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Simple Indirect Spectrophotometric Determination of Amoxicillin in Pharmaceutical Preparations

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Abstract
A simple, rapid and sensitive indirect spectrophotometric method has been described for the determination of Amoxicillin in both pure form and pharmaceutical preparations. The method is based on the reaction of well-known oxidizing agent potassium permanganate with amoxicillin, and then disappearing of violet color of permanganate can be monitored to be proportional to the amount of amoxicilline in the pharmaceutical preparations. The optimum experimental conditions for the reaction have been studied carefully. It was observed that the proposed method has many considerable and reliable figures of merits. Beer's law is obeyed from 0.1-1.6 $mg.dL^{-1}$. The detection limit was 0.04$mg.dL^{-1}$ and the recovery was about 98$mg.dL^{-1}$. Based on the obtained figures of merits, the proposed method is well-suited for the determination of amoxicillin in different pharmaceutical preparations and routine analysis.
Keywords: Determination, Amoxicillin, Spectrophotometry, Potassium Permanganate.

Introduction

Amoxicillin is a moderate-spectrum, bacteriolytic, β-lactam antibiotic used to treat bacterial infections caused by susceptible microorganisms, figure 1. Due to its better absorption following oral administration, amoxicillin is usually the drug of choice than other β-lactam antibiotics.
Amoxicillin is bactericidal against susceptible micro-organisms through the inhibition of biosynthesis of cell wall mucopeptide during bacterial multiplication. (Amin 1994 and Nagaralli 2002).

Pharmaceutical analysis is one of the most important fields in analytical chemistry. The discovery of new drugs and the on-going update of international regulations for the safety and efficacy of pharmaceutical formulations demand the continuous development of new analytical methods. Inevitably, automation plays an important role, especially when a lot of samples have to be analyzed in the minimum of time.(Paraskevas 2007)

In literature survey there are several methods which can be used for assaying amoxicillin inbiological fl uids, drug substances, formulation products . Potentiometric method, (Sutherland 1972) Liquid chromatography (Simarpreet 2011, British pharmacopoeia Commission London 2009), HPLC (Kowalczuk 2012,Mascher 1998, Riediker 2001), RPHPLC (Ahmed 2011,Shaheen 2014), spectofluorimetry (Kamruzzaman 2012 , Kemal 2008), Uv-vis spectrophotometry (Nagaralli 2002,Amin 1994, M.B. Devani 1992 , H.D. Revanasiddappa1999), FIA (Bery 2015,M. Q .Al-Abachi 2009)..

In the present work, simple and sensitive indirect spectrophotometric determination of amoxicillin is described using potassium permanganate as a reagent and oxidizing agent.

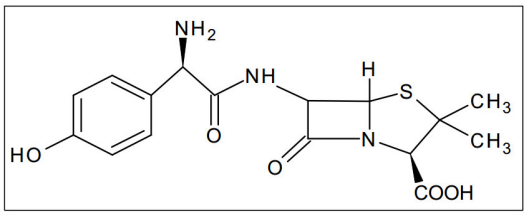

Figure1. Chemical structure of Amoxiciline

Experimental

Instruments

All absorbance spectra were recorded with (Helιasα, UV-Visible spectrophotometer, V4.6) and APEL Spectrophotometer (APEL PD 303 Visible spectrophotometer, Japan) using 1 cm quartz cell.

Chemicals

All solvents and reagents were of analytical grade unless indicated otherwise. Solutions were prepared with deionized water. Potassium Permanganate (Sigma-Aldrich, 99 %,). Amoxicilline was purchased from Samarra

pharmaceutical Manufacturer, Iraq.

Stock solutions

Stock solution of amoxicillin was prepared by dissolving 5 mg of pure form of amoxicillin in 100 mL water to give 5mg/dL. Potassium permanganate 1mM was prepared in deionized water. 0.1M sodium hydroxide (NaOH, Sigma-Aldrich, 98%) and 0.1 H_2SO_4 (H_2SO_4, Fisher scientific, 96.2%) were prepared in deionized water.

Pharmaceutical preparations and drugs

Twenty tablets or the contents of 20 capsules or any contained tablets were weighed and ground into a fine powder. An accurately weighed amount Powder equivalent to 0.005gm of both drugs was transferred quantitatively to dissolved in 50 mL of water, The contents were transferred into a 100 mL volumetric flask, made up to the mark with water. By using 3ml of both drug solution and 5ml $KMnO_4$ are mixed and read absorbance for each solution sample at (λ_{max} =525 nm) of AMX and repeated reading five times.

Preliminary Procedure

Amoxicillin standard solution was mixed with $KMnO_4$ solution in acidic medium. The Absorbance of the resulting solution was recorded at two wavelengths, 380 nm and 525 nm. The serial solutions and samples were all recorded in both wavelengths. Figure (2) shows the absorption maxima of both Amoxicillin reacted with potassium permanganate (dotted line) and with $KMnO_4$ alone (solid line). In terms of quantitative monitoring both wavelengths could be used for analysis, however, the more sensitive wavelength is the one which can be preferred. In our case, 525 nm is the more sensitive wavelength.

Results and Discussion

Potassium permanganate ($KMnO_4$) plays a major role in determination of many chemicals acting as oxidizing agent (M.PATGAR 2014).

Optimization of reaction variables

In order to obtain the best sensitivity, the solutions were read at two wavelengths; the maximum wavelength of $KMnO_4$ (525 nm), and the best wavelength of the reaction products of permanganate with Amoxicillin (380 nm). Figure 2 shows the spectra of $KMnO_4$ and both Amoxicillin and $KMnO_4$. All other variable parameters were optimized, like; reaction time, temperature, pH of the reaction, and all are tabulated in Table 1.

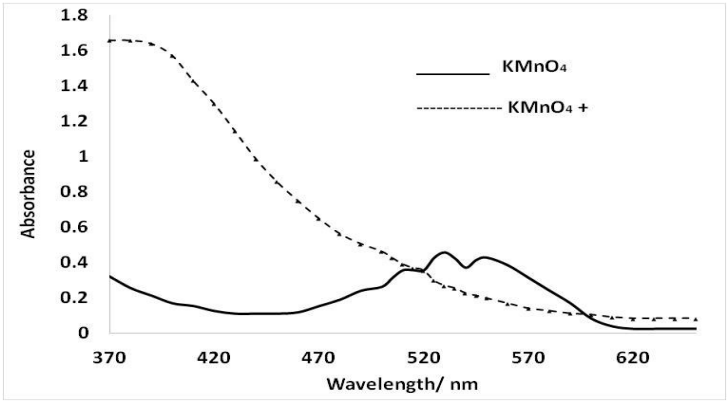

Figure2. Absorption spectra of solution 3ml Amoxicillin with 5ml $KMnO_4$ (dotted line) and for 3ml $KMnO_4$ without Amoxicillin (Solid line).

Table1. Optimized Variable Parameters

Parameters	Best
λ_{max}	525 nm
pH	< 3.0
Temperature	25 ^{0}C
Reaction time	2 min

Standard Procedure

Calibration Curve, Accuracy, Precision and Sensitivity

Standard curve was produced as shown in figure (3) between Absorbance verse concentrations of amoxicilline, it was observed that the line was in a linear relation in the ranges of 0.1-1.6 mg.dL^{-1} at λmax at 525nm. The molar absorpitivity (ε) was 4.6 X 10 ^{5} L.mol^{-1}.cm^{-1}. The relative standard deviations of the proposed method were close to 0.9% (n=7), which indicate the proposed method has a very good reproducibility of the results (high precision).

LOD was calculated based on standard deviation of response and the slope of calibration curve. The limit of detection was expressed as:

D.L. = 3 σ /slope

Where σ is the standard deviation of intercept, S is the slope of calibration curve. Based on our experimental data, the LOD was **0.04** mg.dL^{-1}.

While **LOQ** was calculated based on standard deviation of intercept and slope of calibration curve:

D.L. = 10 σ /slope

Based on the experimental calculation, the LOQ was **0.12** mg.dL^{-1}.

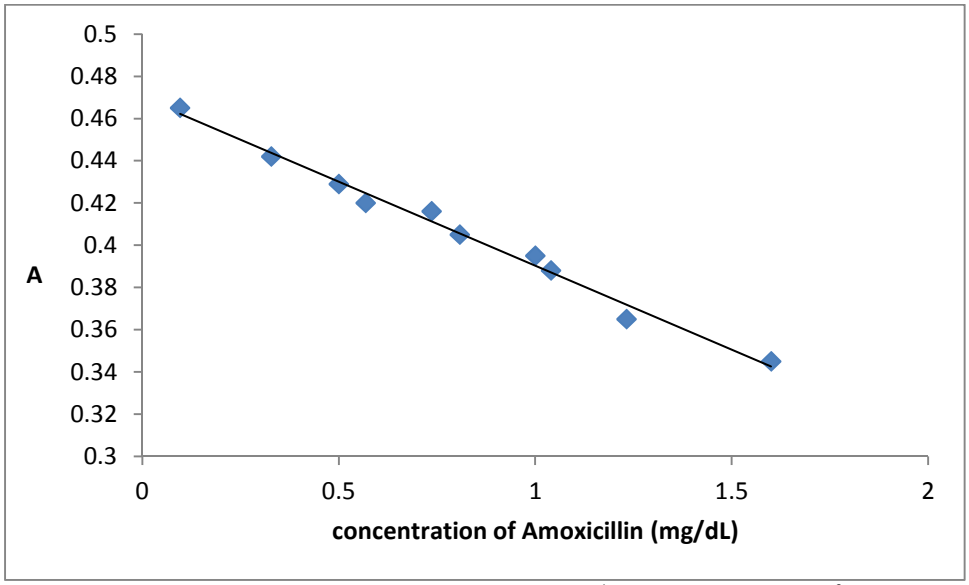

Figure3. Calibration curve of series solution from 5 mg.dL^{-1} of AMX with (1×10^{-3} M) of $KMnO_4$.

Table 2. Parameter of Calibration Curve

Parameters	results
Linear range	0.1-1.6 mg.dL^{-1}
RSD	0.96 (n=7)
Correlation coefficient	0.990
LOD	0.04 mg. dL^{-1}
LOQ	0.12 mg. dL^{-1}
E%	(0.142-1.23)%

Determination of Amoxicillin in Pharmaceutical Preparations

The proposed method was applied to the determination of Amoxicillin in pharmaceutical preparations (tablets and capsule) available in pharmacies and drug stores in Kurdistan region-Iraq in table 3.

Twenty tablets or capsules were weighed and ground into a fine powder. An accurately weighed amount powder equivalent to 0.005gm was transferred quantitatively to dissolved in 50 mL of water, The contents were transferred into a 100 mL volumetric flask, made up to the mark with water. By using 3ml of the solution and 5ml $KMnO_4$ are mixed, waited for 40 minutes and absorbance was recorded at 525 nm.

Table 3. Analysis of Drugs Formulation:

sample	Theoretical weight(mg)	Weight all Capsules (mg)	Sample ± S.D (mg/dL)	% recovery
Amoxicillin (Iran) farabi pharmaceutical (10 capsules)	500	6261	0.8453 ± 0.002729	99.6
Largopen capsule (turkey) (20 capsules)	500	13772	0.6544 ± 0.001303	99.3
Largopen tablet (turkey) (16 tablets)	1000	21729	1.2053 ± 0.00041	98.8
Amoxicillin capsule BP(united kingdom) Bristol(21 capsules)	500	11901	0.68033 ± 0.000168	99.3
Amoxicillin capsule BP(UK Ireland) (POM) (21 capsules)	500	11909	$0.7233 \pm 5.77 \times 10^{-5}$	99.8
Amoxicillin capsule England PL holder & manafu (21 capsules)	500	12182	$0.9201 \pm 1.41 \times 10^{-6}$	99.9
Amoxil(Glaxo simith kline) (10 capsules)	500	5972	0.58259 ± 0.00155	99.5

Conclusion

Simple, accurate, and sensitive method was developed for the quantitative determination of amoxicillin in some pharmaceutical preparations available in Kurdistan region-Iraq. The method was based on indirect spectrophotometric determination of amoxicillin after its reaction of a well-known strong oxidizing agent potassium permanganate, then measuring the absorbance at wavelength of 525 nm. With the proposed method, good analytical values were obtained from the assays of determination of amoxicillin in pharmaceutical preparations. The high value of merits with simplicity in instruments and chemicals suggest the utility of the proposed method in routine analysis.

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