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DEVELOPMENT AND VALIDATION OF UV-VISIBLE SPECTROMETRY METHOD FOR METOPROLOL SUCCINATEIN MODIFIED RELEASE PELLET AND IN MUPS TABLETS

SHINDE MB[*1], SHINDE G[2], PATEL R[1] AND DHARMASI A[1]

1: Department of Pharmaceutics, Parul Institute of Pharmacy, Parul University, Vadodara, Gujarat, India

2: Mil Laboratories Pvt Ltd, Vadodara, Gujarat, India

***Corresponding Author: Mukesh B. Shinde: E Mail: mukeshbshinde@gmail.com**

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ABSTRACT

Objective: A simple, precise, accurate and cost effective UV-visible spectrometry method and validation for water soluble Metoprolol succinate in the modified release pellet formulation and MUPS tablets.

Method: Analytical method development and validation was done as per ICH Q2 (R1) guideline

Results: The absorption maxima of Metoprolol succinate was found to be 274nm for all three solvents. The method represents correlation coefficient (R2 =0.999) at concentration range of 50 to 160 µg/ml. The validation of method was done and the percent relative standard deviation of inter-day and intra-day precision range for 0.1N HCl was (0.24-1.83) & (0.0.91-1.84);for pH 4.5 Acetate buffer was (0.12-1.99) & (0.17-1.89) and for pH 6.8 phosphate buffer was (1.00-1.95) & (0.49-1.92) respectively, which shows the method was precise. The recovery of Metoprolol succinate was found to be 97.08 - 99.03 %.

Conclusion: This validated precise UV- visible spectrometry method can be applied for the estimation of Metoprolol succinate with respect to the determination of assay content for the solid dosage form.

Keywords: Metoprolol succinate, Modified release pellets, Validation, MUPS

INTRODUCTION

Metoprolol succinate (MS), is a β_1- selective adrenoceptor blockingagent, for oral rout of administration. MS chemically is (±) 1 (isopropylamino)-3- [p-(2-methoxyethyl) phenoxy] - 2-propanol succinate (2:1) salt. Its empirical formula is $C_{34}H_{56}N_2O_{10}$and structural formula is shown in **Figure 1**.

MS is a white crystalline powder with a molecular weight of 652.8 g/mol. It is freely soluble in water; soluble in methanol; sparingly soluble in ethanol; slightly soluble in dichloromethane and -2 -propanol; practically insoluble in ethyl-acetate, acetone, diethylether and heptanes **[1-2]**. Some of the physical and biopharmaceutical properties of MS are given in **Table 1**.

Instrumental analysis is very sensitive and accurate measure of estimation of drug content drug release. Hence, UV spectrophotometry, one of the simplest instrumentation methods capable of drug estimation, was used in the present study. Literature survey reveals several analytical methods for the determination of Metoprolol succinate (MS) in pharmaceutical dosage forms and in biological fluids by LC **[2]**, UV **[3-4]**, HPLC **[5]** and LC-MS/MS **[6]**. The main aim of the present study was to develop and validate a simple and accurate UV method for the assay of MS its pellet and in MUPS tablets dosage form with less aggressive solvent system.

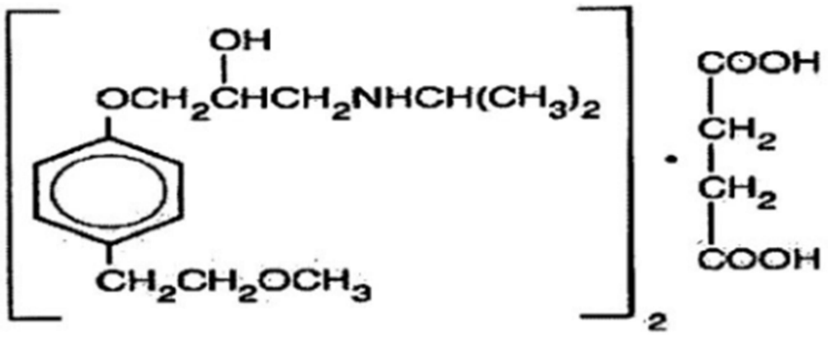

Figure 1: Molecular Structure of Metoprolol succinate

Table 1: Physicochemical properties of MS

Parameter	Observations
Solubility	Freely soluble in water
pH	Between 7.0 and 7.6
Melting point	136-1380C
Volume of Distribution	5.6 L/kg
t½	3 to 7 hours
pKa	9.5
Log P	1.6

MATERIALS AND METHOD

Materials

Metoprolol succinate (MS) was obtained as gift sample from Alembic Pharm, India. All chemicals were used was analytical grade of Merck.

Instruments Used

A double beam UV-visible spectrophotometer (UV-1800 Jasco) with 1 cm path length was used for spectral measurements with 3 cm matched quartz cells. Digital top pan balance (sensitivity- 0.1 mg) Sartorius balance was used for weighing.

Preparation of standard stock solution

Stock solutions were prepared in all the selected solvents. Accurately weighed quantities (100 mg) of drug were transferred to 100 Ml calibrated volumetric flasks and dissolved in different solvents. The volumes were made up to 100 mL with the same solvents. The resulting solutions (1000 µg/mL) were further diluted ten times with the same solvents to get stock solutions (100 µg/mL).(Stock Solution I)

Preparation of standard solutions

From stock solution I aliquots of the stock solutions of MS were transferred to calibrated volumetric flasks and diluted up to the mark with respective solvents to obtain known final concentrations ranging from 10 to 50 µg/mL.

Determination of maximum wavelength (ʎmax)

The spectrum scan of standard solution was recorded using UV Visible spectrophotometer for 200 to 400 nm wavelength range against respective solvent as blank. The wavelengths with maximum absorbance (λmax) were selected as analytical wavelengths.

Preparation of calibration curve

Absorbances of standard solutions were recorded at selected analytical wavelengths and the calibration curves were plotted between standard drug concentrations and observed absorbance.

Estimation of MS in Modified release pellets

An accurately weighed quantity of coated pellets equivalent to 50mg of Metoprolol succinate. Pellets was crushed and finely powdered in mortar pestle and transferred to 100 mL volumetric flask and10mL methanol was added and volumetric flask was shaken for 15 min on cyclomixer or dissolved by sonication with sufficient quantity of phosphate buffer 6.8, volume was made upto mark. A 1 mL portion of the filtrate was further diluted with phosphate buffer 6.8 in a 10 ml volumetric flask upto mark (10µg/mL)

on label claim basis. The absorbance of the resulting solution was measured at 274 nm against solvent blank.

Estimation of MS in Pellet in MUPS tablets formulation

Metoprolol succinate MUPS Twenty tablets were weighed and average weight was determined. Tablets was crushed and finely powdered in mortar pestle. An accurately weighed quantity of tablet powder equivalent to 50mg of Metoprolol succinate was transferred to 100 mL volumetric flask and10mL methanol was added and volumetric flask was shaken for 15 min on cyclomixer or dissolved by sonication with sufficient quantity of phosphate buffer 6.8, volume was made upto mark. A 1 mL portion of the filtrate was further diluted with phosphate buffer 6.8 in a 10 ml volumetric flask upto mark (10µg/mL) on label claim basis. The absorbance of the resulting solution was measured at 274 nm against blank.

VALIDATION OF UV SPECTROPHOTOMETRIC METHODS

Developed UV method for estimation of MS was validated as per ICH guideline for evaluating different parameters like Linearity, Accuracy, Precision, Stability, Sensitivity, Limit of detection (LOD) and limit of quantification (LOQ) **[8-10]**.

Linearity

Linearity of an analytical method is the ability to elicit the test results that are directly orby well-defined transformation proportional to the concentration of the analyte in the samples within the given range **[11]**.

Accuracy

The accuracy of an analytical method is the closeness of test results obtained by that method to the true value **[12]**. The accuracy of the method was determined by calculating the recoveries of the analyte by the method of standard additions different levels of drug concentrations. Known amounts of standard drug (80%, 100% and 120%) were added to the pre-analyzed samples and the absorbance was measured. Accuracy is assessed as the mean % recovery of the added pure drug which was calculated using following equation

$$\% \mathbf{RC} = \frac{\mathbf{SPS} - \mathbf{S}}{\mathbf{SP}} \times \mathbf{100}$$

Where, % RC= Percent recovery; SPS= Amount found in the spiked sample

S= Amount found in the sample; SP= Amount added to the sample

Precision

Precision is a measure of the consistency and reproducibility of a method. The precise analytical method is the degree of agreement among the individual test results gives very

close values for repeated measurement of same sample when the procedure is applied repeatedly to multiple sampling of homogeneous sample **[13]**. Multiple measurements for same standard concentrations were made on same day as well as on three consecutive days to determine intraday and interday precision, respectively. The % Relative Standard Deviation (% RSD) was calculated as a measure of precision.

Sensitivity

Limit of detection (LOD) and limit of quantification (LOQ are used to describe the smallest concentration of an analyte that can be reliably measured by an analytical procedure. So, LOD and LOQ of developed methods were determined using following equations **[13]**,

$$\mathbf{LOD} = \frac{\mathbf{3.3 \times \sigma}}{\mathbf{S}}$$

Where, σ = standard deviation of Y- intercepts, S=Slope

$$\mathbf{LOQ} = \frac{\mathbf{10 \times \sigma}}{\mathbf{S}}$$

Where, σ= standard deviation of Y- intercepts, S=Slope

RESULTS AND DISCUSSION

Determination of maximum wavelength

The maximum wavelength (ʎmax) was evaluated by scanning the full spectrum in the range of 200 nm-800 nm UV-visible ranges **(Figure 2)**. The maximum wavelength (ʎmax) was found 274 nm at three different solvent.

Preparation of calibration curve

The calibration curve of MS was performed in three different solvents and graph plotted concentration vs. absorbance **(Figure 3)**. The absorbance values of different concentration were noted **(Table 2)**.

Estimation of Metoprolol succinate (MS)in Pellet formulation

Assay study of pellet formulation containing metoprolol succinate (MS) was carried out and the percent (%) assay was found to be 97.9 ± 0.2.

Estimation of Metoprolol succinate (MS) in MUPS tablets formulation

Assay study of MUPS Tablets formulation containing metoprolol succinate (MS) was carried out and the percent (%) assay was found to be 100.1 ± 0.2.

VALIDATION OF UV SPECTRO-PHOTOMETRIC METHODS

Linearity

For the linearity of the Metoprolol succinate(MS), twelve point calibrations curve were plotted in a concentration range of 50-160 (µg/ml). From the linearity study it was observed that the drug was found to be linear in the concentration range and the linear regression equation with correlation

coefficient elaborated in **Table 3 and Figure 2**.

Accuracy

Accuracy of the proposed UV method was verified by conducting the recovery studies by using standard addition method. Standard drug concentration at three different percent levels was added to known amount of MS taken from pellets. The percent recovery of added standards was calculated **(Table 3)**. The results showed better % mean recovery for respective percent levels. The % mean recovery values are closer to 100% showed high accuracy of the proposed UV analytical method.

Precision

Intra-day and inter-day precision study of drug were evaluated for the 50µg/ml to 160 µg/ml in three solvents. Absorbance mean, percent assay and percent RSD were calculated for the intra-day as well as inter-day precision study **(Table 5, 6 and 7)**. The RSD values obtained for the analytical methods were within the acceptable range (< 2%) indicating that these methods are precise.

Stability

The stability of the drug in all the three solvents was ascertained over the period of 24h by measuring absorbance of the solution at initial (0hrs) and 24 hrs. Details of results mention in **Table 8, 9 and Figure 4**.

Sensitivity

Limit of Detection (LOD) & Limit of Quantification (LOQ)

LOQ are used to describe the smallest concentration of an analyte that can be reliably measured by an analytical procedure. So, LOD and LOQ of developed methods were determined using following equations **(Table 10)**.

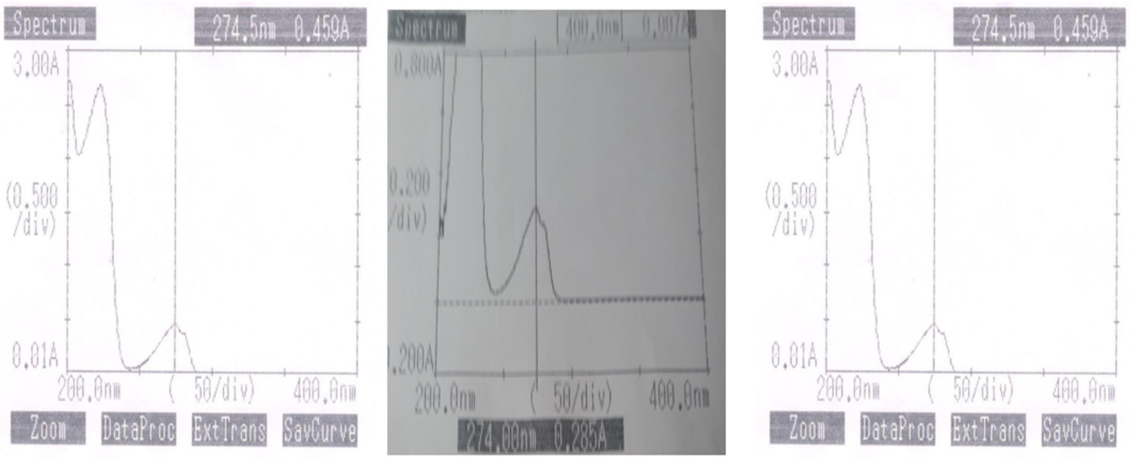

Figure 2: The maximum wavelength (λmax) at different solvents

Table 2: The absorbance values of different concentration in three different solvent

Concentration (µg/mL)	Absorbance		
	In 0.1N HCl	In Acetate buffer pH 4.5	In phosphate buffer pH 6.8
50	0.218	0.229	0.218
60	0.263	0.274	0.267
70	0.312	0.319	0.31
80	0.358	0.365	0.365
90	0.395	0.414	0.399
100	0.436	0.458	0.448
110	0.475	0.507	0.508
120	0.516	0.55	0.549
130	0.557	0.601	0.598
140	0.604	0.652	0.64
150	0.651	0.699	0.695
160	0.709	0.741	0.739

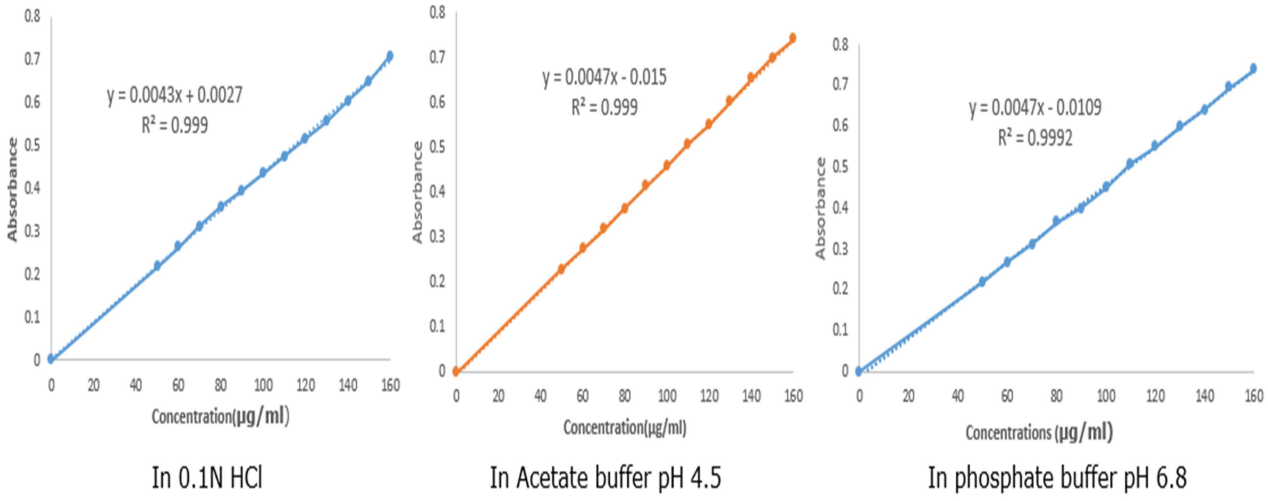

Figure 3: Calibration curve of metoprolol succinate in different solvents

Table 3: Linear regression analysis of calibration data

Solvents	Calibration range (µg/mL)	Regression equation	Correlation coefficient (R2)
In 0.1N HCl	50 to 160	Y=0.043x + 0.0027	0.999
In Acetate buffer pH 4.5	50 to 160	Y=0.0047x - 0.015	0.999
In phosphate buffer pH 6.8	50 to 160	Y=0.047x - 0.0109	0.999

Table 4: Standard addition data to measure accuracy of UV method in different solvents

Solvents	Spiking	Drug in solution (µg/mL)	Spiked drug (µg/mL)	Total drug found# (µg/mL)	% Analytical Recovery#
0.1N Hydrochloric acid	80%	25	20	43.88	97.51
	100%	25	25	48.78	97.56
	120%	25	30	54.69	99.43
Phosphate buffer pH 4.5	80%	25	20	43.98	97.73
	100%	25	25	48.98	97.96
	120%	25	30	54.47	99.03
Phosphate buffer pH 6.8	80%	25	20	43.69	97.08
	100%	25	25	48.92	97.84
	120%	25	30	53.87	97.94

Table 5: Intra-day and inter-day precision study in 0.1N Hydrochloride Acid

Concentration (µg/mL)	Intraday (Absorbance)			% RSD	Inter day (Absorbance)			% RSD
	Set I	Set II	Set III		Day 1	Day 2	Day 3	
50	0.213	0.211	0.218	1.68	0.201	0.196	0.2	1.32
60	0.253	0.246	0.249	1.40	0.256	0.253	0.26	1.37
70	0.3	0.308	0.299	1.63	0.298	0.309	0.302	1.83
80	0.347	0.337	0.344	1.49	0.341	0.342	0.342	0.16
90	0.386	0.379	0.383	0.91	0.379	0.372	0.374	0.96
100	0.423	0.433	0.418	1.79	0.428	0.434	0.436	0.96
110	0.465	0.459	0.476	1.84	0.461	0.463	0.463	0.24
120	0.512	0.509	0.525	1.65	0.519	0.511	0.514	0.78
130	0.578	0.583	0.592	1.21	0.55	0.55	0.552	0.20
140	0.621	0.598	0.612	1.89	0.596	0.602	0.605	0.76
150	0.655	0.675	0.676	1.77	0.621	0.607	0.608	1.27
160	0.762	0.767	0.787	1.71	0.639	0.641	0.643	0.31

Table 6: Intra-day and inter-day precision study in Acetate buffer pH 4.5

Concentration (µg/mL)	Intraday (Absorbance)			% RSD	Inter day (Absorbance)			% RSD
	Set I	Set II	Set III		Day 1	Day 2	Day 3	
50	0.219	0.217	0.225	1.89	0.219	0.225	0.226	1.70
60	0.259	0.26	0.261	0.38	0.265	0.261	0.262	0.79
70	0.3	0.299	0.298	0.33	0.303	0.298	0.3	0.84
80	0.349	0.349	0.348	0.17	0.359	0.348	0.355	1.57
90	0.387	0.397	0.4	1.72	0.393	0.402	0.398	1.13
100	0.431	0.436	0.435	0.61	0.448	0.438	0.436	1.46
110	0.477	0.482	0.477	0.60	0.451	0.469	0.463	1.99
120	0.525	0.511	0.525	1.55	0.478	0.477	0.478	0.12
130	0.557	0.551	0.561	0.90	0.549	0.571	0.562	1.97
140	0.602	0.59	0.6	1.08	0.604	0.611	0.597	1.16
150	0.644	0.632	0.641	0.98	0.66	0.641	0.643	1.61
160	0.689	0.677	0.689	1.01	0.703	0.689	0.684	1.42

Table 7: Intra-day and inter-day precision study in phosphate buffer pH 6.8

Concentration (µg/mL)	Intraday (Absorbance)			% RSD	Inter day (Absorbance)			% RSD
	Set I	Set II	Set III		Day 1	Day 2	Day 3	
50	0.227	0.219	0.224	1.81	0.246	0.238	0.24	1.73
60	0.247	0.247	0.243	0.94	0.249	0.258	0.25	1.95
70	0.298	0.308	0.298	1.92	0.328	0.335	0.33	1.09
80	0.341	0.349	0.339	1.54	0.349	0.356	0.352	1.00
90	0.373	0.375	0.37	0.68	0.385	0.395	0.386	1.42
100	0.426	0.432	0.424	0.97	0.432	0.429	0.443	1.70
110	0.472	0.472	0.468	0.49	0.484	0.5	0.499	1.81
120	0.508	0.52	0.504	1.63	0.538	0.548	0.551	1.25
130	0.543	0.53	0.55	1.88	0.569	0.55	0.56	1.70
140	0.595	0.604	0.59	1.19	0.614	0.627	0.637	1.84
150	0.629	0.644	0.624	1.65	0.659	0.653	0.674	1.63
160	0.663	0.683	0.659	1.92	0.693	0.717	0.714	1.85

Table 8: Stability study

Concentration (µg/mL)	Absorbance in 0.1N HCl		Absorbance In Acetate buffer pH 4.5		Absorbance In phosphate buffer pH 6.8	
	Initial	After 24 Hrs	Initial	After 24 Hrs	Initial	After 24 Hrs
50	0.218	0.21	0.229	0.215	0.218	0.228
60	0.263	0.256	0.274	0.271	0.267	0.27
70	0.312	0.297	0.319	0.311	0.31	0.323
80	0.358	0.345	0.365	0.353	0.365	0.365
90	0.395	0.393	0.414	0.396	0.399	0.412
100	0.436	0.428	0.458	0.444	0.448	0.444
110	0.475	0.464	0.507	0.499	0.508	0.501
120	0.516	0.521	0.55	0.543	0.549	0.534
130	0.557	0.559	0.601	0.591	0.598	0.585
140	0.604	0.6	0.652	0.649	0.64	0.642
150	0.651	0.672	0.699	0.687	0.695	0.678
160	0.709	0.686	0.741	0.732	0.739	0.711

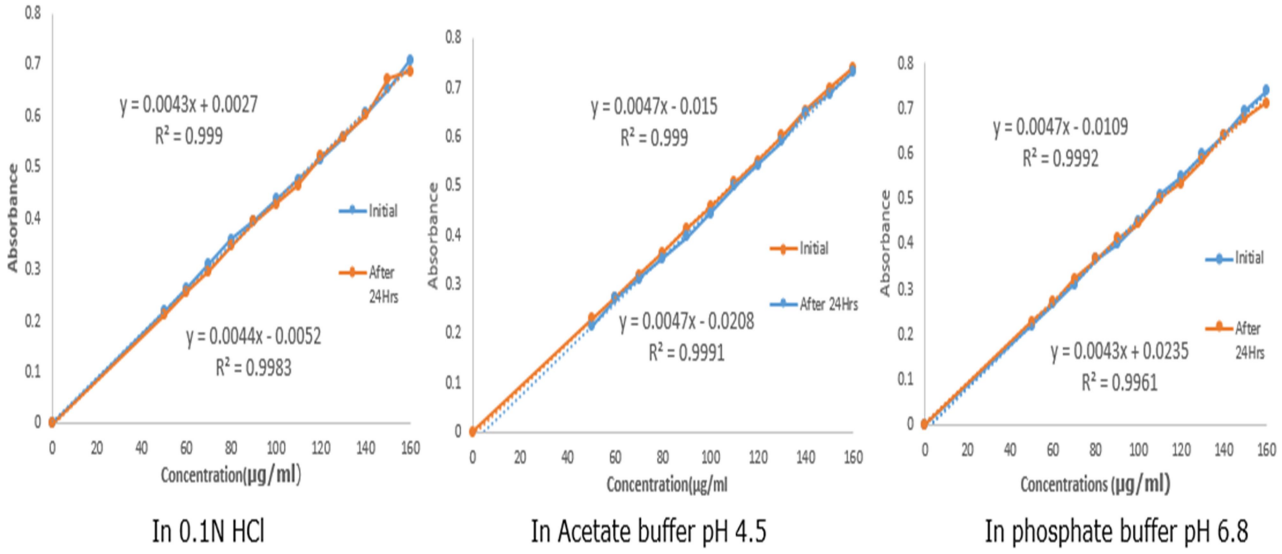

Figure 4: Calibration curve of different solvent at 0 hrs and 24hrs

Table 9: Linear regression analysis of calibration data

Solvents	Calibration range (µg/mL)	Regression equation	Correlation coefficient (R2)
In 0.1N HCl	50 to 160	Initial : Y=0.043x + 0.0027	0.999
		24 Hrs.: Y=0.0044x -0.0052	0.998
In Acetate buffer pH 4.5	50 to 160	Initial Y=0.0047x - 0.015	0.999
		24 Hrs.: Y= 0.0047x – 0.0208	0.0999
In phosphate buffer pH 6.8	50 to 160	Initial Y=0.047x - 0.0109	0.0999
		24 Hrs.: Y= 0.0043x +0.0235	0.996

Table 10: LOD and LOQ calculation from calibration data

Solvents	Slope of line	SD of line	LOD (µg/mL)	LOQ (µg/mL)
0.1N Hydrochloric acid	0.043	0.027	1.883	6.279
Acetate buffer pH 4.5	0.047	0.025	1.595	5.319
Phosphate buffer pH 6.8	0.047	0.022	1.402	4.680

CONCLUSION

A simple, precise and accurate UV-visible spectrophotometric method was developed for the estimation of MS in pellets as well as in MUPS tablets formulation.

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REFERENCES

[1] http://www. Drug Bank Metoprolol (DB00264). Htm 25,November, 2019

[2] United States Pharmacopeia. The Standard USA: US Pharmacopeia-42-National Formulary-37. 2019.

[3] Jadhav AS, Tarkase KN, Deshpande AP. Quantitative Determination of Metoprolol Succinate in bulk and tablet Dosage form through comparative study of UV and derivative Spectroscopy. Der Pharmacia Lettre. 2012; 4(3): 763-7.

[4] Li XN, Guo HX, Heinamaki J. Aqueous coating dispersion (pseudolatex) of zein improves formulation of sustained-release tablets containing very water-soluble drug. Journal of Colloid and Interface science. 2010 May 1; 345(1): 46-53.

[5] Marucci M, Andersson H, Hjärtstam J, Stevenson G, Baderstedt J, Stading M, Larsson A, von Corswant C. New insights on how to adjust the release profile from coated pellets by varying the molecular weight of ethyl cellulose in the coating film. International journal of pharmaceutics. 2013 Dec 15; 458(1): 218-23.

[6] Siddique S, Khanam J, Bigoniya P. Development of sustained release capsules containing "coated matrix granules of metoprolol tartrate". Aaps Pharmscitech. 2010 Sep; 11(3): 1306-14.

[7] Ravishankar H, Patil P, Samel A, Petereit HU, Lizio R, Iyer-Chavan J. Modulated release metoprolol succinate formulation based on ionic interactions: in vivo proof of concept. Journal of controlled release. 2006 Mar 10; 111(1-2): 65-72.

[8] I.E.W. Group, Validation of analytical procedures: Text and Methodology Q2(R1), ICH Secretariat, Geneva, 2005.

[9] Q2A: Text on; Validation of Analytical Procedures. In International Conference on Harmonization. Federal Register 1995; 60(40): 11260-11262.

[10] Q2B: Validation of Analytical Procedures: Methodology, Availability. In International Conference on Harmonization. Federal Register 1997; 62(96): 27463-27467.

[11] Hubert P. Validation of quantitative analytical procedure, harmonization of

approaches. STP Pharma Practices. 2003; 13: 101-38.

[12] Swartz ME, Krull IS. Handbook of Analytical Validation. Taylor & Francis, 2012; 140-14.

[13] Venugopal K, Saha RN. New, simple and validated UV-spectrophotometric methods for the estimation of gatifloxacin in bulk and formulations. Il Farmaco. 2005 Nov 1; 60(11-12): 906-12.