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DETERMINATION OF METOPROLOL IN PHARMACEUTICAL PREPARATIONS BY ZERO-, FIRST-, SECOND- AND THIRD-ORDER DERIVATIVE SPECTROPHOTOMETRIC METHOD

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ABSTRACT

In this study, zero-, first-, second- and third-order derivative spectrophotometry methods were developed for the determination of metoprolol in pharmaceutical preparations. In zero order spectrophotometry, absorbance values were measured at 276 nm in zero order spectra of solution of metoprolol in methanol in the range of 240-310 nm. In first derivative spectrophotometry, absorbance values were measured at 265, 278 and 285 nm. In second derivative spectrophotometry, absorbance values were measured at 276, 279, 287 and 282 nm. In third derivative spectrophotometry, absorbance values were measured at 275, 278 and 281 nm. Parameters such as linearity, precision, accuracy, specificity, stability, limit of detection and limit of quantitation were studied according to the International Conference on Harmonization Guidelines. All the methods developed were successfully applied to two tablet formulation and the results were compared statistically with each other.

KEYWORDS

Metoprolol, Zero-, First-, Second-, Third-order Derivative Spectrophotometric Method and Pharmaceutical Preparation

INTRODUCTION

β-blockers are clinically important drugs and are used in the treatment of disorders such as hypertension, angina pectoris and arrhythmia (1). Metoprolol (Figure 1) is a relatively selective β-1 adrenoceptor antogonist that has been used extensively for more than 25 years to treat such cardiovascular disorders as hypertension, arrhythmia and heart failure[1,2].

Several methods have been reported for determination of metoprolol including gas chromatography-mass spectrometry (GC-MS)[3-5], high-performance liquid chromatography (HPLC)[2,6-10], LC-MS[11-13], LC-MS-MS[14] and spectrophotometry[15].

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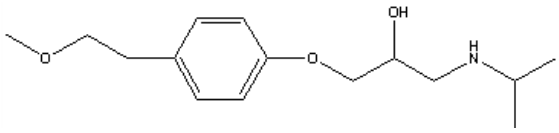

Figure 1. Metoprolol.

But, to our knowledge, there is no individual first-, second- and third-order derivative spectrophotometric method for the determination of metoprolol in pharmaceutical preparation in literature. Derivative spectrophotometry is an analytical technique of great utility for extracting both qualitative and quantitative information from spectra composed of unresolved bands, and for eliminating the effect of baseline shifts and baseline tilts. It consists of calculating and plotting one of the mathematical derivatives of a spectral curve[16]. In the last year, this technique has been rapidly gained its application in the analysis of pharmaceutical preparations.

We wanted to develop new spectrophotmetric methods for the determination of metoprolol in pharmaceutical preparation without the necessity of sample pre-treatment. After developing zero-, first-, second- and third-order derivative derivative spectrophotometric methods were also carried out and all optimization parameters were also considered. Also, the developed zero-, first-, second- and third-order derivative spectrophotometric methods were applied to commercial preparations (Problok and Beloc ZOK) as tablet. The results obtained by these four methods were statistically compared.

MATERIALS AND METHODS

(i) *Chemicals and reagents*:

Metoprolol tartrate was purchased from Sigma-Aldrich (St. Louis, MO, USA). Problok and Beloc ZOK tablets (100 mg metoprolol tartrate) were obtained Terra and Astrazeneca Pharmaceutical Industry (Istanbul, Turkey), respectively.

(ii) *Instrument:*

A Thermospectronic double-beam UV-Visible spectrophotometer (HEλIOSβ) with the local control software was used. Zero-, first-, second- and third-derivative spectra of reference and sample solutions were recorded in 1 cm quartz cells at a scan speed of 600 nm/min, a scan range of 240-310 nm and fixed slit width of 2 nm.

(iii) *Preparations of the standard and quality control solutions*:

The stock standard solution of metoprolol was prepared in methanol to a concentration of 100 µg/mL and kept stored at -20 °C in dark glass flasks. Working standard solutions were prepared from the stock standard solutions. A calibration graph was constructed in the range of 3, 5, 7.5, 10, 12.5, 15 and 20 µg/mL for metoprolol (n=6). For quality control samples containing concentration 4, 9, 17.5 µg/mL of metoprolol, the stock solution was diluted with methanol.

(iv) *Procedure for pharmaceutical preparations:*

The average tablet mass was calculated from the mass of 10 tablets of Problok and Beloc ZOK (100 mg metoprolol tartrate tablet, which was composed of metoprolol tartrate and some commen excipients). They were then finely ground, homogenized and portion of the powder was weighed accurately, transferred into a 50 mL brown measuring flask and diluted to scale with methanol. The mixture was sonicated for at least 20 min to aid dissolution and then filtered through a Whatman No 42 paper.

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Approximate dilutions were made at concentrations of 5 and 15 µg/mL with methanol. Zero-, first- second- and third-order derivative spectra were recorded against methanol.

(v) *Data analysis:*

All statistical calculations were performed with the Statistical Product and Service Solutions (SPSS) for Windows, version 10.0. Correlations were considered statistically significant if calculated P values were 0.05 or less.

RESULTS

1. Method development:

The derivative wavelength difference (Δλ) depends on the measuring wavelength range and n values (smoothing factor). Generally, the noise decreases by increasing Δλ. Optimal wavelength range should be chosen since the broad peaks become sharper, the ratio of signal/noise elevates and the sensitivity of the method increases by controlling the degree of low pass filtering or smoothing. Therefore, a series of *n* values (*n*=1-9) were tested in the first-, second- and third-order derivative spectra of metoprolol in methanol. Optimum results were obtained in the measuring wavelength range 240-310 nm, *n*=5 (Δλ=17.5 nm) for first-, second- and third-order derivative spectrophotometric methods.

Figure 2A presents the overlay of UV spectra of metoprolol in methanol gives two characteristic maxima at 276 and 282 nm. These two shouldered peaks were separated by using derivative spectrophotometer. Figures 2B-D presents the overlay of first-, second- and third-order ultraviolet spectra of metoprolol standard samples in methanol, respectively. As demonstrated in the Figure 2B, the spectra present characteristic a maximum and two minima. Maximum is represented at 265 and minima are shown at 278 and 285 nm. As demonstrated in the Figure 2C, the spectra present characteristic two minima and two maxima. Maxima are represented at 279 and 287 nm and minima are shown at 276 and 282 nm. As demonstrated in the Figure 2D, the spectra present characteristic two minima and a maximum. Maximum is represented at 278 nm and minima are shown at 275 and 281 nm.

As no difference was observed between spectra of metoprolol standard and tablet solutions and in the maxima and minima wavelengths of all spectra, it was suggested that the developed methods allowed complete elimination of the background absorption due to the tablet excipients at the chosen wavelengths both in zero-, first-, second- and third-order derivative spectra of metoprolol (Figures 2A-D).

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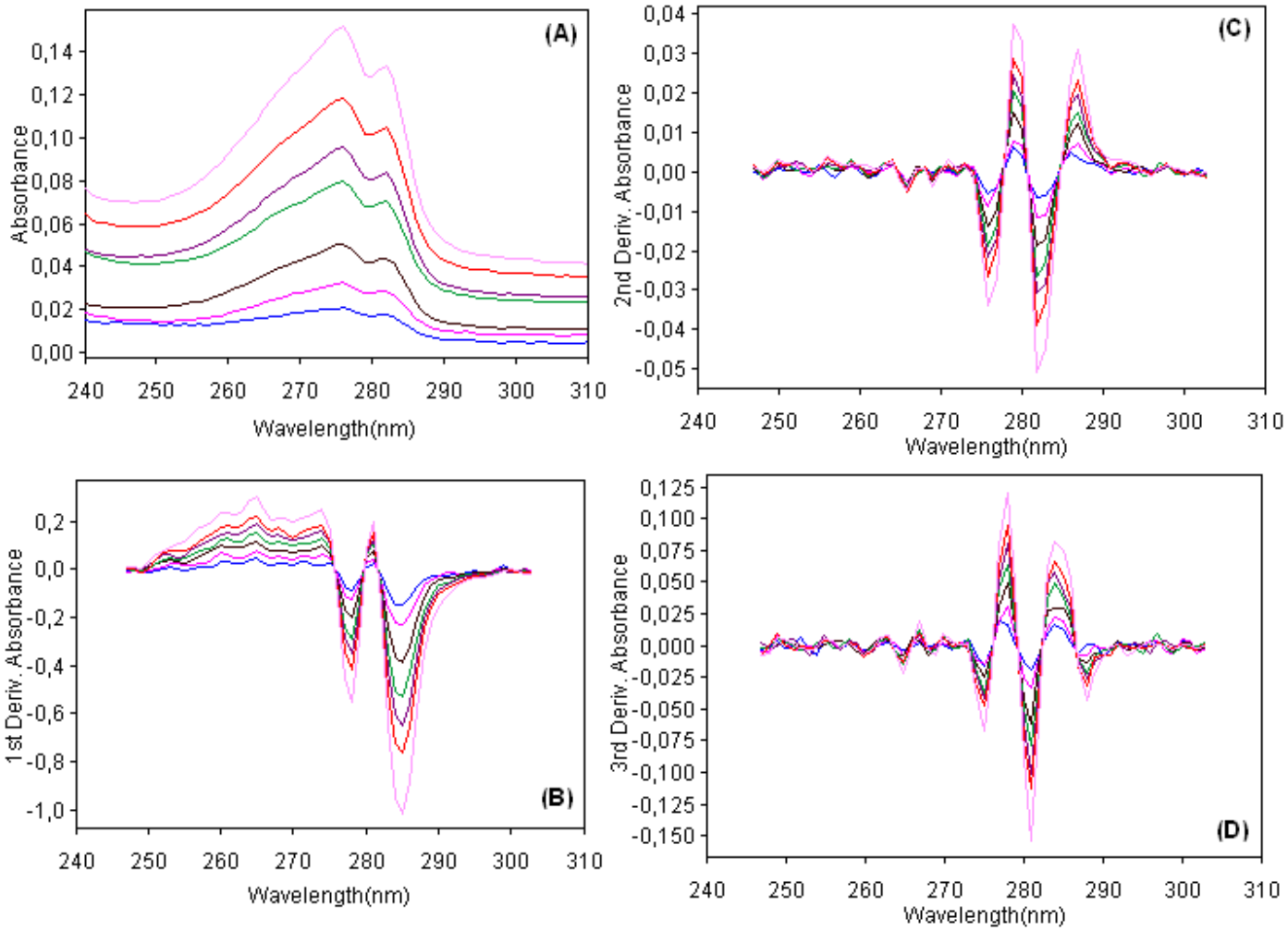

Figure 2. Spectrum of obtaining calibration graph point: (A) Zero-, (B) First-, (C) Second- and (D) Third-order derivative spectrum of standard solution of metoprolol.

2. Validation of the method:

2.1. Linearity

For quantitative analysis of metoprolol, the calibration curves were plotted for each spectrophotometric method over the concentration ranges cited. The peak to zero method for calibration curve in the first-, second- and third-order derivative spectrophotometric methods were used. The linearity ranges of all spectrophotometric methods were found to be 3.0-20 µg/mL (Figures 3A-D). The statistical parameters and regression equations which were calculated from the calibration curves along with the

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standard error of the slope and the intercept are given in Table 1.

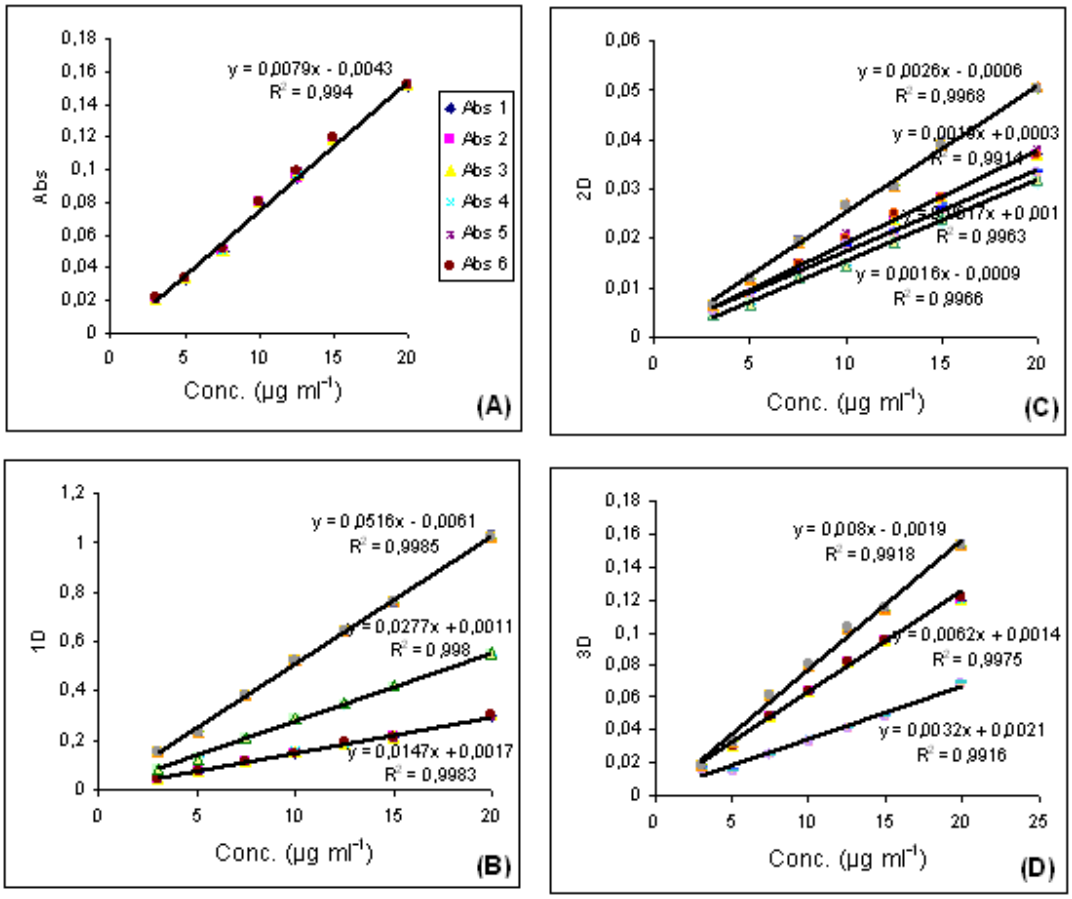

Figure 3. (A) Zero-, (B) First-, (C) Second- and (D) Third-order derivative calibration curves of metoprolol.

2.2. Specificity

Comparison of the zero-, first-, second- and third-order derivative spectrum of metoprolol in standard and drug formulation (Problok and Beloc ZOK tablet) solutions show that the wavelength of maximum and minimum absorbance did not changed (Figures 4A-D). According to the results obtained, the zero-, first-, second- and third-order derivative spectrophotometric methods are able to access the metoprolol in presence of excipients and hence, methods can be considered specific.

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Table 1
Results of regression analysis of metoprolol by the proposed methods.

Method	Range (µg/mL)	LR[a]	Sa	Sb	R^2	LOD	LOQ
Zero-order Spectrophotometric Method	3.0-20	$A_{276\ nm}$=0.0079x-0.0043	0.0036	0.0019	0.9940	0.80	2.42
First-order Spectrophotometric Method	3.0-20	$1D_{265\ nm}$=0.0147x+0.0017	0.0061	0.0032	0.9983	0.73	2.21
		$1D_{278\ nm}$=0.0277x+0.0011	0.0043	0.0060	0.9980	0.72	2.18
		$1D_{285\ nm}$=0.0516x-0.0061	0.0142	0.0109	0.9985	0.70	2.12
Second-order Spectrophotometric Method	3.0-20	$2D_{276\ nm}$=0.0017x+0.001	0.0002	0.0004	0.9963	0.78	2.36
		$2D_{279\ nm}$=0.0019x+0.0003	0.0004	0.0003	0.9914	0.52	1.58
		$2D_{282\ nm}$=0.0026x-0.0006	0.0005	0.0005	0.9968	0.63	1.92
		$2D_{287\ nm}$=0.0016x-0.0009	0.0006	0.0003	0.9966	0.62	1.88
Third-order Spectrophotometric Method	3.0-20	$3D_{275\ nm}$=0.0032x+0.0021	0.0089	0.0007	0.9916	0.72	2.18
		$3D_{278\ nm}$=0.0062x+0.0014	0.0049	0.0012	0.9975	0.64	1.94
		$3D_{285\ nm}$=0.008x-0.0019	0.0152	0.0015	0.9918	0.62	1.88

λ: Wavelength, [a]Based on six calibration curves, LR: Linear regression Sa: Standard deviation of intercept of regression line, Sb: Standard deviation of slope of regression line, R^2: Coefficient of correlation, x: metoprolol concentration (µg/mL), LOD: Limit of detection, LOQ: Limit of quantitation, A: Absorbance, 1D: First-, 2D: Second-, 3D: Third-order absorbance.

2.3. Precision and accuracy:

The precision of the analytic methods were determined by repeatability (within-day) and intermediate precision (between-day). Three different concentrations which were quality control samples (4.0, 9.0, 17.5 µg/mL) were analyzed six time in one day for within-day precision and once daily for three days for between-day precision. Repeatability was ≤2.23 %, ≤3.12 %, ≤3.81 % and ≤3.24 % (n=6) and intermediate precision was ≤3.39 %, ≤3.93 %, ≤3.92 % and ≤4.24 % (n=6) for zero-, first-, second- and third-order derivative spectrophotometric methods, respectively (Table 2). Within- and between-day accuracy of zero-, first-, second- and third-order derivative spectrophotometric methods showed acceptable relative error values were ≤0.25 %, ≤3.25 %, ≤4.00 %, ≤5.25 %, ≤1.44 %, ≤4.25 %, ≤5.25 % and ≤5.50 % (n=6), respectively (Table 2).

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Table 2

Precision and accuracy of metoprolol by the proposed methods.

Method	λ (nm)	Added (µg/mL)	Within-day			Between-day		
			Found±SD (µg/mL)	Accuracy	Precision R.S.D%[a]	Found±SD (µg/mL)	Accuracy	Precision R.S.D%[a]
Zero-order Spectrophotometric Method	$A_{276\ nm}$	4.0	4.01±0.049	0.25	1.22	3.98±0.088	-0.50	2.21
		9.0	9.02±0.192	0.22	2.13	9.13±0.285	1.44	3.12
		17.5	17.54±0.391	0.23	2.23	17.61±0.597	0.63	3.39
First-order Spectrophotometric Method	$1D_{265\ nm}$	4.0	4.02±0.052	0.50	1.29	4.08±0.093	2.00	2.28
		9.0	9.11±0.198	1.22	2.17	9.16±0.281	1.78	3.07
		17.5	17.56±0.409	0.34	2.33	17.51±0.503	0.06	2.87
	$1D_{278\ nm}$	4.0	4.11±0.089	2.75	2.17	4.14±0.118	3.50	2.85
		9.0	9.13±0.209	1.44	2.29	9.11±0.274	1.22	3.01
		17.5	17.66±0.551	0.91	3.12	17.67±0.694	0.97	3.93
	$1D_{285\ nm}$	4.0	4.13±0.088	3.25	2.13	4.17±0.129	4.25	3.09
		9.0	9.11±0.179	1.22	1.96	9.16±0.271	1.78	2.96
		17.5	17.61±0.503	0.63	2.86	17.58±0.569	0.46	3.24
Second-order Spectrophotometric Method	$2D_{276\ nm}$	4.0	3.97±0.078	-0.75	1.96	4.08±0.093	2.00	2.28
		9.0	9.13±0.275	1.44	3.01	9.12±0.289	1.33	3.17
		17.5	17.44±0.499	-0.51	2.87	17.63±0.517	0.74	2.93
	$2D_{279\ nm}$	4.0	4.16±0.948	4.00	2.28	4.21±0.135	5.25	3.21
		9.0	9.22±0.292	2.44	3.17	9.31±0.356	3.44	3.82
		17.5	17.71±0.674	1.20	3.81	17.68±0.695	1.09	3.92
	$2D_{282\ nm}$	4.0	4.20±0.091	5.00	2.17	4.17±0.135	4.25	3.24
		9.0	9.18±0.174	2.00	1.89	9.21±0.348	2.33	3.78
		17.5	17.61±0.532	0.63	3.02	17.78±0.686	1.60	3.86
	$2D_{287\ nm}$	4.0	3.95±0.090	-1.25	2.28	4.13±0.127	3.25	3.08
		9.0	8.87±0.165	-1.44	1.86	9.21±0.274	2.33	2.98
		17.5	17.36±0.415	-0.80	2.39	17.75±0.579	1.43	3.26
Third-order Spectrophotometric Method	$3D_{275\ nm}$	4.0	4.21±0.133	5.25	3.16	3.87±0.127	-3.25	3.28
		9.0	9.17±0.297	1.89	3.24	9.31±0.375	3.44	4.03
		17.5	17.86±0.511	2.06	2.86	17.68±0.568	1.03	3.21
	$3D_{278\ nm}$	4.0	4.16±0.091	4.00	2.19	4.22±0.144	5.50	3.41
		9.0	9.32±0.176	3.56	1.85	9.38±0.259	4.22	2.76
		17.5	17.70±0.566	1.14	3.20	17.81±0.725	1.77	4.07
	$3D_{281\ nm}$	4.0	3.84±0.076	-4.00	1.98	4.03±0.084	0.75	2.08
		9.0	9.23±0.256	2.56	2.78	9.31±0.361	3.44	3.88
		17.5	17.86±0.537	2.06	3.01	17.69±0.750	1.09	4.24

SD : Standard deviation of six replicate determinations, R.S.D: Relative standard derivation, [a]Average of six replicateeterminations, Accuracy: (%relative error) (found-added)/addedx100.

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2.4. Recovery:

To determine the accuracy of the zero-, first-, second- and third-order derivative spectrophotometric methods and to study the interference of formulation additives, the recovery was checked as three different concentration levels (5.0, 10, 15 µg/mL) and analytical recovery experiments were performed by adding known amount of pure drugs to pre-analyzed samples of commercial dosage form (Problok and Beloc ZOK tablet). The percent analytical recovery values were calculated by comparing concentration obtained from the spiked samples with actual added concentrations. The recoveries of zero-, first-, second- and third-order derivative spectrophotometric methods were 99.6 %, 100.3 %, 101.2 % and 100.2 (Table 3).

Table 3

Recovery values of metoprolol in pharmaceutical preparations.

Commercial Preparation	Method	λ (nm)	Added (µg/mL)	Found (µg/mL)	Recorvery (%)	R.S.D[a] (%)
Problok tablet (2µg/mL)	Zero-order Spectrophotometric Method	$A_{276\ nm}$	5	4.97±0.113	99.4	2.27
			10	9.98±0.316	99.8	3.17
			15	14.93±0.318	99.5	2.13
	First-order Spectrophotometric Method	$1D_{285\ nm}$	5	4.92±0.146	98.4	2.96
			10	10.17±0.294	101.7	2.89
			15	15.93±0.289	100.9	1.91
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	5	5.13±0.115	102.6	2.24
			10	10.17±0.312	101.7	3.07
			15	14.91±0.428	99.4	2.87
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	5	4.89±0.138	97.8	2.82
			10	10.14±0.421	101.4	4.15
			15	15.18±0.541	101.2	3.56
Beloc ZOK tablet (2µg/mL)	Zero-order Spectrophotometric Method	$A_{276\ nm}$	5	5.11±0.157	102.2	3.07
			10	10.13±0.427	101.3	4.22
			15	15.22±0.584	101.5	3.84
	First-order Spectrophotometric Method	$1D_{285\ nm}$	5	5.17±0.168	103.4	3.25
			10	9.93±0.306	99.3	3.08
			15	15.21±0.432	101.4	2.84
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	5	5.10±0.154	102.0	3.02
			10	10.08±0.291	100.8	2.89
			15	15.07±0.478	100.5	3.17
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	5	4.90±0.129	98.0	2.63
			10	10.12±0.407	101.2	4.02
			15	15.21±0.509	101.4	3.45

SD: Standard deviation of six replicate determinations, R.S.D: Relative standard derivation, [a]Average of six replicate determinations.

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2.5. Stability:

To evaluate the stability of metoprolol, standard solutions were prepared separately at concentrations covering the low, medium and higher ranges of calibration curve for different temperature and times. These solutions were stored at room temperature, refrigeratory (4 °C) and frozen (-20 °C) temperature for 24 h and 72h. Stability measurements were carried out with zero-, first-, second- and third-order derivative spectrophotometric methods. The results were evaluated comparing these measurements with those of standards and expressed as percentage deviation and metoprolol was found as stable at room temperature, 4 and -20 °C for at least 72h (Table 4).

Table 4
Stability of metoprolol in solution.

Stability (%)		Room temperature stability (Recovery % ± SD)		Refrigeratory stability, +4°C (Recovery % ± SD)		Frozen stability, - 20°C (Recovery % ± SD)	
λ (nm)	Added (µg/mL)	24 h	72 h	24 h	72 h	24 h	72 h
A_{276} nm	5	99.1±0.578	99.3±0.582	101.2±0.639	102.2±1.961	101.1±4.479	102.2±1.817
	10	98.1±0.017	100.4±0.187	99.4±0.077	98.7±0.162	102.3±0.086	98.2±0.742
	20	100.3±0.078	102.4±0.432	102.8±0.215	102.1±0.059	99.5±0.127	101.0±0.087
$1D_{285}$ nm	5	98.5±0.453	98.7±0.113	102.9±0.074	98.7±3.206	100.1±1.025	98.6±0.264
	10	100.2±0.087	101.5±0.084	98.7±4.517	100.9±2.024	99.3±0.095	98.7±0.221
	20	102.6±1.597	102.1±0.088	103.0±1.218	99.1±1.234	102.2±0.086	101.2±0.098
$2D_{282}$ nm	5	99.6±2.541	101.2±2.564	101.1±1.968	104.6±1.310	101.1±2.895	99.7±1.747
	10	101.3±1.876	102.1±2.135	98.87±0.148	103.5±0.093	102.5±0.083	100.4±1.319
	20	99.5±0.724	101.3±2.523	102.0±0.150	98.7±1.028	98.0±0.677	99.7±1.537
$3D_{281}$ nm	5	99.8±2.541	101.2±2.564	101.1±1.988	104.2±1.302	101.2±2.865	99.8±1.776
	10	101.2±1.876	102.1±2.135	98.8±0.158	103.1±0.092	102.5±0.076	100.2±1.317
	20	99.6±0.744	101.2±2.523	102.0±0.130	98.9±1.028	98.4±0.687	99.7±1.593

SD :Standard deviation of six replicate determinations.

2.6. *Limits of detection (LOD) and quantitation (LOQ):*

The LOD and LOQ of metoprolol by the proposed methods were determined using calibration standards. LOD and LOQ values were calculated as 3.3 σ/S and 10 σ/S, respectively, where S is the slope of the calibration curve and σ is the standard deviation of y-intercept of regression equation (n=6)[17] (Table 1).

DISCUSSIONS

Zero-, first-, second- and third-order derivative spectrophotometric methods were applied for the determination of the commercial tablet (Table 5). The results show the high reliability and reproducibility of four methods. The best results obtained at 276 nm, 285 nm, 282 nm and 281 nm for zero-, first-, second- and third-order derivative spectrophotometric methods were statistically

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compared using the F-test. At 95 % confidence level, the calculated *F*-values do not exceed the theoretical values (Table 6). Therefore, there is no significant difference between zero-, first-, second- and third-order derivative spectrophometric methods.

Table 5
Determination of metoprolol in pharmaceutical preparations.

Commercial Preparation	Method	λ (nm)	n	Found ± SD (mg)	Recorvery (%)	R.S.D[a] (%)	Confidence Interval
Problok (100 mg/tablet)	Zero-order Spectrophotometric Method	$A_{276\ nm}$	6	101.3±2.269	101.3	2.24	99.2-101.4
	First-order Spectrophotometric Method	$1D_{285\ nm}$	6	99.5±2.557	99.5	2.57	98.6-102.7
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	6	98.7±2.358	98.7	2.39	97.6-101.8
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	6	100.2±2.194	100.2	2.19	98.6-102.4
Beloc ZOK (100 mg/tablet)	Zero-order Spectrophotometric Method	$A_{276\ nm}$	6	98.7±3.039	98.7	3.08	97.5-101.3
	First-order Spectrophotometric Method	$1D_{285\ nm}$	6	100.3±3.219	100.3	3.21	98.76-102.7
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	6	101.2±2.499	101.2	2.47	99.4-102.7
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	6	99.7±1.924	99.7	1.93	97.9-102.2

SD: Standard deviation of six replicate determinations, R.S.D: Relative standard derivation, [a]Average of six replicate determinations.

There is a study for determination of metoprolol by zero-order derivative spectrophotometry method[15] in literature. In this study, the method is based on the formation of Cu (II) dithiocarbamate complex by derivatization of the secondary amino group of metoprolol with CS_2 and $CuCl_2$ in the presence of ammonia. The copper-bis(dithiocarbamate) complex was extracted into chloroform and the concentration of metoprolol tartrate was determined directly by spectrophotometry.

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Table 6
Statistical comparison (F-test) of the results obtained by proposed methods.

Commercial Preparation	Method	λ (nm)	n	Mean ± SD (mg)	P value	F-test
Problok (100 mg/tablet)	Official method (HPLC)	-	-	100.4±0.85	0.321	F_c=1.97 Ft=3.00
	Zero-order Spectrophotometric Method	$A_{276\ nm}$	6	101.3±2.269		
	First-order Spectrophotometric Method	$1D_{285\ nm}$	6	99.5±2.557		
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	6	98.7±2.358		
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	6	100.2±2.194		
Beloc ZOK (100 mg/tablet)	Official method (HPLC)	-	-	100.4±0.85	0.289	F_c=1.83 Ft=3.00
	Zero-order Spectrophotometric Method	$A_{276\ nm}$	6	98.7±3.039		
	First-order Spectrophotometric Method	$1D_{285\ nm}$	6	100.3±3.219		
	Second-order Spectrophotometric Method	$2D_{282\ nm}$	6	101.2±2.499		
	Third-order Spectrophotometric Method	$3D_{281\ nm}$	6	99.7±1.924		

n: number of determination, SD: Standard deviation of six replicate determinations, Fc: Calculated F values, Ft: Tabulated F values, Ho hypothesis: no statistically significant difference exists between five methods, $F_t > F_c$; Ho hypothesis is accepted ($P > 0.05$).

DETERMINATION OF METOPROLOL IN PHARMACEUTICAL PREPARATIONS BY ZERO-, FIRST-, SECOND- AND THIRD-ORDER DERIVATIVE SPECTROPHOTOMETRIC METHOD

Also, the suggested zero-, first-, second- and third-order derivative spectrophotometric methods were compared with the HPLC method of USPXXII[18]. There was no significant difference between the five methods with respect to mean values and standard deviations at the 95% confidence level (Table 6). Therofore, this is suggested that the four methods are equally applicable.

CONCLUSION

Zero-, first-, second- and third-order derivative spectrophometric methods were developed for the determination of metoprolol in tablet dosage form. Metoprolol can be directly determined in tablets in presence of excipients without sample pre-treatment procedures by using spectrophotometric methods. The apparatus and reagents used seem to be accessible even for the simple laboratories. Also, no significant difference was found between the proposed spectrophotometric methods. Therefore, developed methods can be recommended for routine and quality control analysis of metoprolol.

DETERMINATION OF METOPROLOL IN PHARMACEUTICAL PREPARATIONS BY ZERO-, FIRST-, SECOND- AND THIRD-ORDER DERIVATIVE SPECTROPHOTOMETRIC METHOD

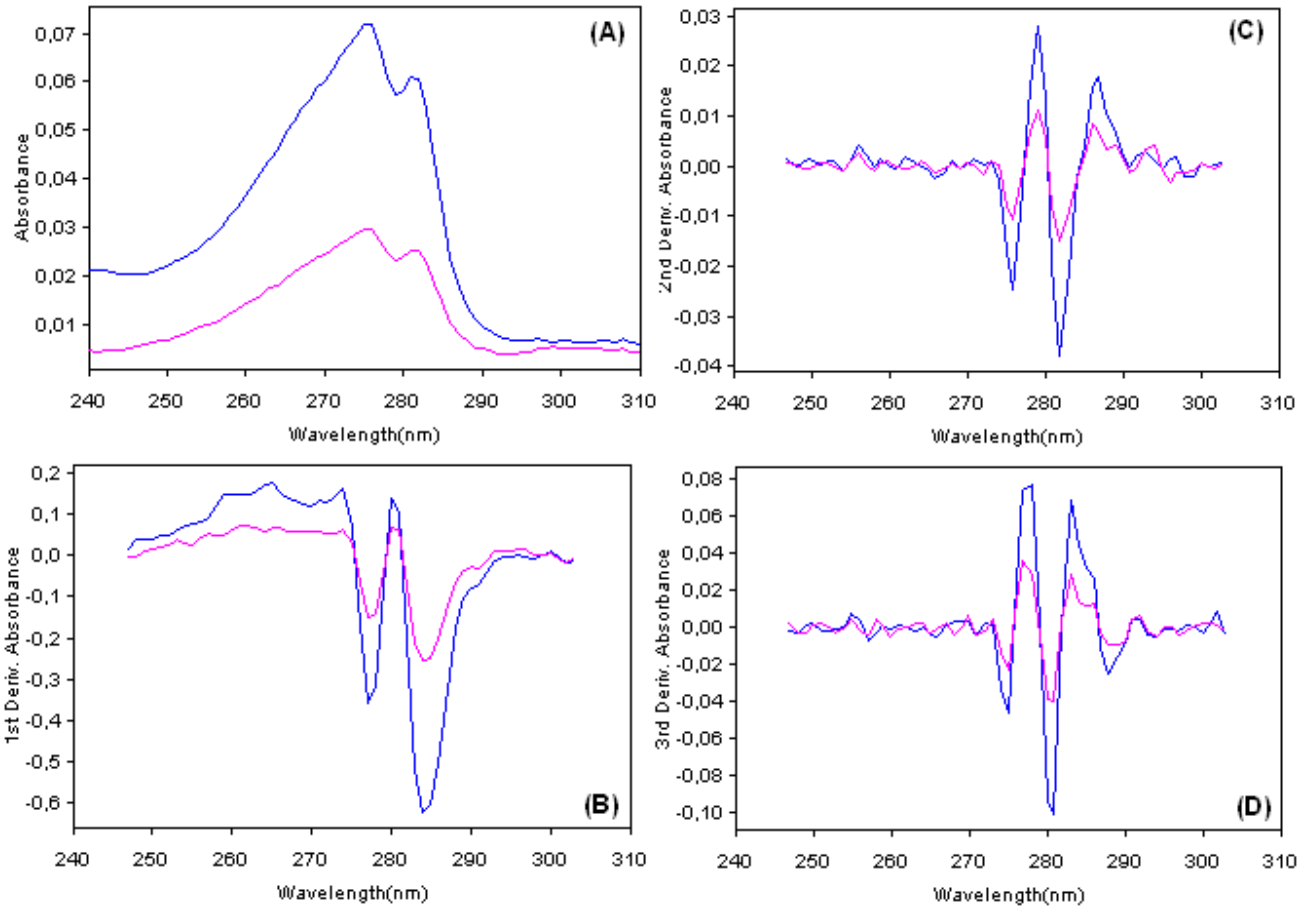

Figure 4. Spectrum of solutions of Problok tablet containing metoprolol (5 and 15 µg/mL): (A) Zero-, (B) First-, (C) Second- and (D) Third-order derivative spectra.

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