INTERNATIONAL JOURNAL OF RESEARCH IN PHARMACY AND CHEMISTRY

Available online at www.ijrpc.com

Research Article

SPECTROPHOTOMETRIC METHODS FOR DETERMINATION OF CILOSTAZOL IN PURE AND DOSAGE FORMS

S. Abd. AlhamideHoballah

National Organization for Drug Control and Research, Egypt.

ABSTRACT

Two, simple, accurate, rapid, sensitive and economic methods have been developed for quantitative determination of cilostazol and its pharmaceutical forms. The first one was based on oxidative coupling using potassium periodate and resorcinol in presence of acid-medium forming pink colored complex which absorbed maximally at 541 nm. The second method was based on formation of metal complexes with cupper (II), cobalt (II) and nickel (II) in phosphate buffer with pH equals = 7.5 with heating at temperature equals 55±5c° for 30±5 minutes forming violet and green colored complexes with absorbed maximally at 572, 504, and 328 nm for cobalt (II). Cupper (II) and nickel (II), respectively. Also cilostazol forms pink complex with iron (III) chloride in acetate buffer at pH equals 4.0 which absorbed maximally at 536nm. Beer's law was obeyed in the concentration ranges of (5-30) µg/ml for the first method and of concentration range of (5-201, (1-20), (3-30), and (5-25) µg/ml for cobalt, copper, nickel and iron (III) chloride in the second method, these two methods can be applied successfully for determination of cilostazol formulations.

Keywords: spectrophotometry, resorcinol. Potassium periodate, cilostazol, cobalt (II).

INTRODUCTION

Cilostazol is chemically 6- [4-1(-cyclohexyl-1H-tetrazol-5- yl-butoxyl] 3,4-dilydro-2(III) quinolinone[1]. Cilostazol and its metabolites are cyclic adenosine monophosphate (CAMP), phosphodiesters III inhibitors, inhibiting phosphodiesterase, activity and suppressing CAM degradation with a resultant increase in CAMP in platelets and blood vessels, leading to inhibition of platelet aggregation and vasodilatation[2], therefore cilostazol is used for the treatment of intermittent claudication resulting from peripheral arterial disease. Cilostazol in commercially available as single and combined formulation cilostazol is official in United States pharmacopeia 2009. USP describes HPLC method for the assay of cilostazol and its tablets using a column packed with actadecylsilanized silica gel with a mobile phase of water, acetonitrile and methanol (10:7:30) equipped with a 254 nm detector and a flow rate of 1ml/min. There are limited reports regarding determination of cilostazol in pharmaceutical dosage forms and biological fluids, these works include HPLC, UV spectrophotometric and potentiometric methods to determine cilostazol in pharmaceutical dosage forms[3-4]. The assay of cilostazol in the human plasma and mouse serum are also reported by HPLC methods[5-13] and electro-chemical methods[14-21]. The present paper aims to evaluate simple accurate, rapid, sensitive and economic methods for determination of cilostazol in pure from as well as in dosage forms and these methods can be use in quality control and quality assurance laboratories.

EXPERIMENTAL PROCEDURES

1. Instruments

A Schimdzu 1660 µV-spectrophotometer with 1-cm matched quartz cells was used to measure absorbance of resulting colored complex.

pHmeter (Hana) was used to adjust pH-values of buffer solutions.

2. Reagents

All chemicals and reagents used were of analytical grade and all solutions were prepared from double distilled water. 0.1 M hydrochloric acid, 0.1% potassium periodate, 0.2% resorcinol 0.1% metal chlorides (cobalt, copper, nickel and iron), phosphate buffer pH-7.5 were prepared in distilled water, phosphate buffer was prepared by adding accurate volume of 0.2M sodium orthophosphate and solution were adjusted to required pH-value using 0.2M sodium hydroxide, acetate buffer was prepared by using exact volume of 0.2M sodium acetate and pH was adjusted to suitable value using 1M hydrochloric acid,

Also 1M solution of potassium chloride was used to adjust ionic strength of solution.

Preparation of Standard Solution of Drug

The standard solution of cilostazol was prepared by dissolving an accurately weight 100 mg of the cited drug in 100 ml of methanol. This stock solution was diluted with methanol to obtain working solution with concentration equals to 100 µg/ml.

Pharmaceutical Preparations

Twenty tablets containing cilostazol were weight and finely powdered. Accurately weighed portions of the powdered equivalent to 100 mg was dissolved in 50-ml methanol and mixed for 3-minutes, then filtered through whatmann filter paper No.42 and solution was completed to 100.0 ml with methanol and this solution was diluted with methanol to obtain solution of concentration of 100 µg/ml and analyzed with the same procedures as the pure drug.

3. Assay Procedures for Pure Drug

Method I

In a series of 10-ml test tubes, aliquots of drug solution equal to (0.1-2.0) ml with concentrations ranges equal to (1-20) µg/ml were transferred, followed by 1.5-ml of 0.1% resorcinol solution, 3.0 ml of 0.1M HCL followed by 2-ml of potassium periodate, the tubes were placed in water bath and heat at temperature equal to 50 C° for 15.0 minutes, the tubes were cooled to room temperature and the contents of all test tubes were transferred carefully to 10-ml measuring flasks and completed to the mark with distilled water and measure absorbance of formed complex against blank, prepared similarly, omitting drug, it is clear that the color complex has absorption maximum at 541 nm.

Method II

Aliquots of drug solution from (0.3-3.0) ml which were equal to (3-30) µg/ml were transferred into series of 10-ml test tubes and added 2.0 ml of 0.2% of solution of cobalt, nickel, cupper chloride, followed by 3.0 ml of phosphate buffer with pH equals 7.0 and 1.0 ml of , 1M potassium chloride for adjustment ionic strength, in case of iron chloride, added 2.0 ml of 0.2% ferric chloride followed by 3.5ml of acetate buffer with pH equals 4.0 and 1.5 ml of 1 M potassium chloride, the contents of all test tubes were heated in water bath for 30-minutes at temperature equal to 55°C , then the test tubes were cooled to room temperature, the contents of all tubes were transferred carefully to 10. ml measuring flask and completed to the mark with distilled water and measure absorbance of resulting complex, against reagent blank, prepared similarly, omitting drug, the formed complexes had absorption maximum at 572, 328, 504 and 536 for cobalt, copper, nickel and iron chlorides, respectively.

4. Recovery Studies

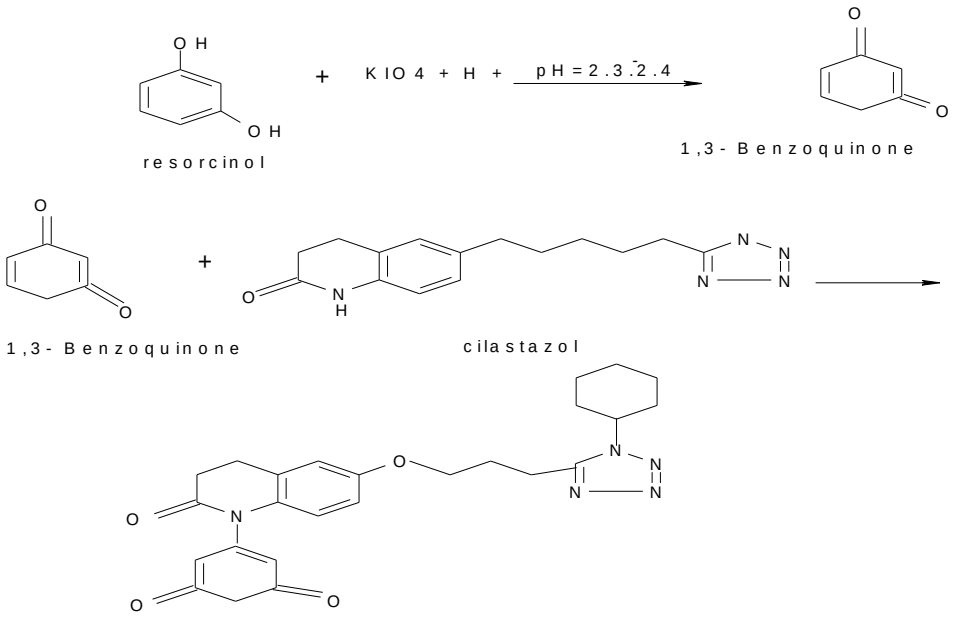
In order to test the accuracy and reproducibility of the proposed methods determined by investigating the recovery of the drug at concentration levels covering the specified range (three replicates of each concentration). The results showed excellent recoveries and precision. The proposed methods can be applied successfully to the cited drug in its pharmaceutical tablet forms.

RESULTS AND DISCUSSION

Method I

Principle of the color reaction

Cilostazol reacts with resorcinol in the presence of potassium periodate in acid medium forming red purple complex which absorbed maximally at 541 nm (Fig. 1). Scheme1 showed the probable reaction mechanism of cilostazol and resorcinol.



Scheme.1. reaction of cilostazol and resorcinol.

1. Effect of oxidizing agents

The effect of some general oxidizing agents on the color of the cilostazol-resorcinol complex was studied by adding 2.0 ml of 0.2% resorcinol solution and 3.0 ml of 0.1% different oxidizing agents like potassium periodate, potassium dichromate, potassium chromate, potassium iodide, N-bromosuccinamide, N-chlorosuccinamide, ferric chloride and ammonium ferric sulphate, 2ml of drug solution (100µg/ml and 2.5 ml of 0.1M hydrochloric acid were placed in 10.0 ml test tubes and were heated in water bath at 50°C for 15-minutes, then the test tubes were cooled to room temperature and carefully transferred to 25-ml volumetric flasks and were completed to the mark with distilled water, the absorbance of formed colored complex was measured at 541 nm, from the experimental data, it can be stated that potassium periodate was gave the best results with low blank absorbance value, which can be selected for the subsequent experiments (Fig.2).

2. Effect of acids

Various acids like hydrochloric acid, nitric acid, sulphuric acid, phosphoric acid, acetic acid and formic acid with different amounts had been investigated to examine their effects on the intensity of the color of the formed complex, from the results, it is clear that 3.0 ml of 0.1 M hydrochloric acid had gave the best and high absorbance of formed colored complex (Fig. 2).

3. Effect of pH

The effect of pH on the absorbance of the formed color complex was studied by transferred 2.0 ml of 0.2% resorcinol, 3ml of 0.1% potassium periodate, 2ml drug solution (100µg/ml) and different volumes of 0.1M HCL and 0.1M NaOH which were added separately and were placed in 10.ml test tubes and were heated at 50°C for 15 minutes, then the test tubes were cooled and their contents were transferred carefully to 25-ml volumetric flasks and were completed to mark with distilled water, from the given results, it was clear that the best results and the high absorbance of colored complex was obtained in acidic medium at pH ranging from (2.3-2.4) further the color of complex was stable, but in basic medium the color of complex was unstable and had low absorbance value.

4. Effect of cilostazol amounts to the reagent amounts

In this study different volumes of 0.2% resorcinol solution ranging from (0.3-3.0) ml and different volumes of cilostazol ranging from (0.5-3.0) ml of (100µg/ml) solution which were equivalent to (50,100, 150, 200, 250, 300) µg/ml, 3 ml 0.1% of potassium periodate, 3.0 ml of 0.1M hydrochloric acid were placed in 10.0 ml test tubes and were placed in water bath at 50°C for 15 minutes, then the tubes were cooled and their contents were transferred carefully to 25-ml volumetric flask and completed to mark with distilled water and

measure absorbance of formed colored complex at 541 nm, the experimental results showed that 1.5ml of 0.2% resorcinol and 2ml of cilostazol solution (100µg/ml) were given the good results and highest absorbance.

5. Effect of temperature

To study the effect of temperature on the absorbance intensity of the formed colored complex, the reaction was carried out at different temperature ranging from room temperature, 40, 50, and 60 for 10 minutes. The given results showed that the highest absorbance of the formed color complex was obtained at 50°C, the effect of heating time on intensity of color of complex was studied by carrying reaction at different time intervals ranging from (0, 5, 10, 15, 20, 25, 30), the results showed that the highest color intensity was obtained by heating reaction mixture at 50°C for 15 minutes.

6. Effect of surfactants

The effect of presence of surfactants on the intensity of the color of the formed complex were carried out by using different surfactants like cetylpyridinium chloride (CPC), cetyltrimethyl ammonium bromide (CTAB) (cationic surfactants), sodium dodecyl chloride (SDC) (anionic surfactants) and tween 80 (Nonionic surfactant), the results reveal that the use of surfactants had no effect on the intensity of the color of formed complex, which can be said that to eliminate the use of surfactants on the reaction mixture.

7. Order of addition

The high intensity of color and subsequent highest absorbance of formed complex was obtained by adding reaction contents by following order: cilostazolfollowed by resorcinol, then 0.1M hydrochloric acid and finally potassium periodate, then the reaction was heated at 50°C for 15 minutes cooled and measured absorbance of formed colored complex which had absorption maximum at 541 nm.

8. Stability of color of formed complex

In order to study effect of time on stability of color of complex, the absorbance of reaction was measured at different time intervals, which indicated that the color of formed complex was stable for at least two hours.

9. Effect of organic solvents

This effect was carried out using different organic solvents like, ethanol. methanol, dioxane, dimethylformamide, acetone and water, the experimental data showed that water was the best solvent used which was given the highest intensity and highest absorbance of the formed colored complex.

10. Nature of cilostazol-resorcinol complex

The nature of the formed cilostazol-resorcinol complex was studied by using jobs method of continuous variation and molar ratio methods in which concentration of cilostazol and resorcinol were equal and [cilostazol] = [resorcinol] = $5x10^{-3}$M, experimental data showed that the complex of cilostazol and resorcinol was formed by the ratio 1:1(Figs.3,4).

Method II

In order to determine optimum conditions of reaction, one parameter was varied and other parameters were remained constant and show the effect of this parameter on the absorbance of formed colored complex. The following are parameters which were affecting intensity of formed color complex.

1. Effect of volumes of reagent

This effect was studied by transferring to 10-ml test tubes 2.0 ml of cilostazol (100µg/ml) and different volumes 0.1% metal chloride ranging from (0.5-3.0) ml, then adding 3.0 ml of phosphate buffer with pH equals 7.0 in case of cobalt, copper and nickel, and 3.5 ml of acetate buffer pH=4.0 in case of iron chloride and 1.0ml, 1.5ml of 1M potassium chloride to adjust ionic strength of solution, these contents of test tubes were placed in water bath and heated at 55°C for 30 minutes, then test tubes were cooled to room temperature and their contents were transferred carefully to 10-ml measuring flasks and completed to the mark with distilled water and measured colored complex at absorption maximum at 572, 328,504 and 536 for cobalt, nickel, copper and iron, respectively experimental data showed that 2.0 ml of 0.1% metal chloride was which was given the best results and highest intensity and absorbance of colored complex.

2. Effect of type of buffer

To study the effect of the type of buffer, this can be done by using different type of buffers like universal buffer, phosphate buffer, acetate buffer, phthalate buffer, the results exhibit that phosphate buffer and acetate buffer were the best ones as they gives highest value of absorbance and highest intensity of color of formed complex for cobalt, nickel, copper and iron, respectively.

3. Effect of pH and volume of buffer

To study this effect on color of formed complex, taking different pH values ranging from (5, 5.5, 6.0, 6.5, 7.0, 7.5) of phosphate buffer and (2.5, 3.0, 3.5, 4.0, 4.5, 5.0) for acetate buffer experimental results showed that pH equals 7.0 and pH equals 4.0 are those which were given the highest color intensity of the formed complex for cobalt, nickel, copper, iron, respectively, to determine the effect of volumes of buffer on formed complex, using different volumes of suitable pH ranging from (0.5-4.0) ml, results showed that 3.0 ml of phosphate buffer pH-7.0 and 3.5 ml of acetate buffer pH=4.0 were the good ones to obtain highest values of absorbance of complex.

4. Effect of values of 1M potassium chloride

This effect was studied by using different volumes of 1M potassium chloride ranging from (0.5-3.0) ml, the results showed that 1.0 ml, 1.5 ml of 1 M potassium chloride were the best to give good results and adjusted ionic strength for reaction.

5. Determination of the stoichiometric ratio of the formed complexes.

The stoichiometric ratio of the complexes formed between cilostazol and cobalt, nickel, copper, iron chlorides were studied using jobs method of continuous variation and molar ratio methods.

Procedures for continuous variation method

The procedure described by Obradovic et al [23] was adopted. $3x10^{-2}$M metalchloride and $3x10^{-2}$cilostazol, solutions of (0-1.0) ml of $3x10^{-2}$M of metal chloride and solutions of (1.0-0) ml of $3x10^{-2}$M of cilostazol were transferred into 10-ml test tubes, keeping the mole fraction of solution constant, then added, 3.0 ml phosphate buffer (pH-7.0) and 3.5ml acetate buffer pH 4.0 for cobalt, nickel, copper and iron, respectively, 1.5 ml of 1M potassium chloride for ionic strength.

The test tubes were heated at 55°C for 30 minutes in water bath and cooled to room temperature and their contents were carefully transferred to 10-ml volumetric flasks and completed to mark with distilled water and measure color of complex at maximum absorbance at 572, 328, 504, 536 for cobalt, nickel, copper and iron, respectively. The results showed that the complexes of cilostazol and metal chlorides were formed by the ratio 1:1.

Procedures for molar ratio method

The procedures described by Vardo and Dauason[24], was adopted. 2.0 ml of $3x10^{-2}$M of metal chloride and different volumes of cilostazol ranging from (0.5-4.0) ml of $3x10^{-2}$M, then the procedures were completed as described under continuous variation, experimental data exhibits that cilostazol-metal chlorides complexes were formed by the ratio of 1:1. (Fig 2)

6. Effect of temperature and heating time:

In order to study this parameter, the reaction was carried out at different temperature (30, 35, 40, 50, 55, 60) at different time intervals (5, 10, 15, 20, 25, 30, 40, 50, 60), the color of formed complex was measured at maximum absorbance, the results showed that heating reaction at 50°C for 30 minutes gives best results.

Validation of the proposed methods

a. Precision

The intra and inter-day precision values were calculated from three concentrations of cilostazol, the RSD values were less than 1.0% indicating that the proposed methods were precise.

b. Accuracy

The accuracy of proposed methods was established by recovery studies. Table.2. results showed that recovery of cilostazol by proposed methods was satisfactory as RSD values were less than 1.0% and mean recoveries between 98.0% -102.0%.

c. Ruggedness and Robustness

The method of robustness and ruggedness was determined by changing the following parameters: pH values in the range of (6.0-7.4) or (3.0-4.0), temperature of heating from (50°C-55°C) and time of heating (20-25) minutes, experimental results indicated no significant change in the absorption maximum and its value of formed colored complex by changing these parameters.

d. Study of interferences

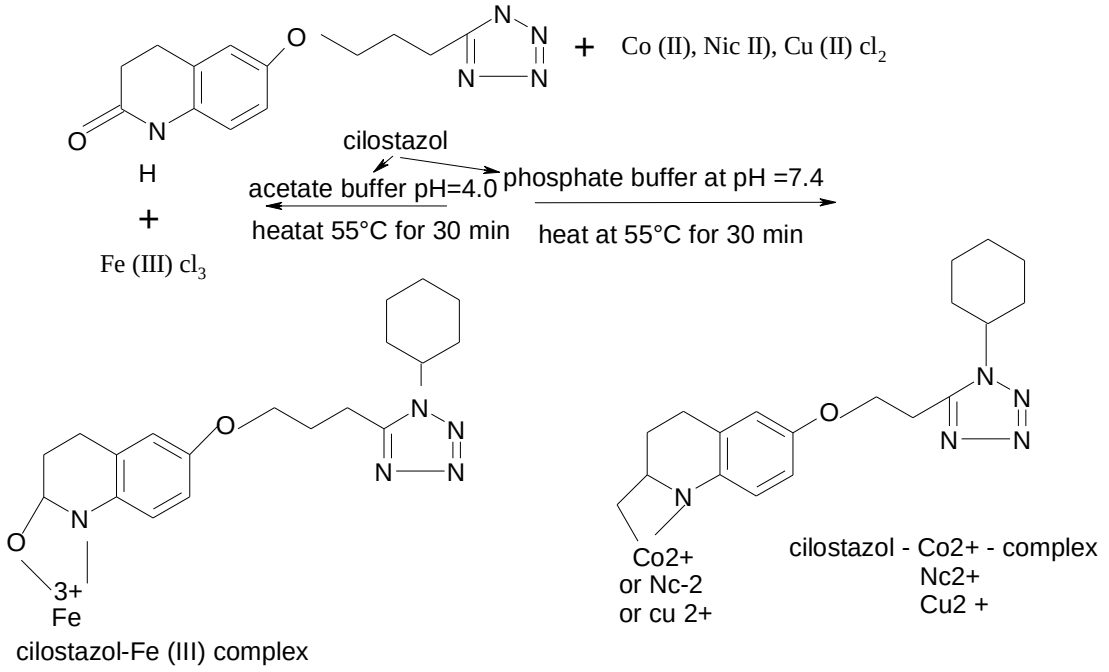
In order to assess the possible analytical application of the present proposed methods, the effect of foreign compounds like lactose, starch, dextrose, glucose, magnesium stearate and cellulose had been studied, the obtained results had been revealed that there is no significant effect of interfering of these excipients with the proposed methods.

Application of the proposed methods to pharmaceutical preparation

The developed methods had been applied to the pharmaceutical preparations (tablets), the results obtained are shown in table.3. And were compared with official method HPLC method, F-test and T-test were calculated for six degrees of freedom at 95.0% confidence limits.

CONCLUSION

The proposed visible spectrophotometric methods are novel, simple, accurate, rapid and sensitive and can be used for routine quality control and quality assurance laboratories in pure as well as in pharmaceutical preparations.



Scheme. 2: Probable mechanism for complexation of cilostazol with Co^{+2}, N^{+2}, Cu^{+2} and Fe^{+3}chlorides

Table 1: Analytical parameters for determination of cilostazol by the proposed spectrophotometric methods

Parameter	1st method oxidative coupling with resorcinol	2nd mothd Complexation of cilostazol with cobalt, nickel, copper in phosphate buffer and with iron buffer in acetate.			
		Co^{+2}	Ni^{+2}	Cu^{+2}	Fe^{+3}
L_{max}	541 nm	572	328	508	536
Color of complex	Red-pink	Violet	Yellowish Green	Green	Violet-pink
Beers law limits µg/ml	(10-25) µg/ml	(5-20)	(1-20)	(3-30)	(5-25)
Ring bon limits µg/ml	(10-25) µg/ml	(8-17)	(5-18)	(5-25)	(7-25)
Molar absorptivity € $L.mol^{-1}.Cm^{-2}$	$3.55x10^3$ $L.mol^{-1}.Cm^{-2}$	$1.98x10^3$	$2\text{-}11x10^3$	$2.27x10^3$	$2.31x10^3$
Sandell sensitivity µg/cm2	0.115	0.212	0.201	0.234	0.137
Limit of detection (LOD)	0.99	0.056	0.068	0.043	0.037
Limit of quantitation (LOQ)	0.173	0.105	0.104	0.133	0.151
Regression equation slope (a)	0.321	0.218	0.323	0.228	0.213
Intercept (b)	0.00111	+0.00212	0.00121	0.0033	0.0012
Relative standard deviation RSD	0.78	0.68	0.37	0.49	0.80
Correlation – coefficient (r)	0.9999	0.9998	0.9997	1.001	0.9996

Table 2: Evaluation of accuracy and precision of determination of cilostazol by 1st proposed method

Table µg/ml	Found µg/ml	Recovery % ±SD	RSD%	S.E%	Confidence limits ±SD (at p= 0.05)
Inter-day precision					
5	5.01	100.01±0.37	0.85	0.199	5.01±0.27
10	10.02	100.02±0.47	0.93	0.199	10.02±0.44
15	14.99	99.99±0.58	0.68	0.066	14.99±0.38
20	19.97	99.97±0.43	0.98	0.150	19.97±0.51
25	24.99	99.99±0.50	0.72	0.04	24.99±0.60
Mean:		99.99±0.47	0.83	0.114	14.99±0.44
Intra-day precision					
5	4.98	99.98±0.43	0.98	0.40	4.98±0.33
10	9.95	99.93±0.37	0.90	0.502	9.95±0.47
13	13.01	100.10±0.36	0.73	0.076	13.01±0.38
20	19.98	99.98±0.71	0.81	0.1	19.98±0.61
25	25.05	100.05±0.62	0.62	0.199	23.03±0.42
		100.0±0.498	0.808	0.265	14.19±0.442

Table 3: Evaluation of accuracy and precision for determination of cilostazol by 2nd proposal method

Method chloride	Taken µg/ml	Found µg/ml	Recovery% ±SD	RSD%	S.E%	Confidence limits ± 0.05
Inter-day precision						
cobalt (II)	5	4.98	99.99±37	0.85	0.4	4.98±0.41
	10	9.97	99.97±0.60	0.55	0.3	9.97±0.33
	15	14.99	99.99±0.58	0.62	0.066	14.99±0.53
Mean			99.98±0.51	0.67	0.21	9.98±0.42
Intra-day precision						
	5	5.01	100.01±0.53	0.65	0.08	5.01±0.44
	10	10.05	100.05±0.67	0.45	0.118	10.05±0.66
	15	15.02	100.02±0.33	0.50	0.202	15.02±0.57
Nickel (II)	10	9.99	99.99±0.48	0.68	0.121	9.99±0.48
	15	15.05	10..05±0.53	0.33	0.218	15.05±0.53
	20	19.98	99.98±0.83	0.78	0.105	19.98±0.68
Mean			100.02±0.81			
Intra-day precision						
	10	10.02	100.0±0.44	0.39	0.09	10.02±0.48
	15	13.03	100.03±0.34	0.46	0.218	13.03±0.54
	20	20.03	100.05±0.40	0.77	0.151	20.05±0.33
Inter-day-precision						
Cupper (II)	10	9.97	99.97±0.53	0.85	0.101	9.97\±0.55
	15	14.95	99.95±0.42	0.95	0.122	14.95±0.45
	20	25.05	100.05±0.39	0.79	0.138	25.05±0.61
Mean						
Intra-day precision						
	10	10.05	100.05±0.32	0.67	0.09	10.05±0.47
	15	14.95	99.95±0.41	0.71	0.105	14.95±0.55
	25	24.96	99.96±0.48	0.33	0.122	24.96±0.60
Inter-day precision						
Fe (III)	10	9.99	99.99v0.57	0.75	0.107	9.99±0.47
	12	11.98	99.98±0.60	0.59	0.202	11.98±0.61
	14	14.01	100.01±0.66	0.68	0.171	14.01±0.70
Mean						
Intra-day precision						
	10	9.98	99.98±0.42	0.58	0.113	9.98±0.44
	12	11.97	99.97±0.53	0.48	0.212	11.97±0.58
	14	13.99	99.99v0.45	0.66	0.119	13.99±0.37
Mean						

Table 4: Determination of cilostazol in tablets using the proposed methods compared with reference one (HPLC)

Pharmaceutical Preparation	% Recovery ± S.D [(a)] 2-nd method					
	1-st method	Co(II)	Ni(II)	Cu (II)	Fe (III)	Reference method (HPLC)
	99.98±0.68	100.01±0.37	100.05±0.55	99.98±0.72	99.99±0.80	99.95±0.65
	t-test-1.36(2.306)	2.11(2.306)	1.97(2.306)	2.18(2.306)	1.51(2.306)	
	F-test= 1.08 (6.39)	0.94(6.39)	2.09 (6.31)	3.59(6.39)	4.34(6.39)	

a = mean ± standard deviation for five determinations.
(2.306) tabulated value for t-test for p = 0.05 and eight degrees of freedom
(6.39) tabulated value for F-test for P=0.05 and eight degrees of freedom.

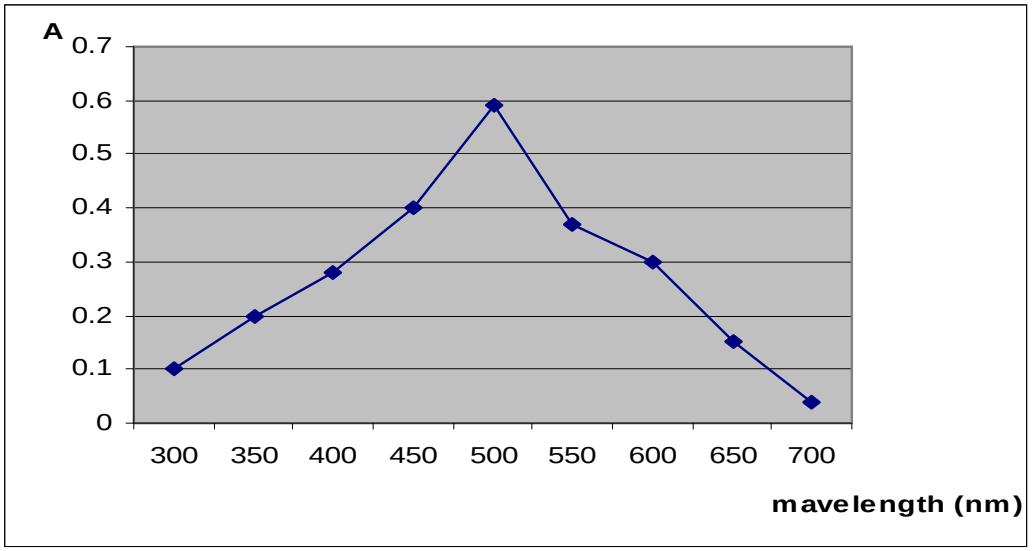


Fig. 1: Absorption spectrum of reaction of cilostazol and resorcinol

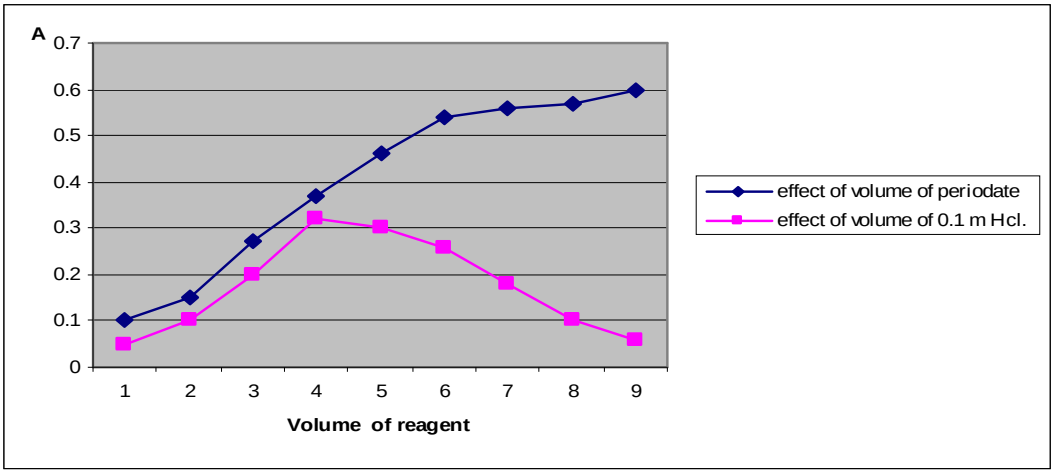


Fig. 2: Effect of volume of periodate and volume of HCl on reaction of cilostazol and resorcinol

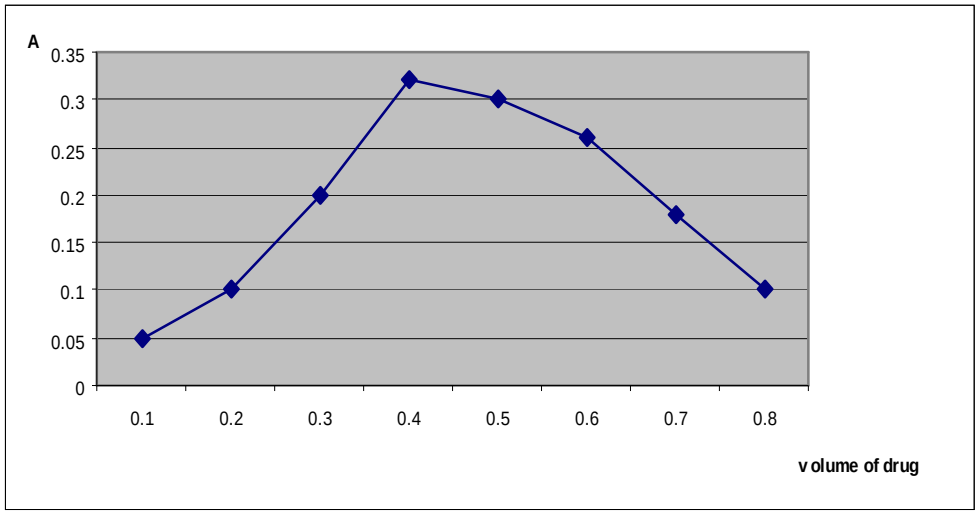


Fig. 3: Continuous variation of reaction of cilostazol with resorcinol

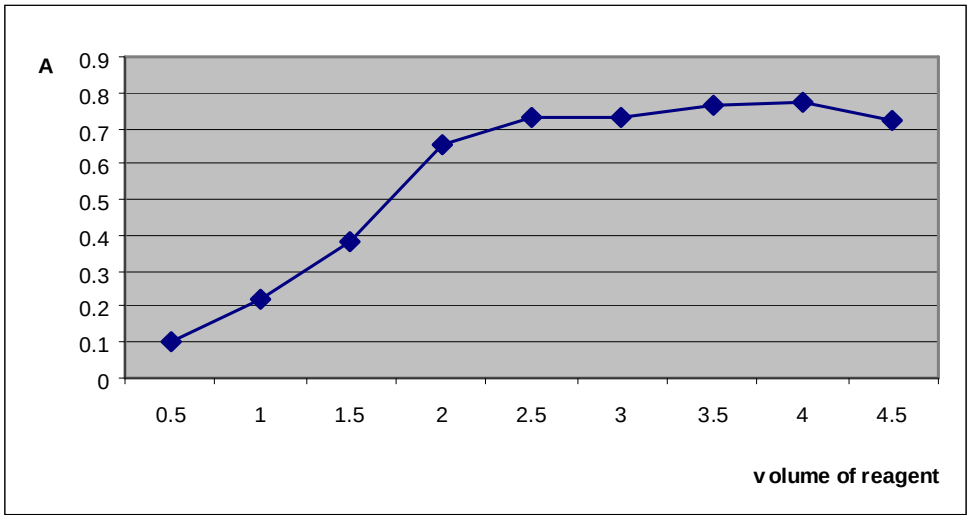


Fig. 4: Molar ratio of cilostazol with resorcinol

REFERENCES

1. United States pharmacopeia 2009 (USP 32 NF 27). United States pharmacopeial convention, rockirlle, Mocyland, USA, 2009.
2. Woo SK, Kang WK and Kwonk I. Clin pharmcol Ther. 2002;71(4):246-252.
3. Sekhar VR, Divya A, geethagss and Ramalingam P. Int J Med Chem Anal. 2013;3(1):22-26.
4. Basmiwal PK and Vinesh Kumar. Trop J Phoim Res. 2010;9(5):499-503.
5. Anwor A wassel, Amin AS. Ahmed 1S, Dessouki HA and Henduray Ham. Anal Bioanal Electrochem. 2012;4(2):197-211.
6. Jinjin Wang, Zhibin WANg, Chen Chen and Zhiyi Wang. Lat Am J Pharm. 2012;31(21):240-244.
7. Jafren Jamal Joti and AhsanulHaque UD. 1J NDD. 2011;3(2):143-148.
8. Kyu-Jeong Yean and Young-Jeou Park. J Liq Chrom Rel Technol. 2004;27(16):2603-3612.
9. Dramer SL, Tata PNV, Vengurbkar SS and Brisson JH. J Pharm Biomed Anal. 2001;26:637-650.
10. Tata PN, Fu CF and Bramer SL. J Pharm Biomed Anal. 2001;24:381-389.
11. Fucj, Tata PN, Okadak, Akiyama H and Braver SL. J Chromatogr B

Biomed Sci Appl. 1999; 728(2):251-262.
12. Tat PN, FucJ, Broundeer NJ, Chour PC and Bramer SL. J Pharm Biomed Anal. 1998;18:441-451.
13. Kurien J and Jayasekhar P. Int J Pharm Bio Sci. 2014;5(1):176-186.
14. Uslu B and OZKau SA. Anal Chim Acta. 2002;49:462.
15. Radi A and El-Sherif Z. Talanta. 2002;58:319.
16. Nigovie B and Simmunie B. J Pharm Biomed Anal. 2005;38:162.
17. Tapsoha I, Belgaied J and Boujlel K. J Pharm Biomed Anal. 2005;38:62.
18. Radi A. Talanta. 2005;65:271.
19. Amhrosi A, Antiochia R, Companella L, Dragone R and Lavagnini I. J Hazard Mater. 2005; 122:219.
20. Ni Y, Wany Y and Kakot S. Talanta. 2006;69:216.
21. Tella AC, Obaleye AJ and obiyenwa GK. J pharm Res. 2011;4(1):241-244.
22. Sarsam LA and Raf. J Sci. 2013;24(1):128-145.
23. Obradovic UV, Mitic SS, Tasic S, Pavlaric AN. J Serh Chem Soc. 2005;70(4):651-659.
24. Narda JV and Dauson JH. Inorg Chim Acta. 1986;123:9-13.