Black Sea Journal of Engineering and Science
doi: 10.34248/bsengineering.1436216

Open Access Journal
e-ISSN: 2619 – 8991

Research Article
Volume 7 - Issue 2: 359-364 / March 2024

A NEW SPECTROPHOTOMETRIC METHOD FOR DETERMINATION OF CARVEDILOL FROM TABLET

Figen EREK[1]*, Işıl AYDIN[2]

[1]*Dicle University, Faculty of Science, Department of Chemistry, Diyarbakır, 21280, Türkiye*
[2]*Dicle University, Faculty of Pharmacy, Department of Analytical Chemistry, Diyarbakır, 21280, Türkiye*

Abstract: In this study, a new spectrophotometric method was developed for the quantitative analysis of Carvedilol and the method was validated. The method depends on the reaction between the carvedilol and 1,2,5,8-tetrahydroxyanthraquinone in methanol to yield colored charge transfer complex giving maximum absorbance at 560 nm. For optimization of the proposed method, several parameters were investigated such as solvent type, reaction time, and quinalizarin concentration. The stoichiometry of colored charge transfer complex was found to be 2:1(reagent: drug) by Job's method. Beer's Law is obeyed in the concentration range of 0.5-60 µg/mL with 0.9986 correlation efficient. Limit of detection (LOD) and limit of quantifiqation (LOQ) were found 0.147 µg/mL, 0.491 µg/mL, respectively. The proposed method can be successfully applied pharmaceutical formulation.

Keywords: Charge transfer complex, Job's method, Molecular absorption spectroscopy

***Corresponding author:** Dicle University, Faculty of Science, Department of Chemistry, Diyarbakır, 21280, Türkiye
E mail: figen.erek@dicle.edu.tr (F. EREK)
Figen EREK https://orcid.org/0000-0002-2861-5504
Işıl AYDIN https://orcid.org/0000-0001-6571-6032

Received: February 13, 2024
Accepted: March 05, 2024
Published: March 15, 2024

Cite as: Erek F, Aydın I. 2024. A new spectrophotometric method for determination of carvedilol from tablet. BSJ Eng Sci, 7(2): 359-364.

1. Introduction

Beta-blockers, which reduce the activity of sympathetic nerves by blocking beta-adrenergic receptors, are in the cardiovascular drug class (Mutlu, 2010; Vale et al., 2019). β-blockers are divided into three groups as first generation non-selective group, second generation β1-selective group, third generation β1-high selective and vasodilator group (Mutlu, 2010).

Carvedilol (CRV), [(6)-1-(carbazole-4-yloxy)-3-((2-(o-methoxyphenoxy)ethyl)amino)-2-propanol], is a third-generation β-blocker agent having nonselective β_1-adrenoreceptor blockade, vasodilation, antioxidant properties (Streim et al., 1987; Brian and William, 2004). CRV is a white powder, whose solubility is very good in dimethylsulfoxide; soluble in methylene chloride and methanol; slightly soluble in ethanol and isopropanol; insoluble in water. Its melting point is in the range of 114-115 °C (Moffat et al., 2004).

Several methods have been developed for analysis of carvedilo in the literature, such as chromatographic (Junwei et al., 2015), spectrophotometric (Verma and Syed, 2007, Rani et al., 2013), electroanalytic (Soleymanpour and Ghasemian, 2015) detections.

Spectrophotometric methods are, espically UV-Visible spectrophotometry, often preferred techniques due to its low cost and simplicity in the determination analysis of pharmaceutical preparations (Ragaa et al., 2013; Kirtimaya et al., 2016). Charge transfer reactions have been commonly studied by UV-Visible spectrophotometic methods. The charge transfer complexes are formed by the molecular interaction of electron donors and acceptors. These colored complexes should absorb at a different wavelength from the donor and acceptor moleculars at the UV-visible region (Ragaaa et al. 2013).

In this study, a new, simple, rapid, validated spectrophotometric method that does not interfere with the excipients has been developed for the determination of carvedilol (CRV) in pharmaceutical formulation. Direct analyzes of Carvedilol with UV-vis spectrophotometer cannot be performed because excipients interfere with pharmaceutical formulations. In this method, direct analysis was performed by extracting carvedilol from pharmaceutical tablet without applying any purification process. The method depends on the reaction between the CRV (π-acceptor group) and 1,2,5,8-tetrahydroxyanthraquinone (π-donor group) in the methanol to yield colored charge transfer complex that is measured at 560 nm. This colored complex made possible the quantitative determination of CRV, spectrophotometrically.

2. Materials and Methods

2.1. Aparatus and Reagents

The UV-VIS spectral measurements were made by using T80+ UV/VIS Spectrometer (PG Instruments Ltd), Varian Cary 100 Bio UV/VIS double beam UV–VIS spectrophotometers equipped with 1 cm matched quartz cells. All solvents (methanol, ethanol, 1-propanol, 2-

propanol, chloroform, N,N-dimethylformamide (DMF), acetone and acetonitrile) used in this work were of HPLC grade and Merck branded. CRV, which is certified to be 99.5% pure, was obtained from Bilim Drug Company, Istanbul, Türkiye. Coronis tablet (Bilim Drug Company) labeled to contain 25 mg CRV per tablet used in the proposed method on was received from in the local pharmacy. Quinalizarin, (1,2,5,8-tetrahydroxy-anthraquinone), was purchased from Sigma Aldrich company. Sucrose, Povidone K25, Magnesium stearate, Lactose monohydrate were purchased from Glentham Life Sciences Company.

The stock solutions 500 µg/mL of CRV and 3.10^{-3} mol/L of Quin were prepared with methanol in 50 mL flasks, daily.

2.2. Construction of Calibration Curves

The aliquots in the range of 0.01-1.2 mL CRV (500 µg/mL) solution were transferred to volumetric flasks (at 27 °C) to obtain concentrations in the range 0.5-60 µg/mL of CRV. Following this, 1.25 mL of 3.0 mmol/L Quin solution was added to these solutions and completed to 10 mL with methanol. These final mixtures were thorougly mixed. Three replications were made for each solution.The absorbance of the prepared solutions was measured at 560 nm against Quin blanks. Finally, the measured absorbances versus the concentration of CRV were plotted.

2.3. Assay Procedure for Tablets

The average weight of a tablet containing 25 mg of CRV was calculated by weighing ten randomly tablets. The weighed ten tablets, then, were finely powdered in a dry and clean agate mortar. Afterwards, an accurately weighed quantity of the powder equivalent to 25 mg of CRV were transferred into the volumetric flask and dissolved in 25 mL methanol. The final mixture was both shaken and sonicated for 30 min, and then filtered by using quantitative filter paper. The volume of filtered solution was completed to the mark with methanol to prepare a stock solution of 250 µg/mL. Then, some working concentrations were prepared from the filtered solution. The proposed method was applied to the prepared solutions. The amount of CRV in the tablet was calculated according to the calibration curve.

3. Results and Discussion

3.1. The Effect of the Solvent Type

In the method based on the formation of colored charge transfer complex, the optimum solvent is to facilitate the charge transfer (Gouda and Kassem, 2012; Mohammed et al., 2018). Therefore the effects of solvents such as Methanol, DMF, Ethanol, Acetonitrile, Chloroform, 1-propanol, 2 -propanol and Acetone, on the charge transfer reaction were investigated.

As seen in Table 1, Methanol, which gave the highest absorbance of the complex, was accepted as the optimum solvent. The charge transfer reaction is thought to proceed through the radical anion. It is thought that the capacity of methanol to form stable hydrogen bonds with the radical anion is higher than that of acetonitrile (Gouda and Kassem, 2012). While Quin gives maximum absorbance at 491 nm in methanol, CRV-Quin complex gives maximum absorbance at 560 nm. The fact that Quin and CRV-Quin charge transfer complex absorb at different wavelengths in the UV-visible region showed that CRV can be determined easily and precisely with Quin (Gouda and Kassem, 2012).

Table 1. The effect of solvents type

Solvents	Absorbance	±SD
Methanol	0.501	0.014
DMF	0.264	0.015
1-propanol	0.142	0.023
Acetonitrile	0.130	0.034
Ethanol	0.123	0.028
2-propanol	0.112	0.016
Acetone	0.061	0.004
Chloroform	0.041	0.002

SD= standard deviation

3.2. The Effect of the Reagent Concentration

The transformation of a very large part of the analyte in solution into the colored complex in a short time, that is, the rapid transfer of charge to equilibrium and achieving the maximum sensitivity of the complex also depends on the concentration of the reagent (Gouda and Kassem, 2012; Mohammed et al., 2018). To determine optimum concentration of reagent, 0.25-2.25 mL of 3.10^{-3} mol/L Quin was added into the CRV solutions. The solution was stirred for 2 min to complete the reaction. At the end of the study, the optimum volume of 3.10^{-3} mol/L Quin solution was found to be 1.25 mL. After this point, it was observed that the absorbance did not change much and reached the maximum density. The related results are shown in Figure 3.

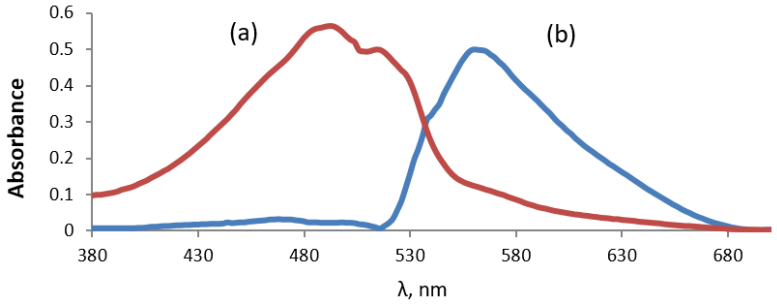

Figure 2. Spectra of Quin (a) and Quin-CRV complex (b) in methanol.

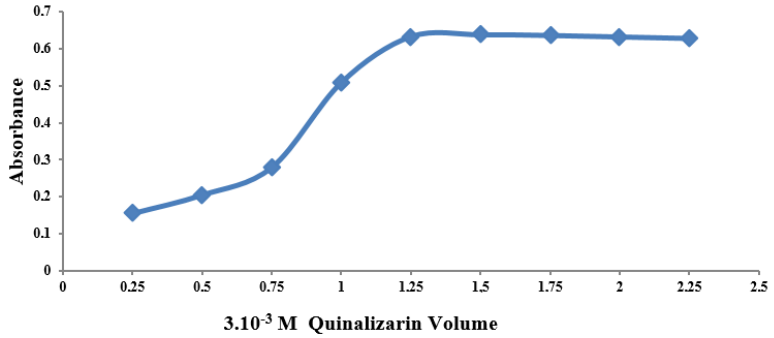

Figure 3. Effect of the quinalizarin concentration.

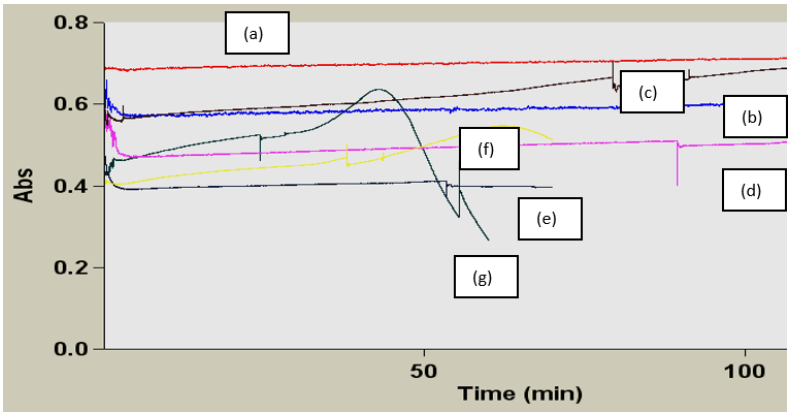

Figure 4. The reaction time and temperature CRV-Quin charge transfer complex (27 ˚C: (a), 35 ˚C: (b), 40 ˚C: (c), 45 ˚C: (d), 50 ˚C: (e), 55 ˚C: (f), 60 ˚C: (g)).

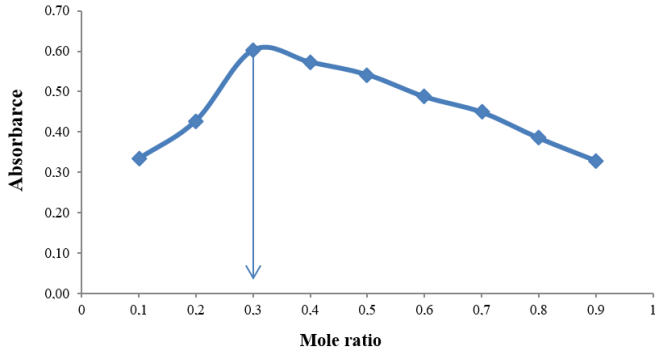

Figure 5. Job's method to the reaction between Quin and CRV (λ= 560 nm).

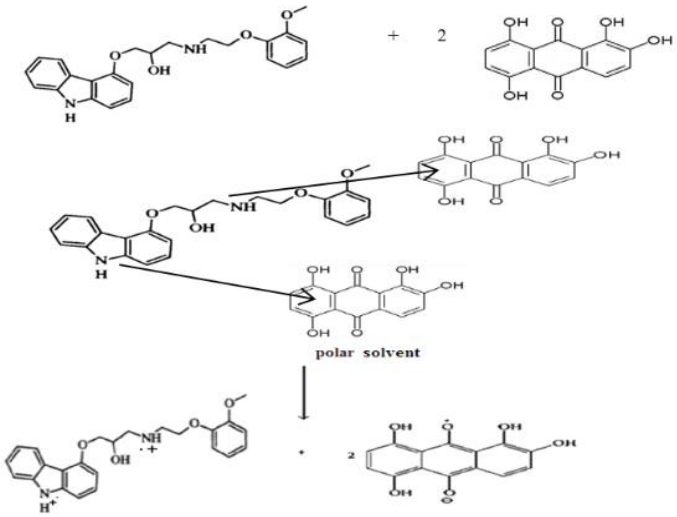

Figure 6. Possible mechanism of radical anion formation from Quin and CRV reaction.

3.3. The Effect of the Reaction Time and Temperature

It was observed that the absorbance of the colored complex was both the most stable and the maximum at 27±2 °C (laboratory temperature). For this reason, it was decided that the optimum temperature was 27±2 °C. The absorbance of the charge transfer complex remained unchanged from the beginning for 120 min at 27 °C. Therefore, the optimum reaction time was kept shorter than 1 min at 27 °C. The absorbance of the complex was immediately measured. As the temperature increased, the stability of the complex deteriorated (Figure 4).

3.4. Characterization of CRV-Quin Charge Transfer Complex

Job's continuous variation method (Kelani et al., 1997) was used to determine the stoichiometry of the charge transfer complex in methanol medium. As seen in Figure 5, it has been determined that CRV and Quin react at a rate of 1-2. The proposed mechanism for this reaction is shown in Figure 6. It was thought that the charge transfer complex (n-π) was formed by transferring the loads in the -NH and -OH groups of the drug to the center with load deficiency in Quin. According to the literature, the mechanism related to n-π type interaction has also been proposed (Gouda et al., 2012).

3.5 Validation

3.5.1. Analytical performance of developed method

Firstly, the linear working range was found that this range is 0.5-60 µg /mL. The equation of the calibration curve is y = 0.0169x + 0.0778 with 0.9981 correlation coefficient (y=Absorbance of complex, x=Concentration of CRV (µg/mL)). In this method, LOD and LOQ at 95 % confidence level were found to be 0.147 µg/mL, 0.491 µg/mL, respectively. LOQ and LOD were calculated according to the Equation 1:

$$LOD=3s/b,\ LOQ=10s/b \quad (1)$$

where *s*: standard deviation of absorbances of blank solution *b:* the slope of the calibration curve (URL1).

All parameters related to calibration are given in Table 2.

Accuracy: In the recovery study with pure CRV, values were obtained between 98.8 and 100.8% (Table 3). In the standard method of addition, recovery was obtained in the range of 98.4 to 99.4% (Table 4). In these two methods, RE % was lower than 1.6% and RSD % was lower than 1.5%. The high and good accuracy of the method depends on the low RSD % and RE %. RE % and RSD % were found to be low in the proposed method. At 95% confidence level and 4 degrees of freedom, the experimental t was calculated as 1.97 and the recovery was found to be 100.7%. The theoretical t value is taken as 2.77. The experimental t value being less than the theoretical t value indicates that there is no systematic error in the method and it is applicable for the determination of CRV in tablets (URL1).

Precision: In the precision study, the solutions prepared at any concentration within the working range were analyzed at 3 different times within the same day (in 1 day) and on 3 different days, and the results were evaluated (Table 6). When the proposed method is applied to the solutions of CRV prepared in two different concentrations during the intraday and inter days, it is seen that the difference between absorbances is not much. RSD % is lower than 2% (URL1).

Stability: In the stability study, after applying the proposed method to 15 µg/ mL, 45 µg/mL solutions, and absorbance measurements of each solution were taken at 24 h intervals for 72 h. The low change in absorbance of the solutions for 72 h indicates that the complex is very stable (URL1).

Specificity: The effects of the excipients such as Lactosemonohydrate, Sucrose, Povidone K25, Magnesium stearate (Mg-St.) on the absorbance were investigated by applaying proposed method to solution 45 µg/mL of CRV (Table 7) (Korkmaz et al., 2005). In this method, it has been observed that these excipients do not have a significant effect on absorbance.

Robutness: Small changes were made in the optimization parameters in the robustness study of the proposed method (URL1, Altunay et al., 2023). In the study, changes of ±0.05 mL in Quin volume and ±2ºC in temperature were examined. In both parameters, recoveries were obtained in the range of 99.00-100.22 %. RSD % was lower than 1% and RE % was less than 1.05 % (Table 8). It was observed that the method was not affected by small changes in optimization parameters.

Table 2. Optimum conditions and analytical parameters

Parameters	Values
λ_{maks}	560
Linearity dynamic range (µg/mL)	0.5-60
Correlation coefficient (r^2)	0.9981
Regression equation	y=0.0169x+0.0778
Slope	0.0169
Interception	0.0778
LOD (µg/mL)	0.147
LOQ (µg/mL)	0.491
Molar Absorptivity ($L.mol^{-1}.cm^{-1}$)	2737

Table 3. Recovery of CRV in proposed method

Taken (µg/mL)	Found Conc. (µg/mL)	Recovery%	SD	RSD%	RE%
15	14.82	98.83	0.07	0.46	1.17
30	29.79	99.32	0.4	1.35	0.68
45	44.57	99.04	0.43	0.96	0.96
60	59.87	99.79	0.84	0.84	0.21

Table 4. the standard addition technique for the determination of the CRV in pharmaceutical preparation

Taken	Added (µg/mL)	Recovery%	SD	RSD%	RE%
20µg/mL	8	98.6	0.19	0.69	1.41
	12	99.4	0.33	1.02	0.57
	16	99.1	0.39	1.11	0.93
	20	98.4	0.53	1.34	1.57
	24	99.0	0.39	0.89	0.97

Table 5. t-test for pharmaceutical preparation of CRV

Commercial Brand	Found Calculated ±SD	Recovery%	t test
Coronis (25 mg CRV)	25.17 ± 0.19	100.68	1.97

Table 6. Intra-day and inter-day precision of proposed method (Taken Concentration: 45 µg/mL and 15 µg/mL)

İntra-day			İnter-day		
Found Conc. (µg/mL)	44.79	15.00	Found Conc. (µg/mL)	44.57	14.96
±SD	0.06	0.12	±SD	0.18	0.21
RSD%	0.14	0.83	RSD%	0.41	1.40

Table 7. Effect of interfer substances in proposed method

$İ_1$:$İ_2$ (µg/mL)	LMH Recovery%	Sucrose Recovery%	Povidon K25 Recovery%	Mg-St Recovery%
0:45	100	100	100	100
45:45	98.6	99.8	100.1	95.2
450:45	98.8	100	100.2	103.0
4500:45	100.6	99.8	96.6	103.2

$İ_1$: interfere s. conc., $İ_2$: CRV Conc.

Table 8. The effect of small change in Quin volume and reaction temperature on absorbance

		Recovery%	SD	RSD%	RE%
Quin Volume (mL)	1.25 + 0.05	99.00	0.36	0.80	1.05
	1.25 – 0.05	100.22	0.21	0.47	0.22
Temperature (°C)	27 + 2	99.04	0.37	0.83	0.96
	27 – 2	99.22	0.15	0.33	0.78

4. Conclusion

In this study, spectrophotometric method based on charge transfer reaction between CRV and Quin was developed for determination of CRV from tablet formulation. The analytical procedure of this study was applied by optimizing several parameters such as solvent type, reagent concentration, reaction temperature and time, stoichiometric ratio. Then the method was validated. The stoichiometry of colored charge transfer complex was found to be 2:1(reagent:drug) by Job's method. Beer's Law is obeyed in the concentration range of 0.5-60 mg/L with 0.9986 correlation efficient. LOD and LOQ were found 0.147 mg/L, 0.491 mg/L, respectively. The proposed method has some advanteges such as being very simple and accurate, robust, without interference and economic. In addition, it does not require extra processes such as purification, heating, a buffer.

Author Contributions

The percentage of the author(s) contributions is presented below. All authors reviewed and approved the final version of the manuscript.

	F.E.	I.A.
C	90	10
D	100	
S	100	
DCP	90	10
DAI	80	20
L	90	10
W	70	30
CR	50	50
SR	100	
PM		100
FA		100

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The authors declared that there is no conflict of interest.

Ethical Consideration

Ethics committee approval was not required for this study because of there was no study on animals or humans.

Acknowledgements

We would like to thank to Dicle University for supporting the subject scientific study (Project number: ECZ: 19.001).

References

Altunay N, Haq HU, Castro-Mu˜noz R. 2023. Optimization of vortex-assisted hydrophobic magnetic deep eutectic solvent-based dispersive liquid phase microextraction for quantification of niclosamide in real samples. Food Chem, 426: 136646.

Briann D, William TA. 2004. Pharmacology of carvedilol. Am J Cardiol, 93: 3B–6B.

Gouda AA, Kasssem M. 2012. Novel spectrophotometric methods for determination of desloratidine in pharmaceutical formulations based on charge transfer reaction. Arabian J Chem, 9: 1712-1720. https://doi.org/10.1016/j.arabjc.2012.04.050.

Junwei L, Wang L, Wang S, Chen M, Hu EG, Ge R 2015. Simultaneous quantification of carvedilol and its metabolites in rat plasma by ultra-performance liquid chromatography tandem mass spectrometry and pharmacokinetic application. J Chromatograp B, 974: 138-146. https://doi.org/10.1016/j.jchromb.2014.10.037.

Kelani K, Bebawy LI, Abdel-Fattah L, Ahamad AS. 1997. Spectrophotometric determination of some n-donating drugs using DDQ Anal. Letters, 10(30): 1843–1860.

Kirtimaya M, Kumar BK, Kumari MM, Subrahmanyam BSS. 2016. New analytical method development and validation of chlorpheniramine maleate by using UV-visible spectrophotometry. Indo American J Pharmaceut Sci, 7: 767-772.

Korkmaz D, Demir C, Aydın F, Ataman OY. 2005. Cold vapour generation and on-line trapping of cadmium species on quartz surface prior to detection by atomic absorption spectrometry. J Anal At Spectrom, 20: 46-52.

Moffat AC, Osselton MD, Widdop B. 2004. Clarke's analysis of drugs and poisons. Pharmaceutical Press, London, UK, pp: 2736.

Mohamed EM, Frag YZ, Hathoot AA, Shalaby EA. 2018. Molecular and biomolecular spectroscopy. Spectrochimica Acta Part A: 189: 357-365. https://doi.org/10.1016/j.saa.2017.08.027.

Mutlu B. 2010. Vasodilatator Beta blockers in cardiovascular disease. Trakya Univ J Medic Fac, 27: 26-30.

Ragaa El-S, Ayman AG, El-Azzazy R. 2013. Spectrophotometric Study on the charge transfer complex between sumatriptan succinate and some Π-acceptors and alizarin derivatives. Chem Indust Chem Eng Quart, 4: 529-540. https://doi.org/10.2298/CICEQ1205133087E.96.

Rani YN, Ravi BV, Kumar V, Smitapadma M. 2013. Development and validation of new analytical methods for the estimation of carvedilol in bulk and pharmaceutical dosage. Asian J Pharmaceut Clin Res, 6: 2.

Soleymanpour A, Ghasemian M. 2015. Chemically modified carbon paste sensor for the potentiometric determination of carvedilol in pharmaceutical and biological media. Measurement, 59: 14-20. https://doi.org/10.1016/j.Measurement.2014.09.046.

Streim K., Sponer, G, MullerBeckmann, B, Bartsch, WJ. 1987. Cardiovasc. Pharmacol, 10: 33.

URL1: https://database.ich.org/ (accessed date: November 12, 2023).

Vale GT, Ceron CS, Gonzaga NA, Simplicio JA, Padovan JC. 2019. Three Generations of β-blockers: History, class differences and clinical applicability. Curr Hypertens Rev, 15: 22. https://doi.org/10.2174/1573402114666180918102735.

Verma JK, Syed HA. 2007. Extractive spectrophotometric method for determination of carvedilol in tablets. Indian J Pharm Sci, 2: 303-304. https://doi.org/10.4103/0250-474X.33166.