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Development and Validation of Stability Indicating Assay Methods (SIAMs) for Citalopram HBr by Using UV-Visible Spectrophotometer and RP-HPLC

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ABSTRACT

A simple, fast, accurate and precise method has been developed UV-Vis spectrophotometric method for the estimation of citalopram HBr in bulk and pharmaceutical dosage form. The solvent used was distilled water and the absorption maxima. (λ max) of drug was found 239 nm. A linear response was observed in the range of 4-20 µg/ml with a regression coefficient of 0.999. Citalopram HBr was subjected to acid hydrolysis, alkali hydrolysis, oxidation, photo and thermal degradation. The citalopram shows relative high thermal and photo stability. i.e.(0.15% at 48 hours and 0.21% at 72 hours) as compared to other stability conditions. The reversed-phase high performance liquid chromatography method has been developed for citalopram HBr. The separation and degradation product was carried out on C_{18} column Phenomenex-250×4.6mm×5µm using mobile phase consisting of acetonitrile: water (75:25%v/v), at a flow rate of 1.5 ml/min and with detection wave length 239 nm. The retention times for citalopram HBr was 5.56 min. The calibration plots were linear over the concentration range of 10-60 µg/ml with regression coefficient of 0.999 for citalopram HBr. Citalopram HBr was subjected to acid and alkali hydrolysis, oxidation, and photo and thermal degradation. The citalopram shows relative high thermal and photo stability. i.e.(0.64% at 48 hours and 0.44% at 72 hours) as compared to other stability conditions. This method was then validated for different parameters as per the international conference for harmonization guidelines (ICH). This HPLC and UV methods can be used for the determination for the stability indicating assay methods for citalopram HBr & its formulations.

Key words: Citalopram HBr, SIAMs, stress conditions studies, validation, and UV-Vis spectroscopy, HPLC

INTRODUCTION

Citalopram HBr is chemically known as 1-[3-(dimethyl amino) Propyl]-1-(4-Flurophenyl)-1, 3- dihydro-2 isobenzofuran-5-Carbonitrile.Citalopram is a widely used antidepressant comes under the class of selective serotonin reuptake inhibitors (SSRIs) having broad spectrum of therapeutic activity against depressive disorders. It exerts effect through a selective inhibition for reuptake of neurotransmitter serotonin by the pre-synaptic receptors. It is prescribed in the treatment of related disorders, such as obsessive-compulsive disorder, anxiety, social phobia, and posttraumatic stress, but has many concentrations dependent adverse effects also [1] .Citalopram is a Pgp substrate and is actively transported by that protein from the brain. The efficiency of citalopram in people possessing a certain version of P-gp (genetic TT-allele) is likely to be diminished. This suggests that in non-responders to citalopram a switch to antidepressant which is not a Pgp substrate, such as fluoxetine (Prozac, Fontex) or mirtazapine (Remeron) but not to venlafaxine (Effexor), amitriptyline (Elavil) or paroxetine (Paxil), which are Pgp substrate may be beneficial. Distinct from some other agents in its class, citalopram exhibits linear pharmacokinetics and minimal drug interaction potential, making it a better choice for the elderly or comorbid patients [2]. Citalopram is available in 10 mg, 20 mg, and 40 mg tablets, as well as 10 mg containing oral solutions in more than 30 drug formulations[3].Presence of nitrile group, fused teterahydrofuran ring, and C-N linkage in structure of the drug

makes it susceptible to degradation due to chemical reactivties of these groups under hydrolytic, oxidative, and photolytic environments, which will eventually produce varied impurities or degradation products[4].Hence, the present study is undertaken to conduct ICH prescribed systematic forced degradation study on citalopram to identify its potential degradation products and to develop and validate a SIAM by using UV-Vis and RP-HPLC for accurate determination of citalopram in the presence of its impurities and degradation products. The aim is to provide new and demonstrate the present data on impurities and its determination methods. Chemical structure of citalopram HBr as shown in figure no.1

MATERIALS AND METHODS

UV Method

1. UV Method development

Materials and methods

The drug, citalopram HBr was procured as a gift sample from Gen Pharma Pvt. Ltd. Pune. The instrument used for the present study was a Jasco V-630 UV-Vis Double Beam Spectrophotometer. The solvent used was distilled water. The chemicals NaOH, HCl, and H_2O_2 used for present study are of AR grade and these chemicals are purchased from Merck Chemicals (Mumbai, India).

Preparation of stock solution

Standard stock solution of citalopram HBr was prepared by dissolving 10 mg citalopram HBr in 100 ml of distilled water which gives 100 µg/ml solution.

Preparation of series of dilution

From the above stock solution 2 ml was transferred into 10 ml volumetric flask and volume was make up with distilled water up to which it gives 20 µg/ml. Citalopram HBr was scanned with UV-Visible Spectrophotometer in the range 200-400 nm against distilled water as blank and the wavelength corresponding to maximum absorbance was noted which is its max i.e. at 239 (figure no. 2).

Preparation of calibration curve

0.4 ml-2 ml of 100 µg/ml solution were diluted and the volume was made up to 10 ml using distilled water to produce 4-20 µg/ml sub stock solutions respectively. Then the absorbance of solution was taken. The calibration curve of citalopram HBr was plotted by taking concentration on X-axis and absorbance on Y-axis (figure no. 3). This drug shown line in concentration range of 4-20 µg/ml and correlation coefficient was found to be 0.999.

2. UV Method Validation [5-7, 9-12]

Linearity

Various aliquots were prepared from the stock solution (100µg/ml) ranging from 4-20 µg/ml. The samples were scanned in UV-Vis Spectrophotometer against distilled water as blank. It was found that the selected drug shows linearity between the ranges of 4-20 µg/ml (Table no.7 and figure no. 3).

Accuracy

Solutions were prepared in triplicate at levels 80%, 100%, and 120% of test concentration using citalopram HBr working standard as per the method and taken absorbance of each solution in triplicate. The recovery result showed that the proposed method has an acceptable level of accuracy for citalopram HBr which is from 80% - 120% of test concentration is from 99.45 % - 99.98% (Table no. 2).

Precision

Precision of the method was demonstrated by intraday and interday variation studies. In intraday variation study six different solutions of concentration 12, 16, 18µg/ml were analyzed three times in a day i.e. at morning, afternoon and evening. In the interday variation studies, solution of same concentration i.e. 12, 16, 18 µg/ml were analyzed three times for the three consecutive days and the absorbance result, mean, standard deviation and % RSD was calculated (Table no. 3,4).

Robustness

Robustness of the method was determined by carrying out the analysis under temperature condition i.e. at 18°C.The respective absorbance's of 12 µg/ml were noted and the result was indicated as % RSD (Table no. 5).

Ruggedness
Ruggedness of the method was determined by carrying out the analysis by different analyst and the respective absorbance of 18µg/ml was noted. The result was indicated as % RSD (Table no. 6).

Limit of Detection (LOD)
The limit of detection (LOD) was separately determined based on the standard deviation of response of the calibration curve. The standard deviation of the y intercept and slope of the calibration curve were used and it was found to be 0.08662µg/ml (Table no.7).

Limit of Quantification (LOQ)
The LOQ is the concentration that can be quantification reliably with a specified level of accuracy and precision. The LOQ was calculated using the formula involving standard deviation of response and slope of calibration curve and it was found to be 0.2625µg/ml (Table no.7).

3. Degradation studies by UV [6-14]
Acid Degradation
In six different volumetric flasks, 2 ml of stock solution of citalopram bulk drug was added and mixed with 3 ml of 0.1N, 1N, 2N, 3N, 4N, 5N hydrochloric acid in each volumetric flask respectively. The volumetric flask was kept for 3 hours at room temperature. After every 1 hour time interval, solution was neutralized and diluted with distilled water in order to make the volume up to 10 ml and the dilution was carried out to achieve the appropriate concentration (20µg/ml). The degradation was observed in the 5N hydrochloric acid. (Table no.8 and Figure no. 4)

Alkali Degradation
In six different volumetric flasks 2 ml of stock solution of citalopram bulk drug was added and 5 ml of 0.1N, 0.5N, 1N, 2N, 3N, 3.5N sodium hydroxide was added in each volumetric flask respectively. The volumetric flask was kept for 1 hour at room temperature. After every 1 hour time interval, solution was neutralized and diluted with distilled water in order to make the volume up to 10 ml and the dilution was carried out to achieve the appropriate concentration (20µg/ml). The degradation was observed in the 1 N NaOH. (Table no.8 and Figure no. 5)

Oxidative degradation
2 ml of stock solutions of citalopram HBr and 2 ml of 30% H_2O_2 was added in 10 ml of volumetric flask and volumetric flask was kept at normal condition for 3 hours. After 3 hours time interval diluted with water in order to make the volume it gives 20 µg/ml solutions. (Table no.8 & Figure no. 6)

Thermal degradation
Citalopram HBr sample were taken in a petri plate and exposed to a temperature of 80°C for citalopram was for 48 hours in an oven. After 48 hours, 10 mg of citalopram was diluted with water in order to make the volume up to 10 ml which gives 20 µg/ml solution and the solution was scanned for the UV–Vis analysis. (Table no.8 and Figure no.7)

Photo-Degradation
Sample of citalopram was exposed to UV Light for 72 hours, 10 mg of citalopram was dissolved in water and volume was made up to 10 ml which gives 1000 µg/ml solution and scanned for the UV (239 nm) analysis.0.2 ml of this solution was diluted to 10 ml to attain concentration 20 µg/ml. (Table no. 8 & Figure no. 8)

HPLC Method
1. HPLC Method Development
Materials & Methods
Active pharmaceutical ingredient of citalopram HBr was received as gift samples from Gen pharma Ltd. Pune. Acetonitrile (HPLC grade), methanol (HPLC grade) and Chloroform (AR grade) were procured from Merck Chemicals, India.

Equipment and chromatographic conditions
The HPLC system used was a cyber-lab HPLC system equipped with a Rheodyne injector (loop size-20 µl) and UV detector. Mobile phase was prepared by mixing acetonitrile: water (pH adjusted 5 with ortho phosphoric acid) (75:25 v/v), filtered through 0.45µ membrane filter paper and then sonicated on ultrasonic water bath for 30 min. The injection volume was 20 µl and eluted at a flow rate of 1.5 ml/min. The detection wavelength was 239 nm.

Preparation of Stock Solution

10 mg of citalopram HBr bulk drug was weighed accurately and transferred in 100 ml volumetric flask. Drug was dissolve in acetonitrile: water (75:25 v/v) and volume was made up to 100 ml with same solvent so as to get the concentration 100 ug/ml. 2 ml standard stock solution of citalopram HBr was then diluted in 10 ml acetonitrile: water (75:25 v/v) to get working standard solution 20 ug/ml.

Preparation of mobile phase
Mobile phase was prepared by mixing acetonitrile:water (pH adjusted 5 with ortho phosphoric acid) (75:25 v/v), filtered through 0.45 µ membrane filter paper and then sonicated on ultrasonic water bath for 30 min.

Chromatogram of Citalopram HBr
The column was saturated with the mobile phase (indicated by constant back pressure at desired flow rate). Standard solution of citalopram HBr was injected to get the chromatogram. The retention time for citalopram HBr was found to be 5.56 min. Chromatogram of citalopram HBr is shown in (Figure no.9)

2. **HPLC Method Validation**[9-12]

Linearity
1-6 ml of 100 µg/ml solution was diluted and the volume was made up to 10 ml using mobile phase to produce 10-60 µg/ml solutions respectively. The peak area was measured and standard calibration curve was plotted as concentrations Vs Peak area. This straight line obeyed linearity in the concentration range of 10-60 µg/ml. The correlation was found to be 0.99. (Table no.14 and Figure no.10)

Precision
The precision of the method was demonstrated by intra-day and inter-day variation studies. In the inter day studies, 3 different concentrations 30, 40 and 50 µg/ml were injected in stabilized chromatographic conditions and were analyzed in triplicate. The percentage RSD was calculated. The result obtained for intraday and interday variations are shown in (Table no.10 and11)

Accuracy
To check accuracy of the method, recovery studies were carried out by mixing standard drug solution to pre-analyzed sample solution at three different levels 80, 100 and 120%. Basic concentration of sample chosen was 25 µg/ml of citalopram HBr bulk drug solution to which 25, and 30 µg/ml of citalopram HBr tablet solution was added. These solutions were injected in stabilized chromatographic conditions in triplicate to obtain the chromatograms. The drug concentrations of citalopram HBr were calculated by using linearity equation. The results obtained are shown in table no.12.In pure drug proportion was 25 µg/ml i.e. 2.5 ml; consider as 100 % so calculate the 80% and 120% level of recovery and calculated how much standard (pure drug) solution was added into the tablet.

Limit of Detection (LOD)
The limit of detection (LOD) was separately determined based on the standard deviation of response of the calibration curve. The standard deviation of the y intercept and slope of the calibration curve were used and it was found to be 0.06052µg/ml (Table no.14).

Limit of Quantification (LOQ)
The LOQ is the concentration that can be quantification reliably with a specified level of accuracy and precision. The LOQ was calculated using the formula involving standard deviation of response and slope of calibration curve and it was found to be 0.1834µg/ml (Table no.14).

Robustness
Robustness was performed by injecting the citalopram HBr standard solution in to the HPLC by altering the flow rate, changing the pH and changing the composition of the organic solvent from the normal chromatographic conditions. The results are tabulated in (Table no.13)

3. Degradation Studies by HPLC [14-15]
Acidic degradation
In 4 different 10 ml volumetric flask 5ml of working standard solution (100 ppm) was mixed with 2ml of 0.1N, 0.5N, 1N, 1.5N, 2N, 3N, 4N hydrochloric acid and kept for 5hours. After 5hours solution was neutralized with sodium hydroxide then solution was diluted to 10 ml acetonitrile: water (75:25 V/V) and injected in stabilized chromatographic conditions. Under this condition, degradation was observed at 4N hydrochloric acid. (Table no.15)

Alkaline degradation
In 5 different 10 ml volumetric flask 5ml of working standard solution (100 ppm) was mixed with 2ml of 1N, 1.5N, 2N, 2.5N, 3N sodium hydroxide respectively and kept for 3 hours. After 3 hours solution was neutralized with dilute hydrochloric acid then solution was diluted to 10 ml with acetonitrile:water (75:25) and injected & degradation was observed at 1N sodium hydroxide.(Table no.15)

Oxidative degradation
5ml of working standard solution was mixed with 3ml 30% hydrogen peroxide respectively .The solution was diluted to 10ml with acetonitrile: water (75:25 V/V) and kept at room temperature for 3 hours. The solution was injected in stabilized chromatographic conditions. The degradation was found at 30% hydrogen peroxide. (Table no.15)

Thermal degradation
Thermal studies were performed by keeping drug sample in oven (100°C) for a period of 48 hours. 10 mg of exposed drug was weighed accurately after every 1 hour and transferred to a 100 ml of volumetric flask and dissolved in acetonitrile:water (75:25 V/V), the volume was made up with acetonitrile: water (75:25) to get concentration of 100µg/ml.(Table no.15) 5 ml standard stock solution of citalopram HBr was then diluted in 10 ml acetonitrile: water (75:25 V/V) to get working standard solution 50 µg/ml. The solution then injected in stabilized chromatographic conditions.

Photo-degradation
The photochemical stability of the drug was studied by exposing the drug sample to UV light for 72 hours 10 mg after exposure, accurately weighed 10mg of drug in 100 ml of distilled water to get concentration 100 µg/ml. 5ml standard stock solution of citalopram was then diluted in acetonitrile: water (75:25 V/V) 10 ml to get working standard solution 50 µg/ml and was then injected in stabilized chromatographic conditions. (Table no.15)

RESULTS AND DISCUSSION

The developed method was found to be precise as the % RSD values for intraday and inter-day were found to be less than 2%. Good recoveries (99% to 100.42%) of the drug were obtained at each added concentration, indicating that the method was accurate. The method was also found to be specific indicated by the % recoveries ranging from 99.45% to 99.98%. The LOD and LOQ were found to be in sub-microgram level indicating the sensitivity of the method. The method was also found to be robust and rugged as indicated by the %RSD values which are less than 2%. The summary of validation parameters of proposed spectrophotometric method is shown in Table no.1 and 7. The stress degradation studies showed that citalopram HBr undergoes degradation in acidic, oxidative, alkaline and photo, thermal condition (17.45%, 26.75%, 33.7%, 0.21%, 0.15%) respectively.

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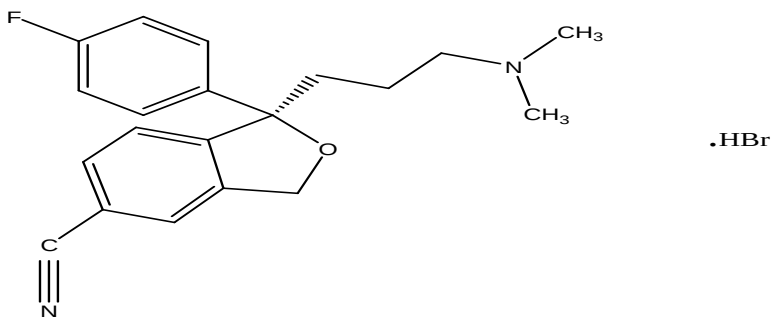

Figure no. 1: Structure of Citalopram HBr

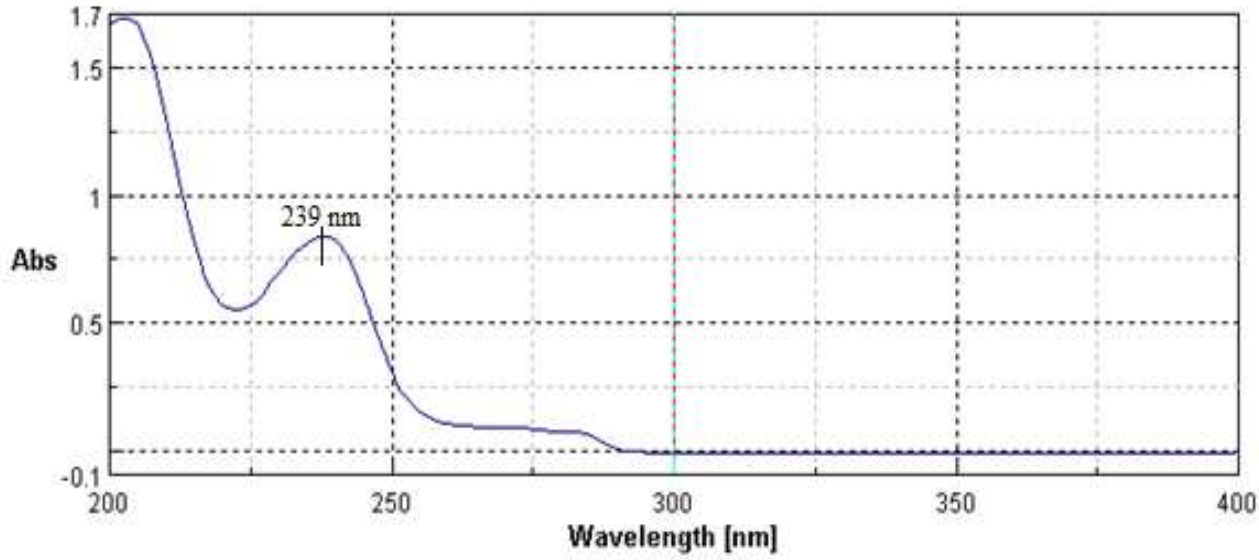

Figure no. 2: λ max of Citalopram HBr

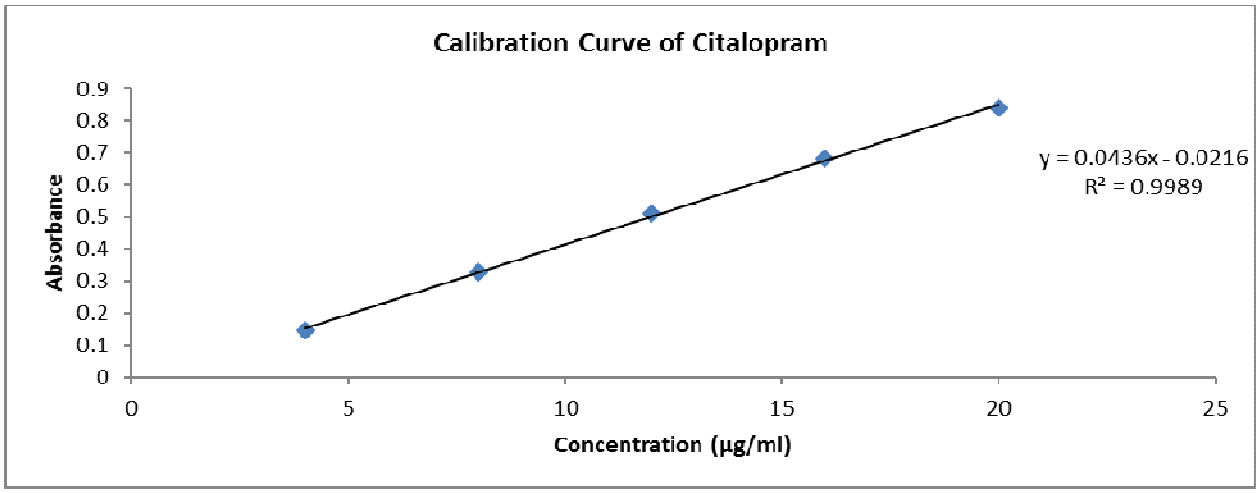

Figure no.3: Calibration curve for citalopram HBr

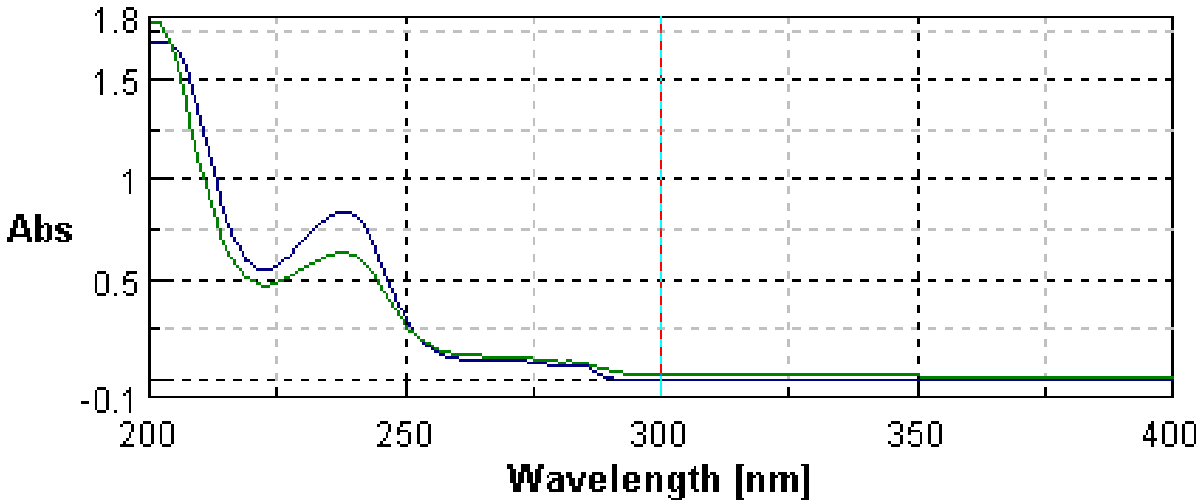

Figure no. 4: Comparison between standard Citalopram (20µg/ml) and acid degraded sample of Citalopram (20µg/ml). After 3 hours Drug got degraded and its λ max shifted from 238 nm to 236 nm

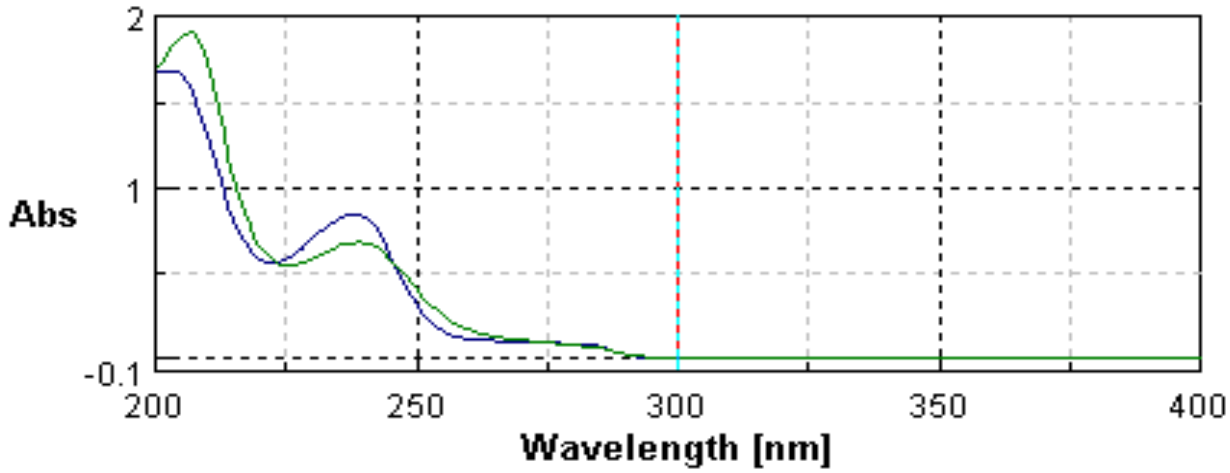

Figure 5: Comparison between standard Citalopram HBr (20µg/ml) and alkali degraded sample of Citalopram (20µg/ml). After 1 hours Drug got degraded and its λ max shifted from 238 nm to 235 nm

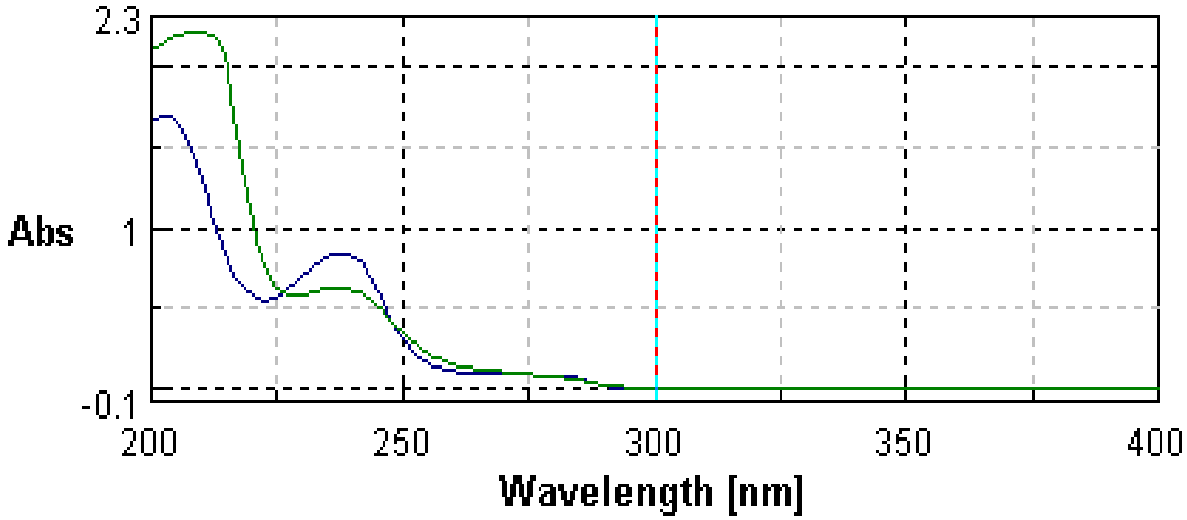

Figure 6: Comparison between standard Citalopram (20µg/ml) and oxidative degraded sample of Citalopram HBr (20µg/ml). After 3 hours Drug got degraded and its λ max shifted from 238 nm to 218 nm

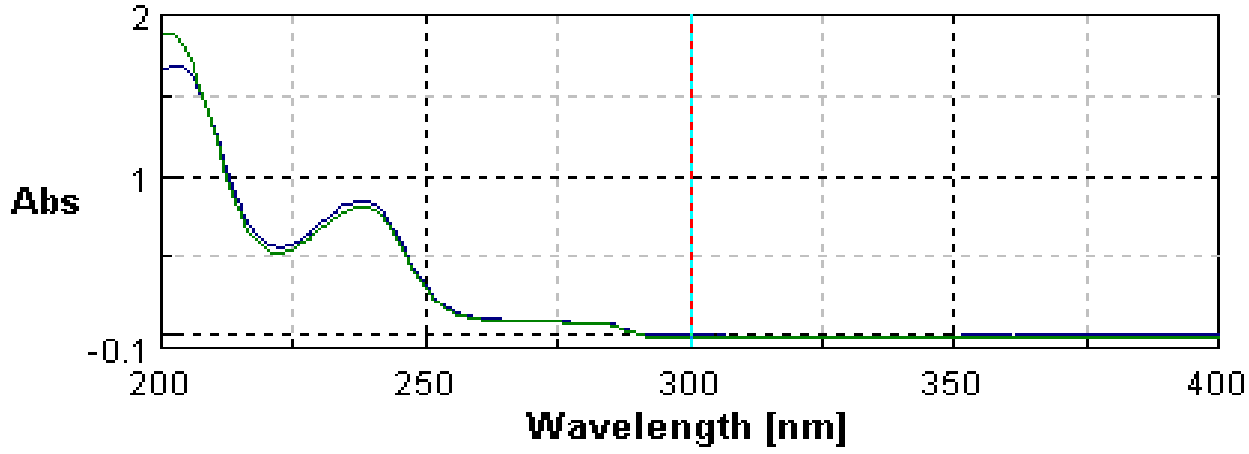

Figure 7: Comparison between standard Citalopram (20µg/ml) and Thermal degraded sample of Citalopram (20µg/ml).

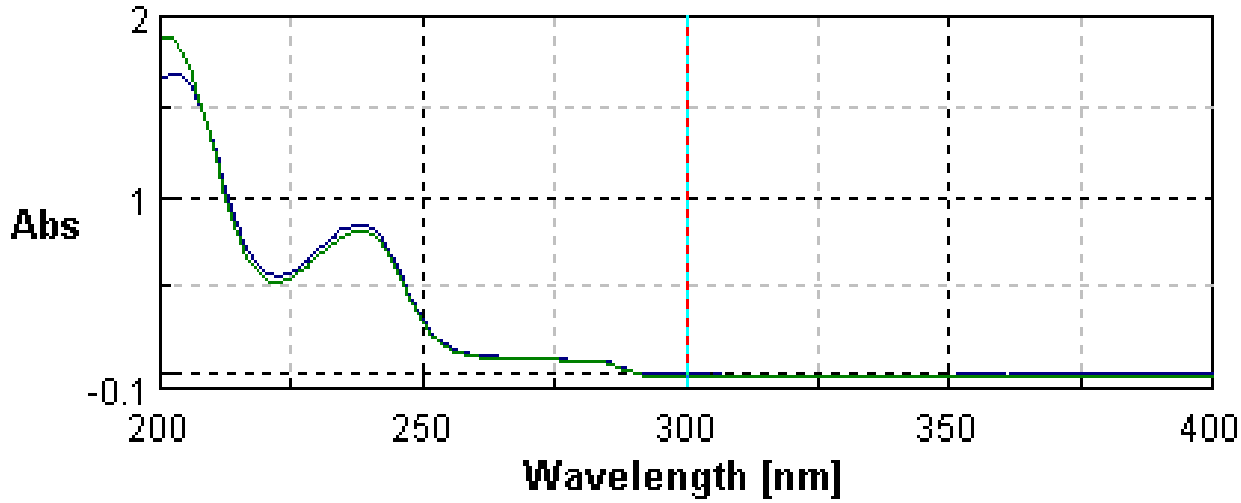

Figure 8: Comparison between standard Citalopram HBr (20µg/ml) and Photo degraded sample of Citalopram (20µg/ml). After 72 hours Drug got degraded and its λ max shifted from 238 nm to 234 nm

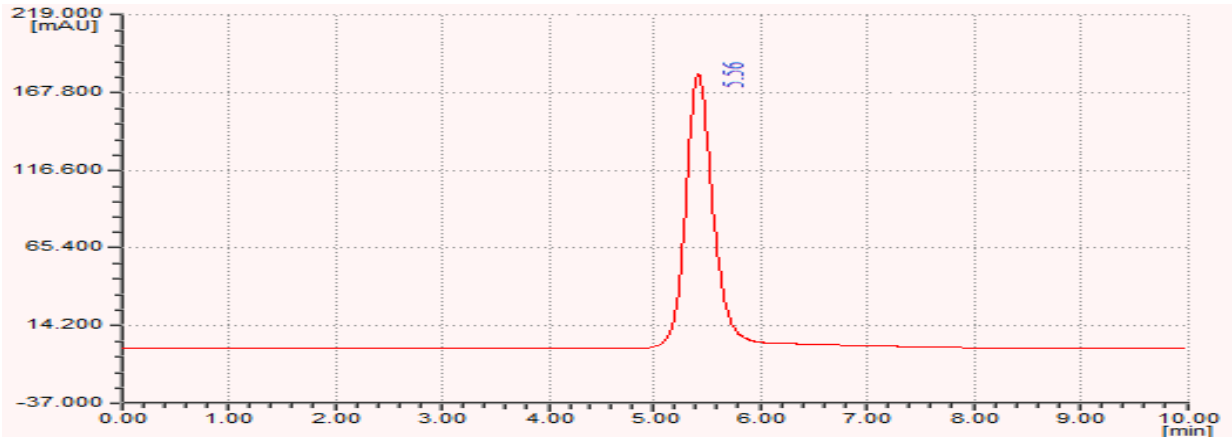

Figure no.9: Chromatogram of Citalopram HBr (50µg/ml, RT= 5.56) acetonitrile: water (75:25)

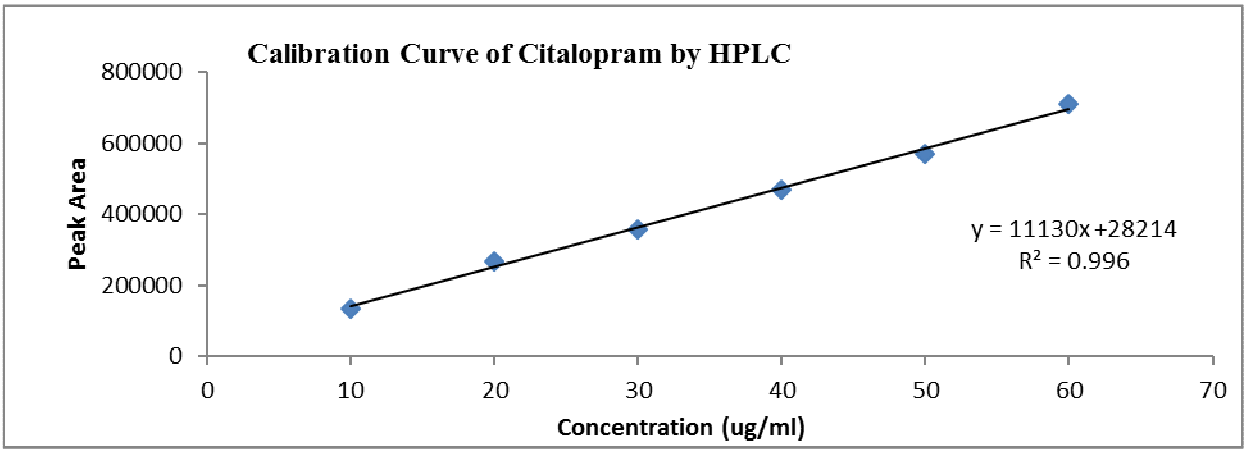

Figure no.10: Calibration curve of Citalopram HBr by HPLC

Table no. 1: optical characteristics

Parameters	Results
Beer's law limit (µg/ml)	4-20 (µg/ml)
Correlation Coefficient	0.998
Regression Equation (Y*)	0.043x-0.021
Slope(a)	0.043
Intercept(b)	0.021

Table no. 2: Accuracy reading of Citalopram HBr

Level	Concentration Formulation	Pure Drug	% Recovery	Mean
80%	10	8	99.50	99.45
	10	8	99.15	
	10	8	99.69	
100 %	10	8	99.87	99.90
	10	8	100.12	
	10	8	99.70	
120%	10	8	100.103	99.98
	10	8	99.85	
	10	8	100	

Table 3: Inter-day Precision of Citalopram HBr

Concentration (µg/ml)	Absorbance(nm)			Mean	Standard Deviation	% Relative standard deviation	Average of % RSD
	Day 1	Day 2	Day 3				
12	0.50	0.51	0.51	0.51	0.0058	1.14	0.91
16	0.68	0.67	0.67	0.67	0.0058	0.87	
18	0.82	0.82	0.83	0.82	0.0058	0.71	

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no. 4: Intraday Precision of Citalopram HBr

Concentration µg/ml	Absorbance(nm)			Mean	Standard Deviation	% Relative Standard deviation	Average of %RSD
	1	2	3				
12	0.50	0.51	0.51	0.51	0.0058	1.14	0.56
16	0.68	0.67	0.68	0.68	0.0058	0.85	
18	0.83	0.83	0.82	0.83	0.0058	0.70	

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no. 5: Robustness study of Citalopram HBr

Sr. No.	Concentration (ppm)	Absorbance		Average % Relative standard deviation
		At Room Temp.	At 18ºC	
1	12	0.5124	0.5113	0.235
2	12	0.5115	0.5118	
3	12	0.5112	0.5132	
4	12	0.5132	0.5141	
5	12	0.5142	0.5114	
6	12	0.5142	0.5122	
7	Mean	0.5128	0.5123	
8	Standard Deviation	0.001303	0.001106	
9	% Relative Standard deviation	0.2541	0.2159	

Average of six determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table 6: Ruggedness study of Citalopram HBr

Sr. No.	Concentration (ppm)	Absorbance		Average % Relative Standard deviation
		At Jasco	At Shimadzu	
1	12	0.5124	0.5142	0.5651
2	12	0.5120	0.5122	
3	12	0.5092	0.5085	
4	12	0.5145	0.5165	
5	12	0.5112	0.5185	
6	12	0.5095	0.5180	
7	Mean	0.5115	0.5147	
8	Standard deviation	0.001971	0.003834	
9	% Relative standard deviation	0.3853	0.7449	

Average of six determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no. 7: Summary of validation of Citalopram HBr

Sr.no	Parameters	Result
1	Linearity Indicated by Correlation Coefficient	0.998
2	Linear Regression Equation	0.043x-0.021
3	Range	4-20ug/ml
4	Interday Precision (% RSD)	0.91
5	Intraday Precision (%RSD)	0.56
6	Limit of detection	0.08662 µg/ml
7	Limit of Quantification	0.2625 µg/ml
8	Robustness indicated by % RSD	0.46535
9	Rugedness indicated by % RSD	0.5651

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no. 8: Summary of results of stress degradation studies

Stress condition	Time	Condition	Observation	% Degradation
Acidic Degradation	**3 Hours**	5 N HCl	λmax Shifted	**17.45%**
Alkali Degradation	**1 Hours**	1 N NaOH	λmax Shifted	**33.07%**
Oxidative Degradation	**3 Hours**	30 % H_2O_2	λmax Shifted	**26.75 %**
Thermal Degradation	**48 Hours**	Oven	λmax was not Shifted	**0.15 %**
Photo Degradation	**72 Hours**	UV Light	λmax was not Shifted	**0.21 %**

Table no.9: Optical Characteristics

Parameters	Results
Beer's law limit (µg/ml)	4-20 (µg/ml)
Correlation Coefficient	0.998
Regression Equation (Y*)	0.043x-0.021
Slope(a)	0.043
Intercept(b)	0.021

Table no.10: Intra-day precision studies for Citalopram HBr bulk drug

Conc. (ng/ml)	Peak area			Mean	SD	% RSD
	Trial 1	Trial 2	Trial 3			
30	355921	355985	355745	355883	124.28	0.034
40	469865	469654	469652	469723	122.40	0.026
50	570012	570155	570726	570297	39.80	0.0069
Average of % RSD = 0.0223						

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no.11: Interday precision studies for Citalopram HBr bulk drug

Conc.(ng/ml)	Peak area			Mean	SD	% RSD
	Trial 1	Trial 2	Trial 3			
30	355921	355815	355564	355766	183.34	0.052
40	469865	469715	469682	469754	97.53	0.021
50	570012	570065	570045	570040	26.76	0.0047
Average of % RSD =0.0259						

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no.12: Recovery studies of Citalopram HBr bulk drug

Level	Conc.(µg/ml) Std Sample	Area	Mean	Recovered Conc.(µg/ml)	% Recovery
80	25 + 20	475637 475590 475450	475559	40.19	98.45
100	25 + 25	476684 475678 478809	477057	40.33	99.56
120	25 + 30	485564 485581 485583	485564	41.09	100.5

Table no.13: Robustness studies of Citalopram HBr bulk drug

Flow Rate	Tailing Factor	Peak area			Mean	SD	% RSD
		Trial 1	Trial 2	Trial 3			
1.3	1.73	576545	575645	574164	575451	1202.26	0.2089
1.4	1.65	575485	575644	576554	575894	576.29	0.1001
1.5	1.45	576545	578541	576549	577211	1151.24	0.1994
Average % RSD= 0.1694							

Mobile Phase Composition	Tailing factor	Peak area			Mean	SD	% RSD
		Trial 1	Trial 2	Trial 3			
75:25	1.45	576424	578546	574566	576512	1991.4	0.3454
70:30	1.89	575496	574568	576965	575676	1208.6	0.2098
80:20	1.93	575644	572458	574958	574353	1676.8	0.2919
Average % RSD= 0.2824							

pH of Mobile Phase	Tailing factor	Peak area			Mean	SD	% RSD
		Trial 1	Trial 2	Trial 3			
5.0	1.36	574589	576895	576499	575994	1233.05	0.2141
7.0	1.85	576648	575899	574555	575701	1060.50	0.1842
6.0	1.56	574589	575648	574959	575065	537.44	0.093
Average % RSD= 0.1638							

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no.14: Summary of validation studies of Citalopram HBr drug

Sr. No	Parameter	Result
1	Linearity indicated by correlation coefficient	0.996
2	Linear regression equation	11130x+28214
3	Range	10 – 60µg/ml
4	Interday Precision (%RSD)	0.0259
5	Intraday Precision (%RSD)	0.0223
6	Limit of Detection	0.06052 µg/ml
7	Limit of Quantification	0.1834 µg/ml
8	Robustness indicated by % RSD	0.2089,0.1001,0.1994

Average of three determinations, S.D. is Standard deviation; RSD is the Relative Standard deviation.

Table no.15: Summary of stress degradation studies of Citalopram HBr bulk drug

Sr. No.	Stress degradation Parameter (50 ppm)	Change in Peak area	Condition	Concentration of degraded(µg/ml)	% degradation
1	Standard	570012		00	00
2	Acid degradation	496456	4 N HCl	6.61	13.22%
3	Alkali degradation	396345	1N NaOH	15.6	31.2%
4	Oxidative degradation	443456	30% H_2O_2	11.37	22.74%
5	Thermal degradation	526515	Oven	0.32	0.64%
6	Photo degradation	502355	UV Light	0.22	0.44%

CONCLUSION

All these factors lead to the conclusion that the proposed method is accurate, precise, simple, sensitive, rugged and rapid and can be applied successfully for the stability study of citalopram HBr in pharmaceutical formulations without interference and with good sensitivity; hence it can be used for the routine analysis in quality control department.

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