



Spectrophotometric Determination of Chloramphenicol by Oxidative Coupling Reaction with Naphthalene-1, 5-Diamine in the Presence of Potassium Iodate

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ABSTRACT

A simple, accurate, and sensitive method for the spectrophotometric determination of Chloramphenicol (CAP) in pharmaceutical preparations. The method is based on the oxidative coupling reaction of Chloramphenicol after reducing the nitro group in the drug into an amino group with Naphthalene-1,5-diamine as a reagent in the presence of potassium Iodate as an oxidizing agent formed violet dye that is soluble in water and have a maximum absorption at 568nm. Beer's law is obeyed in the concentration range of 6-27 $\mu\text{g}.\text{mL}^{-1}$ with a molar absorptivity of $0.4805 \times 10^4 \text{ L}.\text{mol}^{-1}.\text{cm}^{-1}$, and Sandell's sensitivity was $0.061 \mu\text{g}.\text{cm}^{-2}$, respectively. The correlation coefficient was 0.9994, with recovery average % was 100.05. Detection limit (D.L) and quantitative limit (Q.L) were $0.241 \mu\text{g}.\text{mL}^{-1}$ and $0.804 \mu\text{g}.\text{mL}^{-1}$. The product was stable for 70 minutes with relative error (RE) % of -0.12 to -0.22 and a relative standard deviation (RSD) % of 0.476 to 0.362. The method was successfully applied to the analysis of Chloramphenicol in pharmaceutical preparations (Eye drop).

Introduction

Chloramphenicol (CAP) is 2,2-dichloro-N-[(1R,2R)-2-hydroxyl(hydroxymethyl)-(4-nitrophenyl) ethyl] acetamide freely soluble in methanol, ethanol, butanol, ethyl acetate, and in propylene glycol. Slightly soluble in water, and ether, and insoluble in benzene, and petroleum ether. Its chemical structure is $\text{C}_{11}\text{H}_{12}\text{Cl}_2\text{N}_2\text{O}_5$ and molecular weight is 323.1 $\text{g}.\text{mol}^{-1}$ [1]. CAP is an antibacterial compound, originally derived from Venezuela bacterium isolated by the scientist David Gottlieb, introduced as a drug in 1949 [2]. It is a broad-spectrum antibiotic used in medicine and veterinary medicine as a food additive to improve growth, it is very effective as it can eliminate many types of microorganisms, it is an inhibitor of the growth and reproduction of very microorganisms through its effectiveness in inhibiting protein biosynthesis, and on the other hand, it also has toxic effects on human organisms [3-4]. It has

also been widely used as an antiseptic to prevent disease or as a chemotherapeutic agent to fight diseases, that likely it's causative in aplastic anemia and is the condition in which the bone marrow cannot produce enough new cells to regenerate blood cells [5-7]. The structural formula of CAP is shown in Figure (1).

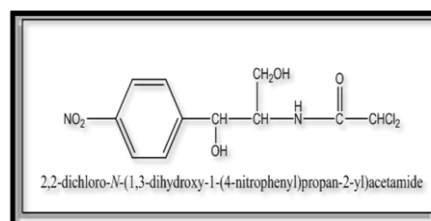


Fig. 1: Chemical Structure of Chloramphenicol

Chloramphenicol was estimated by different analytical methods, such as Spectroscopic methods

[8-13], high-performance liquid chromatography [14-16], chemiluminescence [17-19], and electrochemical [20-22]. In this paper a new method was developed for the reduction of the nitro group of Chloramphenicol (CAP) drug by concentrated hydrochloric acid and zinc powder and then reaction with Naphthalene-1,5-diamine as a reagent to form violet dye and measured the absorbance of the colored product by using a UV-visible Spectrophotometer.

Materials and Methods

Instrument:

UV-visible double beam (T92+ Spectrophotometer, China) with 1cm matched quartz cells.

Reagent:

All chemicals used in this study were of a high degree of purity. A pure Chloramphenicol and pharmaceutical formulation (eye drops) were kindly provided by state company for Drug Industries and Medical appliance-(SDI) Sammara-Iraq. Naphthalene-1,5-diamine, 1-Naphthylamine-4-sulfonic acid, Sulphanilic acid, and P-aminophenol were obtained from Fluka. Potassium Iodate, Potassium persulfate, Ammonium persulfate, and Ammonium ferric sulfate were obtained from BDH. Zinc powder (Zn) and hydrochloric acid are from Poueler.

Chloramphenicol Standard Solution (CAP) 1000 $\mu\text{g/ml}$ ($4.3 \times 10^{-3} \mu\text{g/ml}$):

The solution was prepared by dissolving 0.1 g of pure Chloramphenicol powder in distilled water in a beaker, then the solution was placed in a water bath for 5 minutes at a temperature of 50 °C with stirring. After cooling the solution was placed in a volumetric flask of 100ml and completing the volume to the mark with the same solvent.

Chloramphenicol Reduced Solution (CAPR) 150 $\mu\text{g/ml}$

The solution was prepared by taking 15ml of Chloramphenicol Standard Solution 1000 $\mu\text{g/ml}$ in a volumetric flask of 100ml, then 0.3g of zinc powder and 15ml of concentrated hydrochloric acid (11.8 N), were added. The mixture was shaken for 20 minutes, to complete the reduction process and then completed the volume to the mark with distilled water.

Solution of Potassium Iodate:

This solution was prepared by dissolving 0.428g of potassium Iodate powder ($2 \times 10^{-2} \mu\text{g/ml}$) in distilled water in a 100ml volumetric flask and then completing the volumetric flask to the mark with the same solvent.

Hydrochloric Acid (1 M):

This solution was prepared by mixing 8.5 ml of hydrochloric acid (11.8 N) with distilled water in a 100 ml of volumetric flask and filling the flask to the mark with distilled water.

Solution of Naphthalene-1,5-diamine ($3 \times 10^{-2} \mu\text{g/ml}$):

This solution was prepared by dissolving 0.474g of Naphthalene-1,5-diamine in amount of distilled water

in a volumetric flask with a capacity of 100 ml and then completing the volume to the mark with the same solvent.

Pharmaceutical Preparation of Chloramphenicol:

Samaphenicol (eye drops): Each 1 ml contains 50 mg of Chloramphenicol was prepared by diluting 1ml of eye drop with an amount of distilled water and then this solution was transferred to a volumetric flask with a capacity of 100ml and completed volume to the mark with distilled water to obtain a concentration of 150 $\mu\text{g/ml}$.

General process:

1ml of Naphthalene-1,5-diamine (0.03 $\mu\text{g/ml}$) as reagent was added to CAPR solution (150 $\mu\text{g/ml}$) in 25 ml volumetric flask and 1ml of potassium Iodate (0.02 $\mu\text{g/ml}$) as oxidizing agent and completed the volume with distilled water to the mark, were violet dye was formed.

Preliminary study:

It was observed that when CAPR solution was mixed with the reagent solution Naphthalene-1,5-diamine in the presence of potassium Iodate, a violet dye is formed. Therefore, the optimum conditions for the coupling reaction were studied to obtain the best possible results to develop a simple and sensitive spectrophotometric method for the determination of Chloramphenicol.

Results and discussion

The Ultimate Absorption:

After reaching the optimum conditions, use 1ml of CAPR solution of 150 $\mu\text{g/ml}$, 1ml of reagent solution of Naphthalene-1,5-diamine of $3 \times 10^{-2} \mu\text{g/ml}$, 1ml from as an oxidizing agent potassium Iodate solution of $2 \times 10^{-2} \mu\text{g/ml}$ at room temperature, and left the solution for 10 minutes for the completion and stability of the reaction after completing 25ml volumetric flask with distilled water to the mark, the final absorption spectrum for the dye formed was measured against reagent blank solution, and it was observed that the maximum absorption at 568nm, whereas the blank solution has no absorption in this region. As shown in Figure (2).

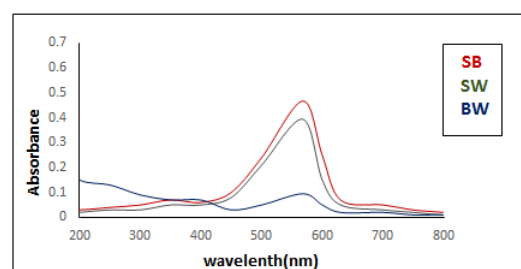


Fig. 2: Final absorption spectrum for the determination of CAPR by reaction with Naphthalene-1,5-diamine in the presence of Potassium Iodate

SB: It represents the absorption spectrum of the product formed versus the blank solution.

SW: It represents the absorption spectrum of the product formed versus distilled water.

BW: Represents the absorption spectrum of the blank solution versus distilled water.

Study of optimal conditions:

Subsequent experiments were carried out using 1 ml of the CAPR solution with a concentration of 150 $\mu\text{g/ml}$, 1 ml of reagent Naphthalene-1,5-diamine, 1 ml of the oxidizing agent solution potassium Iodate and distilled water, in a final volume of 25ml. The absorbance was measured versus blank solution, and the formed dye gave a maximum absorption at 568nm.

Choosing the best coupling reagent: 1 ml of each of the used reagents solutions were taken at a

concentration of $3 \times 10^{-2} \mu\text{g/ml}$, 1 ml of the oxidizing agent solution potassium Iodate with a concentration of $2 \times 10^{-2} \mu\text{g/ml}$, 1ml of CAPR solution 150 $\mu\text{g/ml}$ and distilled water. The results listed in Table (1), showed that the Naphthalene-1,5-diamine reagent gave a maximum absorbance of the dye formed at 568 nm against the blank solution, the blank solution did not show any absorption at this wavelength, so this reagent was chosen in the subsequent experiments.

Table 1: Choosing the best reagent

Reagents (1×10^{-2}) M	V Variable	A Absorbance	$\lambda_{\text{max}}(\text{nm})$
Naphthalene-1,5-diamine	SB	0.467	568
	BW	0.095	510
1-Naphthylamine-4-sulfonic acid	SB	0.381	490
	BW	0.084	435
Sulphanilic acid	SB	0.256	462
	BW	0.072	428
P-aminophenol	SB	0.178	485
	BW	0.064	410

SB: It represents the absorption spectrum of the product formed versus the blank solution.

BW: Represents the absorption spectrum of the blank solution versus distilled water.

Impact of Changing the Concentration of Reagent:

When different concentrations of reagent Naphthalene-1,5-diamine were added to 1ml of CAPR solution 150 $\mu\text{g/ml}$ and 1ml of the oxidizing agent solution potassium Iodate with a concentration of $2 \times 10^{-2} \mu\text{g/ml}$ and then distilled water. The maximum absorption was observed at concentration $3 \times 10^{-2} \mu\text{g/ml}$. The results were listed in Table (2).

Table 2: Impact of Changing the Concentration of Reagent

M of Reagent	Absorbance	
	SW	SB
0.01	0.088	0.361
0.02	0.092	0.384
0.03	0.095	0.466
0.04	0.090	0.452
0.05	0.086	0.448

Impact of Volume Changing of Reagent:

A study was conducted to stabilize the optimal amount of the reagent solution Naphthalene-1,5-diamine of $3 \times 10^{-2} \mu\text{g/ml}$, which gives the maximum absorption of the dye formed, as increasing volumes (0.1-2ml) were added from the reagent solution, 1ml of the CAPR solution of 150 $\mu\text{g/ml}$ and 1ml of the as an oxidizing agent solution of potassium Iodate of $2 \times 10^{-2} \mu\text{g/ml}$ and then Distilled

water, where the results indicate that 1ml of the volume used is the optimal volume of the reagent, so 1ml of this reagent was used in the following experiments. The results are indicated in Table (3).

Table 3: Impact of the reagent volume

ml of Reagent 3×10^{-2} M	Absorbance	
	BW	SB
0.1	0.073	0.216
0.3	0.080	0.283
0.5	0.088	0.347
0.7	0.090	0.395
1	0.095	0.467
1.3	0.093	0.378
1.5	0.086	0.291
1.7	0.080	0.210
2	0.075	0.186

Choosing the best oxidizing agent:

Several oxidizing agents, each in a volume of 1ml was utilized at a concentration of $2 \times 10^{-2} \mu\text{g/ml}$. In a volumetric flask with a capacity of 25ml, 1 ml was added of Naphthalene-1, 5-diamine to 1ml of CAPR solution 150 $\mu\text{g/ml}$ and distilled water, then the absorbance of each sample was measured against the blank solution. It was observed that the best oxidizing agent was potassium Iodate, which gave the highest absorption to the dye formed at the wavelength of 568nm. The results are indicated in Table (4).

Table 4: Choosing the Best Oxidizing Agent

Oxidizing agent 2×10^{-2} M	Absorbance		Ma $\lambda_{\text{max}}(\text{nm})$
	Blank	Sample	
Potassium Iodate	0.095	0.466	567
Potassium persulfate	0.086	0.325	531
Ammonium persulfate	0.078	0.274	482
Ammonium ferric sulfate	0.067	0.193	435

Impact of Changing the Concentration of Oxidizing Agent:

When different concentrations of oxidizing agent potassium Iodate were added to 1ml of CAPR solution of 150 $\mu\text{g/ml}$ and added to it 1ml of the reagent Naphthalene-1,5-diamine solution with a concentration of $3 \times 10^{-2} \mu\text{g/ml}$ and distilled Water, it gave a maximum absorption at concentration $2 \times 10^{-2} \mu\text{g/ml}$. The results are indicated in Table (5).

Table 5: Impact of Changing the Concentration of Oxidizing Agent

M of Oxidizing agent KIO_3	Absorbance	
	BW	SB
0.01	0.090	0.351
0.02	0.095	0.466
0.03	0.088	0.442
0.04	0.082	0.441
0.05	0.077	0.441

Impact of the oxidizing agent volume:

This study was conducted to choose the best volume of oxidizing agent solution with a concentration of $2 \times 10^{-2} \mu\text{g/ml}$, as different volumes were used (0.2-2ml) and it was found that the volume of 1ml of the solution of potassium Iodate is the best volume that was used in the following experiments, and the results are indicated in Table (6).

Table 6: Impact of the oxidizing agent volume

ml of KIO_3 $2 \times 10^{-2} \text{M}$	Absorbance	
	BW	SB
0.2	0.068	0.235
0.4	0.077	0.296
0.6	0.082	0.384
0.8	0.089	0.408
1	0.095	0.467
1.2	0.086	0.415
1.4	0.080	0.362
1.6	0.072	0.325
1.8	0.066	0.271
2	0.060	0.212

Impact of the sequence of additions:

When mixing, the order in which solutions are added can have a significant impact on the end product. As

a result, a series of laboratory tests with various additions sequences were conducted. Table (7) shows that arrangement (I) gave the highest absorption for the dye formed, indicating that the reagent is first oxidized and subsequently mixed with the drug, and has thus been employed in the following experiments.

Table 7: Impact of sequence of additions

Numbering Order	Order Of addition	Absorbance	
		BW	SB
I	D+R+O	0.094	0.466
II	O+R+D	0.088	0.413
III	O+D+R	0.080	0.372
VI	R+D+O	0.074	0.330

Potassium Iodate solution (O), reagent solution Naphthalene1, 5diamine (R), Reduced Chloramphenicol (D).

Impact of time:

The time required to complete the reaction was studied by taking a series of volumetric flasks with a capacity of 25 ml containing 1 ml of CAPR solution at a concentration of 150 $\mu\text{g/ml}$ and added to it 1 ml of the reagent solution Naphthalene-1,5-diamine with a concentration of $3 \times 10^{-2} \mu\text{g/ml}$ and then added 1 ml was from the oxidizing agent solution potassium Iodate at a concentration of $2 \times 10^{-2} \mu\text{g/ml}$ and distilled water to the mark, the solutions were left for periods, then the absorbance of the solutions was measured at the wavelength of 568 nm against their blank solutions. The results are indicated in Table (8).

Table 8: The Effect of time

Time/min	0	5	10	15	20
Absorbance	0	0.382	0.466	0.435	0.412

Temperature's Impact:

The effect of temperatures (10-60 $^{\circ}\text{C}$) on the oxidative coupling reaction has been investigated using the optimum conditions obtained from previous experiments, note in this study that the absorbance reached its maximum at 25 $^{\circ}\text{C}$ degrees, so the laboratory temperature was used in subsequent experiments. The results are indicated in Table (9).

Table 9: The Influence of Temperature

Temperature $^{\circ}\text{C}$	10	15	20	25	30	35	40	45	50	55	60
Absorbance	0.294	0.342	0.388	0.468	0.428	0.378	0.351	0.332	0.310	0.281	0.279

The Product Stability:

When comparing the absorbance of the product formed against the blank solution at different time intervals at a temperature of 25 $^{\circ}\text{C}$ to determine the product's stability, and to know the stability of the formed product, were take three different volumes of the CAPR solution 150 $\mu\text{g/ml}$ which are (2,3,4 ml), the final concentration was (12,18,24) $\mu\text{g/ml}$, were

adding 1ml of the reagent solution Naphthalene-1,5-diamine of $3 \times 10^{-2} \mu\text{g/ml}$, 1ml of the oxidizing agent solution potassium Iodate of $2 \times 10^{-2} \mu\text{g/ml}$ in a 25 ml volumetric flask, then completed the volume to the point of the mark with distilled water. The results in table (10) show that the product formed was observed to be stable for 70 minutes. As in previous drugs paper⁽²³⁾

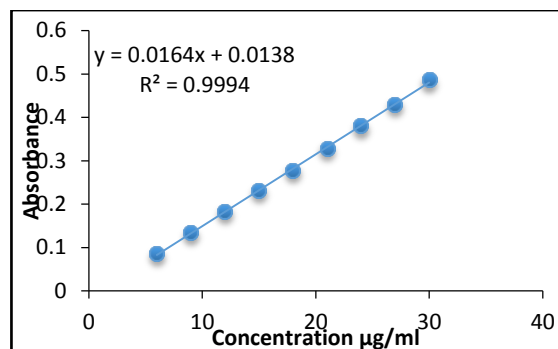
Table 10: The Stability of the Reaction Product

$\mu\text{g/ml}$ of CAPR	Absorbance Reaction / Time						
	10	20	30	40	50	60	70
12	0.254	0.271	0.286	0.287	0.288	0.287	0.288
18	0.328	0.345	0.364	0.365	0.365	0.364	0.365
24	0.446	0.462	0.470	0.484	0.483	0.484	0.484

Calibration Curve:

After Knowing the optimum conditions of the method the calibration curve was plotted by took different concentrations of CAPR and measured the absorbance of colored product, which was formed by reacting the drug with the reagent. In a series of 25 ml of volumetric flasks. Different volumes (1-5ml) were taken from a solution of CAPR with a concentration of $150 \mu\text{g/ml}$ representing (6-30 $\mu\text{g/ml}$), added 1ml of the reagent solution Naphthalene-1,5-diamine of $3 \times 10^{-2} \mu\text{g/ml}$, then add 1ml of the as an oxidizing agent potassium Iodate solution of $2 \times 10^{-2} \mu\text{g/ml}$, left the solutions for 10 minutes to complete the reaction and stabilize then complete the volume to the point of the mark with distilled water, then measured of absorption of the solutions at wavelength 568nm versus the blank solution. The results indicated in Figure (3), shows that the calibration curve follows Beer's law in the range of (6-27 $\mu\text{g/ml}$), and a deflection occurs at concentration 30 $\mu\text{g/ml}$, the value of the molar absorption coefficient of the method $0.4805 \times 10^4 \text{ L.mol}^{-1}.\text{cm}^{-1}$, and Sandell's sensitivity

was $0.061 \mu\text{g}.\text{cm}^{-2}$, the value of the correlation coefficient is 0.9994.

**Fig. 3: Calibration curve for the determination of CAPR****Method validation**

The accuracy of the method represented by Relative Error RE%, and Recovery percentage (R%) was calculated the precision of the method represented by Relative standard Deviation RSD% was calculated of three different concentrations (9, 18, 27 $\mu\text{g/ml}$), the accuracy and precision of the procedure are demonstrated by the results in Table (11).

Table 11: The Accuracy and compatibility of the Method

Concentration. of CAPR $\mu\text{g}.\text{mL}^{-1}$	Concentration of Measured $\mu\text{g}.\text{mL}^{-1}$	RE %	Recovery %	Average of Recovery %	RSD %
9	8.9	-0.12	99.88	100.05	0.473
18	18.1	0.5	100.5		0.430
27	26.94	-0.22	99.77		0.382

*Average of six determinations

Limit of Detection:

At a wavelength of 568 nm, the detection limit was calculated by measuring the absorption of the lowest

concentration from the calibration curve (6 $\mu\text{g/ml}$) for six readings under identical conditions and displays the results in Table (12).

Table 12: Limits of Detection

Conc. of $\mu\text{g/ml}$	$\bar{X}$	S	D.L. $\mu\text{g/ml}$	Q.L. $\mu\text{g/ml}$
6	0.6873	0.009214	0.241	0.804

The Nature of the product Formed:

To find out the nature of the formed product and the ratio of the drug's binding to the reagent, where the two continuous changes methods (Job's method) and the molar ratio method were applied in both methods, the concentration of each of the CAPR solution and the reagent solution Naphthalene-1,5-diamine $3 \times 10^{-2} \text{M}$, in the continuous changes method (Job method) Different volumes of the drug solution, ranging from (1-9ml) were put into volumetric flask with a capacity of 25ml, then the rest of the additions were completed with the optimum volumes according to the method

of work, then they were diluted with distilled water to the mark limit and measured the absorptions of these solutions at 568 nm in comparison to their blank solutions, the ratio is 1:1, as seen in Figure (4).

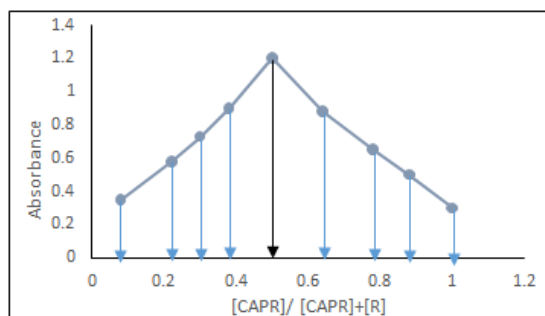


Fig. 4: Job's method for the determination of Chloramphenicol with a reagent Naphthalene-1,5-diamine in the presence of oxidizing agent potassium Iodate

To confirm that the reaction ratio between CAPR and the reagent Naphthalene-1,5-diamine is 1:1, the molar ratio method was used, as 1ml of the CAPR drug solution was placed in a series of 25ml volumetric flask, and the reagent solution was added in different sizes (0.2-3) ml, then the rest of the additions were complete of the optimal sizes and were diluted with distilled water to the mark, and the absorbance of solutions was compared to the blank solution at a wavelength of 568 nm. The molar ratio was determined to be consistent with the continuous changes method, as shown in Figure (5), and the ratio

is 1:1 between the CAPR and the reagent Naphthalene-1,5-diamine.

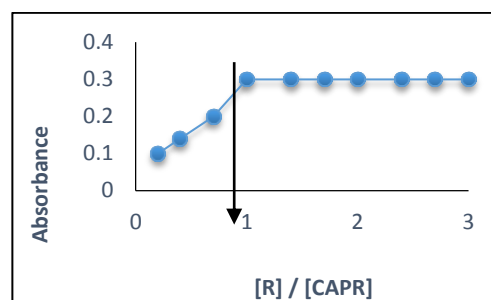


Fig. 5: The molar ratio method shows that the ratio is 1:1 between Chloramphenicol and the reagent Naphthalene-1,5-diamine

Reaction Mechanism

The results obtained in this method were based on oxidation of the reagent first, followed by coupling reaction of reductive chloramphenicol (CAPR) with Naphthalene-1,5-diamine, and potassium Iodate to form violet dye, that exhibited maximum absorption at 568 nm versus the blank solution. The functional group used for the color development for this method was primary amine group. A schematic reaction mechanism of CAPR with Naphthalene-1,5-diamine reagent was shown in Figure (6).

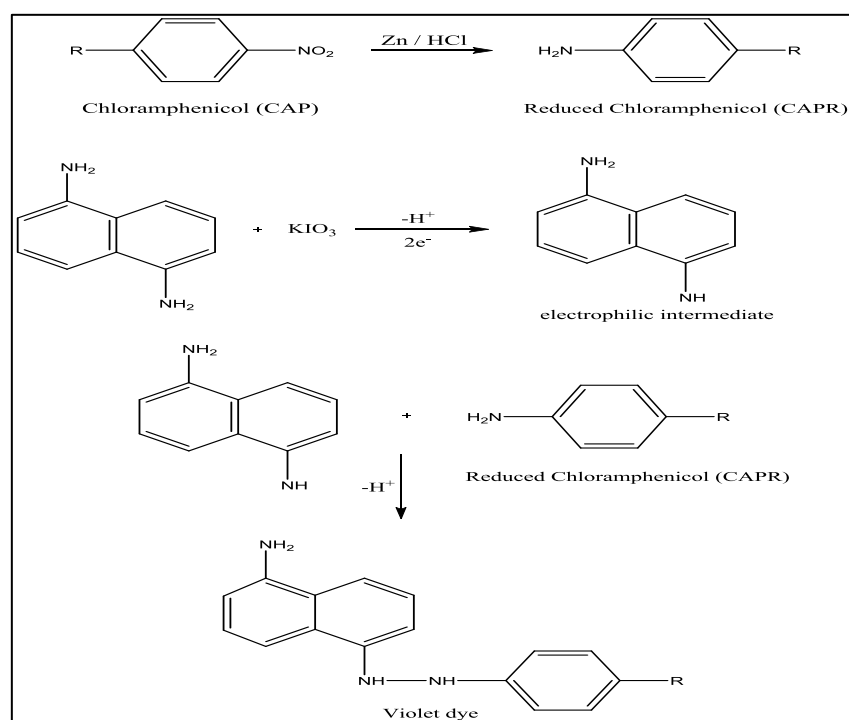


Fig. 6: A schematic reaction mechanism of CAPR with Naphthalene-1,5-diamine

Applications:

The method could be applied to pharmaceutical preparations containing chloramphenicol, which is a Samaphenicol pharmaceutical preparation in the form of eye drops containing 50mg of chloramphenicol.

The Direct method:

Three different concentrations of the preparations solution prepared (Eye drops) were taken (its preparation is indicated in page 3 Pharmaceutical

Preparation of Chloramphenicol) which is (15, 21, 27 $\mu\text{g/ml}$) and by the same steps followed when preparing the calibration curve, and then the absorption was measured at wavelength 568 nm versus the blank solution, and the average of six was calculated Measurements for each concentration, then the recall was calculated, and the results indicated in Table (13).

Table 13: The Direct Method

Concentration of CAP (Eye drops) $\mu\text{g/ml}$	Concentration of Measured $\mu\text{g/ml}$	RE %	Recovery %	Average Recovery %	RSD %
15	15.02	0.13	100.13	99.99	0.381
21	21.01	0.04	100.04		0.345
27	26.95	-0.19	99.81		0.266

The results in table (13) show that the proposed method has succeeded in appreciating the pharmaceutical preparation that contains Chloramphenicol. Where the value of the average recovery was 99.99%.

Conclusions

An easy, simple and highly sensitive spectroscopic method has been developed for the determination of

Chloramphenicol based on the oxidative coupling reaction of the reducing Chloramphenicol drug with the reagent Naphthalene-1,5-diamine in the presence of the oxidizing agent potassium Iodate to form violet dye dissolved in water and stable, give the maximum absorption at 568 nm. The proposed method has been successfully applied in the determination of preparation containing Chloramphenicol.

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التقدير الطيفي للكلورامفينيكول عن طريق تفاعل الإقتران التأكسدي مع الكاشف نفتالين -5,1- ثنائي

أمين بوجود يودات البوتاسيوم

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الملخص

يتضمن البحث تطوير طريقة بسيطة ودقيقة وحساسة لتقدير الكلورامفينيكول (CAP) في المستحضرات الصيدلانية. تعتمد الطريقة على تفاعل الإقتران التأكسدي للكلورامفينيكول بعد اختزال مجموعة النيترو في الدواء إلى مجموعة أمينية مع النفثالين -5,1- ثنائي أمين ككاشف بوجود يودات البوتاسيوم كعامل مؤكسد مكوناً صبغة بنفسجية قابلة للذوبان في الماء لها أقصى امتصاص عند 568 نانومتر. يطبق قانون بير في النطاق التركيز 6-27 مايكروغرام. مل⁻¹ مع امتصاص مولاري 0.4805×10^4 لتر.مول⁻¹.سم⁻¹، وحساسية ساندل 0.061 مايكروغرام.سم⁻²، معامل الارتباط 0.9994، معدل الإسترجاعية 100.05%. كان حد الكشف 0.241 مايكروغرام.مل⁻¹ وحد الكمي 0.804 مايكروغرام.مل⁻¹. كان المركب مستقرًا لمدة 70 دقيقة مع الخطأ النسبي من 0.12- إلى 0.22% والانحراف القياسي النسبي من 0.476 إلى 0.362%، تم تطبيق الطريقة بنجاح لتحليل الكلورامفينيكول في المستحضر الصيدلاني (قطرة العين). الكلمات الرئيسية: الكلورامفينيكول، جهاز مطيافية الأشعة فوق البنفسجية والمرئية، تفاعل الإقتران التأكسدي، مستحضر الصيدلاني.