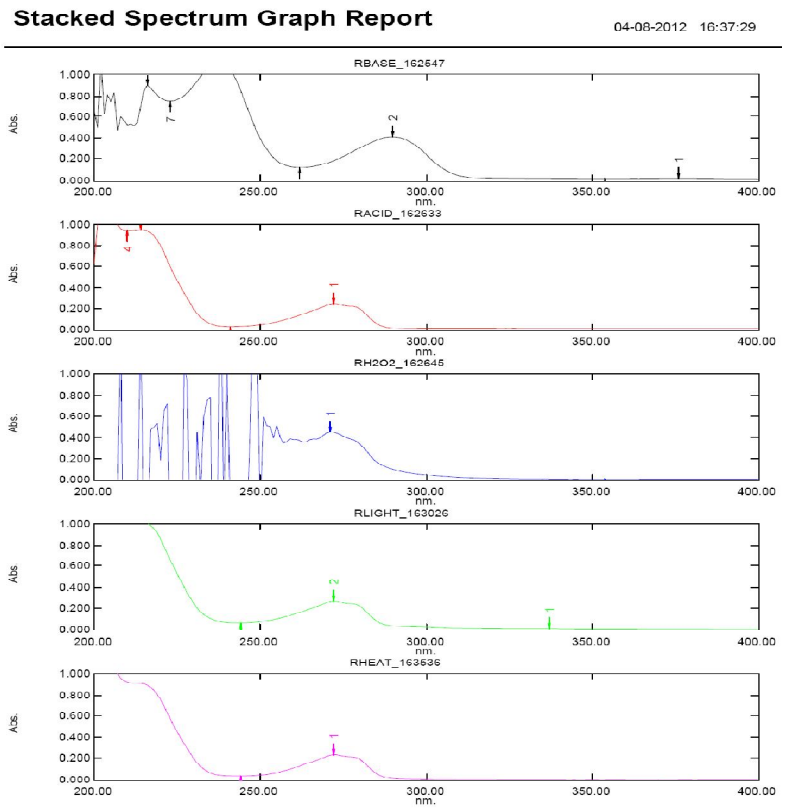


Figure 3: spectrum of Degradation Report

2. Accuracy:

LQC (20µg/ml), MQC (35µg/ml) and HQC (50µg/ml) were prepared, that solutions are spiked with standard nominal concentration (35µg/ml). Absorbance values of these solutions were measured at 272nm.The recovery studies were performed.

Table 2: Accuracy table

No. of preparations	Concentration (µg/ml)		Asorbance	Sample Recovery	Mean	% recovery
	Sample	Spiked Standard				
A1:LQC1	20	35	0.378	20.06	20.29	101.4
A2:LQC2			0.394	20.92		
A3:LQC3			0.374	19.89		
B1:MQC	35	35	0.480	35.65	35.21	100.6
B2:MQC			0.470	34.92		
B3:MQC			0.472	35.06		
C1:HQC1	50	35	0.575	50.07	50.21	100.4
C2:HQC2			0.582	50.62		
C3:HQC3			0.574	49.96		

The difference between the assay obtained from average of 9 determinations (3conc/3replicates) were within the limits. The %Recovery is 98%-102%.

3 Precision:

Precision of the method was demonstrated by Repeatability, Intraday and Interday variation studies using six replicates of MQC (35µg/mL) and Absorbance values of these solutions were measured at 272nm. Results were determained in the form of %RSD.

3.1 Intermediate precision

Intermediate precision was performed by preparing MQC (35µg/ml) and measuring absorbance in different days, different analysts, different equipment, etc. Results were determained in the form of %RSD.

3.2 Reproducibility

Reproducibility was performed by preparing MQC (35µg/ml) and measuring absorbance in different laboratories. Results were determained in the form of %RSD.

Table 3 precision table

Concentration (µg/ml)	Absorbance	S.D	%RSD
35	0.245	0.002074	0.84
	0.246		
	0.245		
	0.248		
	0.250		
	0.245		

The assay results of 6 determinations of 35µg/ml concentrations show results within limits. The RSD was found to be 0.84.

3.3 Intraday assay precision

Table 4: intraday precision table

	Time in Min.	
Concentrations(µg/ml)	10.00Am	4.00pm
35	0.248	0.246
35	0.251	0.245
35	0.245	0.245
35	0.245	0.248
35	0.248	0.250
35	0.250	0.245
%RSD	1	0.84

The intraday precision of 6 determinations having the same concentrations shows results within the limits.

3.4 Interday assay precision

Table 5 interday precision table

Concentrations (µg/ml)	%RSD	
	Day1	Day2
35	0.95	0.84

The interday assay precision results was found to be within the limits.

4 Linearity profile

From the stock solution various dilutions were made to obtain solutions of 15, 25, 35, 45, 55 and 65µg/ml. Absorbance values of these solutions were measured at 272nm.The calibration curve was plotted between concentration of Tapentadol hydrochloride and respective measured absorbances. And R^2 was found to be within acceptable range.

Table 6 Linearity Table

Concentration (µg/ml)	Absorbence
15	0.108
25	0.175
35	0.235
45	0.304
55	0.372
65	0.436

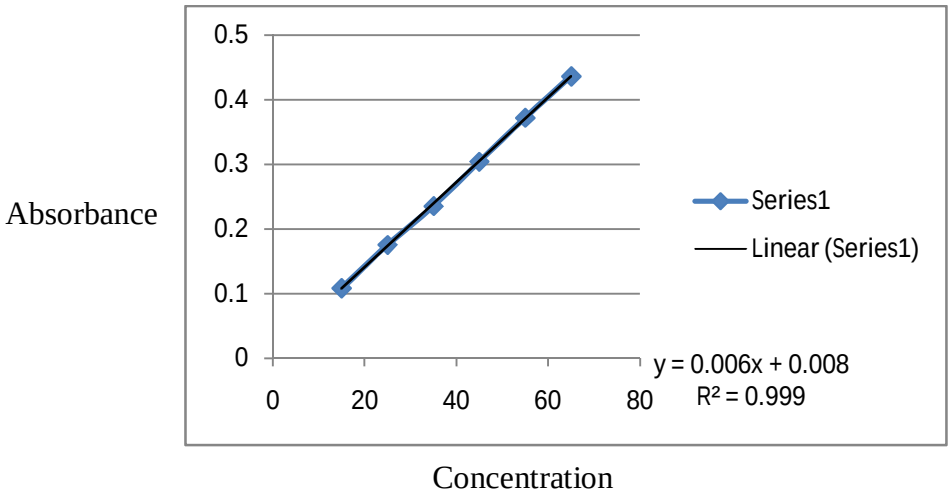


Figure 4: Linearity Curve

Table 7: statistical parameters

Statistical parameters	Results
Wavelength	272nm
Slope	0.006
Intercept	0.008
Regression coefficient	Y=0.006x+0.008
Correlation coefficient	0.999

Tapentadol Hydrochloride was found to be linear over the range of 15 -65 µg/mL and coefficient of variation was found to be 0.999.

5 Limit of Detection:

Limit of detection was measured on the basis of standard deviation of the response and slope and slope is estimated from the calibration curve of the analyte and standard deviation is calculated from calibration curve (standard deviation of y- intercepts of regression lines).

LOD=3.3*S.D/slope

6 Limit of quantification:

Limit of detection was measured on the basis of standard deviation of the response and slope and slope is estimated from the calibration curve of the analyte and standard deviation is calculated from calibration curve (standard deviation of y- intercepts of regression lines).

LOQ=10*S.D/slope

Table 8 LOD&LOQ table

Parameter	Tapentadol Hydrochloride (µg/ml)
Standard deviation	0.00433
LOD	2.113
LOQ	6.405

LOD=3*SD/slope of calibration curve

LOQ=10*SD/slope of calibration curve

Standard deviation (SD)=0.00433

Slope (b)=0.006

Limit of detection and limit of quantitation was found to be 2.113 and 6.405 respectively.

7. Robustness:

Robustness of the method was determined by carrying out the analysis under different wavelength conditions and variation in mobile phase dilution. The respective absorbances were noted and the result are reported as in the terms of %RSD.

Table 9 Robustness table

S.NO.	Parameter	Absorbance		
		Minimum	Optimum	Maximum
1	Variation in mobile phase	+1ml	0ml	-1ml
		0.232	0.237	0.236

S.NO.	Parameter	Absorbance		
		Minimum	Optimum	Maximum
1	Wavelength(nm)	At 271	At 272	At 273
		0.244	0.249	0.248

The robustness was carried out by changing parameters like variation in mobile phase and Wavelength. It was found that these parameters were within the acceptable limits.

Assay of marketed Formulation:-Tydol-50mg

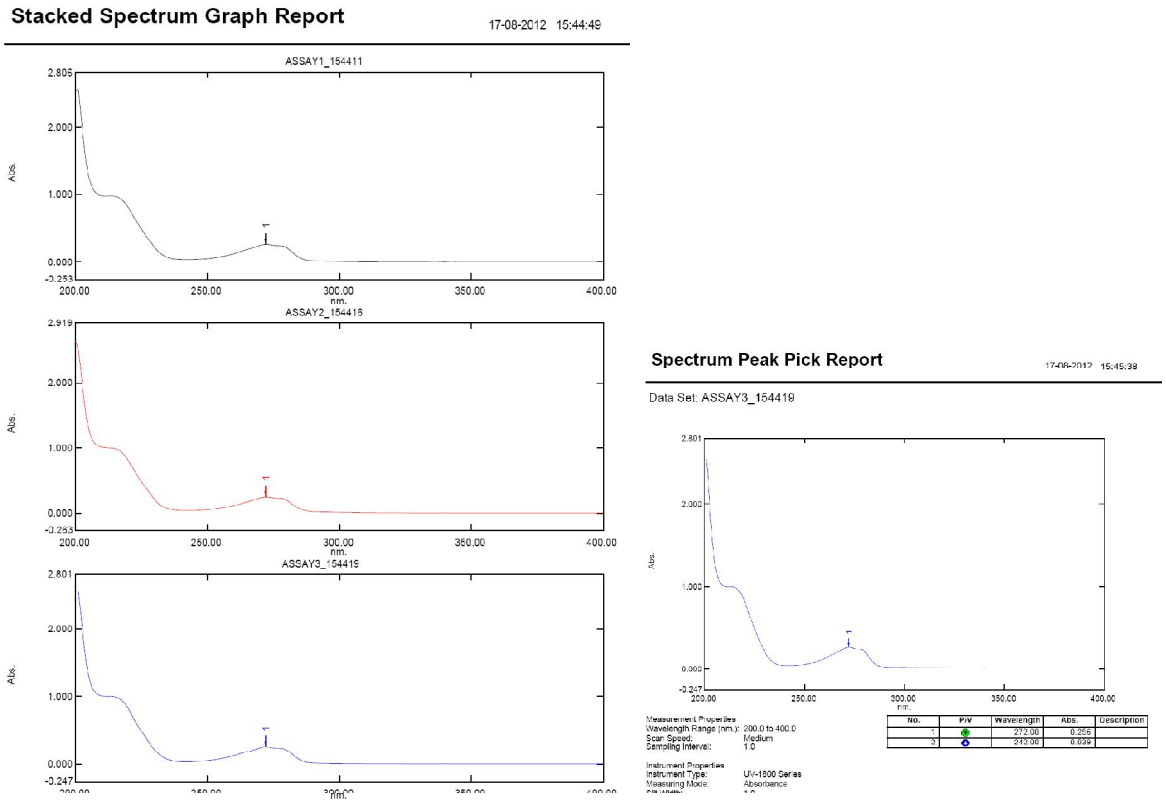


Figure 5: Assay Reports

Spectrum Method Report 17-08-2012 15:45:24

Data Set: ASSAY3_154419

Measurement Properties
Wavelength Range (nm.): 200.0 to 400.0
Scan Speed: Medium
Sampling Interval: 1.0

Instrument Properties
Instrument Type: UV-1800 Series
Measuring Mode: Absorbance
Slit Width: 1.0

Software Information
Software Name: UV-1800 Series
Version:
Mode: Normal Mode

Data Information
Data is: Modified
Analyst:
Date/Time: 08-08-2012 15:39:31
Comments:

Instrument Information
Instrument Name: UV
Instrument Type: UV-1800 Series
Model (S/N):

Table 10 Assay table

S. No	Concentration in µg/ml	Absorbance	Recovary in µg/ml	%Purity
Assay1	35	0.251	34.4	98.29
Assay2		0.248	35.7	102.12
Assay3		0.256	35.14	100.42

The percentage purity of marketed formulation of tapentadol was found to be With in the acceptable limits.

CONCLUSION

The developed UV spectroscopic method shows linearity in the range of 15-65µg/mL concentration of Tapentadol hydrochloride by using water as solvent medium. This method was proved to be accurate as the percentage recovery values were found to be with in the acceptable limits. The %RSD values for repeatability, intraday precision and inter day precision lie within the limits.LOD and LOQ values were found to be 2.113 and 6.405 respectively. The developed method was highly specific, robust and can be used for routine analysis of tapentadol hydrochloride in tablet formulations.

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