RESEARCH ARTICLE

Spectrophotometric Determination of Phenylephrine Hydrochloride in Pharmaceutical Preparations by Oxidative Coupling Reaction

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ABSTRACT

Phenylephrine is a medication mainly applied as a decongestant to relieve hemorrhoids, increase blood pressure, and dilate the pupil. It is the first-line choice for prevention and treatment of hypotension during spinal anesthesia for cesarean section. However, many methods were used for the determination of this drug and its properties. This paper proposes a method that uses N, N dimethyl-p-phenylenediamine dihydrochloride as a new chromogenic reagent (NNDPH). This method is based on the oxidative coupling reaction of phenylephrine with N, N dimethyl-p-phenylenediamine dihydrochloride with ferric chloride in basic media to form green-blue soluble dye product. The outcomes demonstrate that the maximum absorption is at 680 nm, molar absorptivity of 5.54×10^3 $mol^{-1}.cm^{-1}$, and Sandell's sensitivity of 0.038 $\mu g.cm^{-2}$. The concentration range of 4 to 22 $\mu g.mL^{-1}$ of the product is conformable to Beer's law. Furthermore, it is applied successfully for estimating the drug properties on a simple condition or pharmaceutical preparations.

Keywords: Absorptivity, Oxidative coupling, Pharmaceutical, Phenylephrine hydrochloride, Spectrophotometric.

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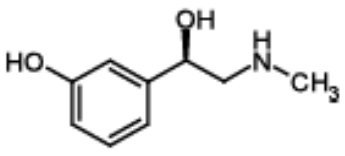
INTRODUCTION

Phenylephrine hydrochloride (PEH) is used as an effective remedy for narrowing the blood vessels in the nasal mucosa that works to relieve nasal congestion.[1] This drug is also used to dilate the pupil and relieve the symptoms of colds and rhinitis.[2] PEH, chemically known as (R)-1-(3-hydroxyphenyl)-2-methylamino ethanol hydrochloride, is a sympathomimetic drug (α-adrener).[3] This drug may be useful in the treatment of low blood pressure and relief of symptoms of external or internal hemorrhoids (Figure 1).[4,5]

In the literature, there are many methods used to estimate phenylephrine, including the following:

- Chromatographic[6-8]
- Spectrophotometric[9-12]
- Electrical[13-16]
- Flow-injection-chemiluminescence[17,18]



.HCl

Figure 1: $C_9H_{13}NO_2$.HCl, M. wt 203.705 $g.mol^{-1}$

Additionally, phenylephrine can be determined by using nanotubes.[19,20] Recently, the method has been adopted by using an oxidative coupling reaction with N, N-dimethyl-p-phenylenediamine dihydrochloride in the presence of ferric chloride in a strong base medium. The proposed method was sensitive, accurate, as well as, offering the advantage of high sensitivity for the developers.

MATERIALS AND METHODS

PEH Solution

A solution of 1,000 μg/mL PEH was prepared by adding 0.1-gram of phenylephrine in 100 mL of H_2O. 25 and 10 mL of the prepared solution, which was diluted to 100 mL with distilled water to obtain 250 and 100 μg/mL, sequentially.

Pharmaceutical Formulation

Tussiram syrup was considered as a sample, by taking 50 mL of it. Then, the oxidizing agent was prepared by dissolving 0.23-gram of $FeCl_3$ in 100 mL of distilled water (Beijing Solarbio Science and Technology Co.). Sodium hydroxide solution was prepared by dissolving 0.4-gram in 100 mL of distilled water (Beijing Solarbio Science and Technology Co.).

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Reagent Solution

The required concentration of the reagent was prepared by dissolving 0.209-gram of N, N dimethyl-p- phenylenediamine dihydrochloride 0.01M in 100 mL of distilled water. (Beijing Solarbio Science and Technology Co.).[21]

RESULTS AND DISCUSSION

The optimal conditions to determine the best quantity of materials that affect the intensity of the product color and the absorption was studied by using 2 mL of 250 µg/mL concentrated drug.

Effect of Oxidizing Reagents Amount

The oxidizing reagent was prepared by mixing 1-mL of ferric chloride oxidant solution, 2 mL of phenylephrine solution, and 1-mL of sodium hydroxide solution. A series of reagent volumes, ranging between 0.5 and 2 mL, was prepared. It was found that 1.5 mL of the reagent volume accorded the higher absorption, as shown in Table 1. Therefore, it was used in recent work.

Table 1: Effect of oxidizing reagent amount

Amount of oxidizing reagent (mL)	*Absorbance*	
	BW	*SB*
0.5	0.045	0.266
0.8	0.051	0.342
1	0.073	0.355
1.5	0.057	0.551
2	0.083	0.411

Table 2: Impact of amount of coupling reagent

Amount of coupling reagent (mL)	*Absorbance*	
	BW	*SB*
0.5	0.055	0.321
0.8	0.042	0.322
1	0.06	0.421
1.5	0.083	0.501
2	0.044	0.611

Table 3: Effect of amount of base

Amount of NaOH 1M (mL)	*Absorbance*	
	BW	*SB*
0.5	0.081	0.312
1	0.083	0.41
1.5	0.074	0.561
2	0.012	0.402
2.5	0.031	0.31

Table 4: Effect of order of addition

Order number	*Order of addition*	*Absorbance*
I	O+D+R+B	0.411
II	D+R+O+B	0.522
III	O+R+D+B	0.601
IV	R+D+O+B	0.333

Impact of Amount of Coupling Reagent

To study the effect of the coupling reagent, a series of 0.5 to 2 mL of the oxidized agent ferric chloride, 1-mL of N, N dimethyl-p-phenylenediamine dihydrochloride, and 1-mL of sodium hydroxide solution was prepared. 2 mL of the prepared series absorb the higher amount of the absorption (Table 2).

Effect of Amount of Base

The best solution volume of alkaline solution sodium hydroxide was 1.5 mL, due to giving higher absorption (Table 3).

Impact of Order of Addition

The results showed that the best addition sequence is O+R+D+B, which gave the highest absorption, as shown in Table 4, where (O) oxidizing agent, (C) coupling reagent, (D) drug, and (B) base. Therefore, this sequence is chosen in this study.

Stability of Reaction Product

As listed in Table 5, the results showed that the absorption value is stable, and almost constant at 5–70 minutes. This duration is sufficient for many measurements.

Temperature Impact

The range of temperatures was varied, ranging between 5 to 50ºC. The optimum temperatures that affected the absorption of the colored product were ranging between 10–30ºC, as shown in Table 6.

Medium Effect

Several media were tested. The best solvent was the distilled water, which gave the highest rate of absorbance (Table 7).

Final Absorption Spectrum

The final absorption showed that the optimal condition for the best absorbance for N, N-dimethyl-p-phenylenediamine

Table 5: Stability of reaction product

Time (min)	*Absorbance*
5	0.601
10	0.61
15	0.61
20	0.612
30	0.601
40	0.6
50	0.6
60	0.61
70	0.611

Table 6: Effect of temperature

Temperature (ºC)	*Absorbance*
5	0.6
10	0.61
15	0.61
20	0.612
30	0.613
40	0.582
50	0.301

dihydrochloride is 2 mL amount, 1.5 mL for the oxidizing agent, and 2 mL of the base, and the best temperature is 25ºC (Table 8; Figure 2).

Calibration Graph

A series of PEH solution 100 µg/mL (1–6.5 mL) was prepared to study the compatibility of the prepared solution with Beer's law. The linear regression was used to calculate the constants of the equation. As shown in Figure 3, the coefficient of determination (R^2) was (Y =0.0272x – 0.0115), with a range of 4 to 22 µg.mL^{-1} of PEH. The analytical data obtained from the calibration graph are shown in Table 9. Sandell's index 0.038 µg.cm^{-2}.[22,23]

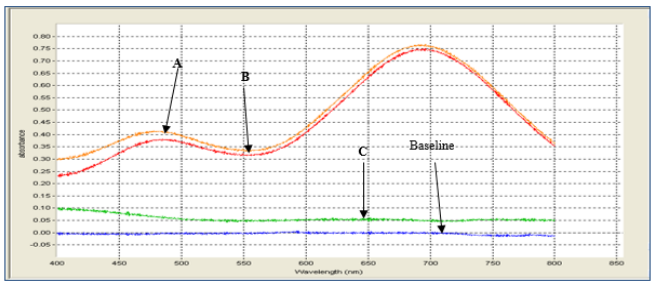

Figure 2: Final absorption spectrum; **A.** Absorption spectrum of chlorpromazine *vs.* distilled water; **B.** Chlorpromazine *vs.* blank; **C.** Blank *vs.* distilled water

Table 7: Solvent effect

Solvent	*Absorbance*
Methanol	0.12
Propanol	0.14
Water	0.622
Butanol	0.162

Table 8: Final absorption spectrum

λ_{max} (nm)	670
Amount (mL) of 1 × 10^{-2}M N,N-dimethyl-p-phenylenediamine dihydrochloride	2 mL
Amount (mL) of 1 × 10^{-2}M ferric chloride $FeCl_3$	1.5 mL
Amount (mL) of 0.1M sodium hydroxide solution	2 mL
Temperature, solvent	25ºC, water

Table 9: Analytical data for determination of PEH

Analytical data	*Value*
Linear range	0–30 µg.mL^{-1}
Correlation coefficient	0.9991
Regression equation	Y = 0.0272x − 0.0115
Molar absorption coefficient	0.54 × 10^3 L.mol^{-1}.cm^{-1}

Precision and Accuracy

Three different concentrations of PEH solution of 100 µg/mL have accuracy and compatibility with the calibration curve (Table 10).[24]

Detection Limit

The detection limit was calculated for 4 µg/mL, by measuring the absorbance for lower concentration in the calibration curve. This concentration was at 680 nm (Table 11).[25]

Stoichiometry of PEH-N, N dimethyl-p-phenylenediamine dihydrochloride complex

To determine the nature of the produced product and the correlation ratio of PEH solution with reagent solution N, N dimethyl-p-phenylenediamine dihydrochloride, the continuous change method (job method) and mole ratio method were applied, after completing the additions, depending on the procedure followed in these studies.[10,26] As depicted in Figures 4 and 5, the absorption for solutions was measured against blank solutions at 680 nm, where the conjugated ratio was 1:1. Finally, the proposed equation of interaction with the product is shown in Figure 6.

Applications

NNDPH was applied to pharmaceutical preparation (Tussiram) syrup containing phenylephrine (5 mg/10 mL).

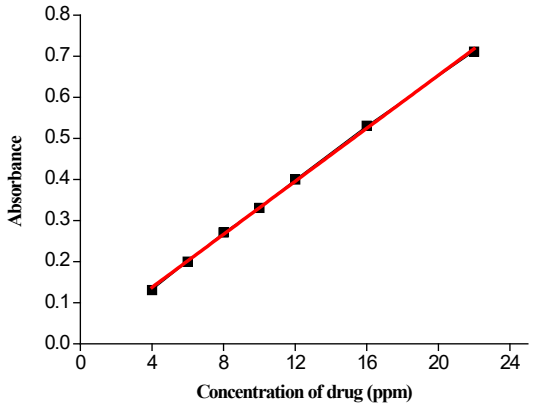

Figure 2: Calibration graph for determination of PEH

Table 10: Precision and accuracy

Concentration of PEH (µg/mL)	RE (%)	Recovery (%)*	Average of recovery (%)	RSD (%)
8	0.7	100.7	100.8	2.25
12	1.5	101.5		0.9
	22	0.4	100.4	1.6

*Average of five determinations

Table 11: Detection limit

Concentration (ppm)		*S*	*D.L. ppm*
4	0.1137	0.00014	0.015

Table 12: Direct method

Concentration of PEH (µg/mL) (tablet)	*RE (%)*	*Recovery (%)*	*Average recovery (%)*	*RSD (%)*
26	1	101		1.6
20	-1	99	100.04	0.83
8	0.19	100.12		0.39

Table 13: Statistical evaluation

		Recovery % ± RSD	
Preparation	*Nominal value*	*Proposed method*	*Literature method*
Tusculum (syrup)	5 mg/5 mL	100.12 ± 0.39 t = 1.24 F = 3.22	105 ± 1.82 t = 2.776 F = 6.36

Table 14: Comparison of method

Analytical parameters	*NNDPH*	*Literature method*[27]
ε.L.mol^{-1}.cm^{-1}	0.554×10^4	0.662×10^4
λ_{max} (nm)	680	510
Beer's law range (ppm)	4–22	0.4–11.2
Stability (hr)	60	48
Reagent	N, N dimethyl-p-phenylenediamine dihydrochloride	2-aminobenzothiazole
Solvent	Water	Water
Temperature	Room temperature	Room temperature

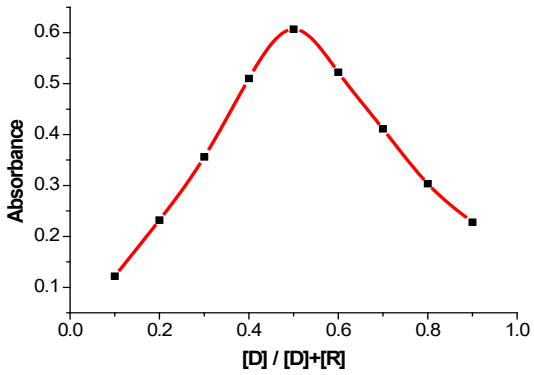

Figure 3: Job method for determining PEH with p-nitroaniline and ceric (IV) ammonium nitrate

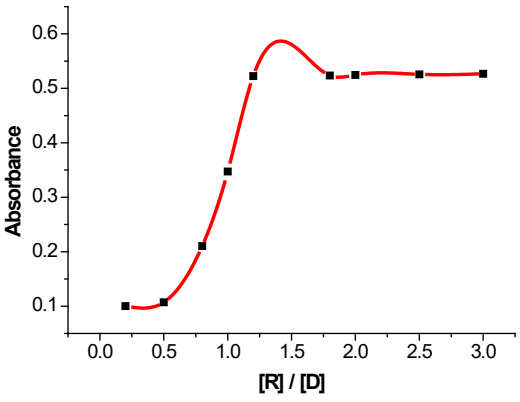

Figure 4: Mole ratio method for determining PEH with N, N dimethyl-p-phenylenediamine dihydrochloride

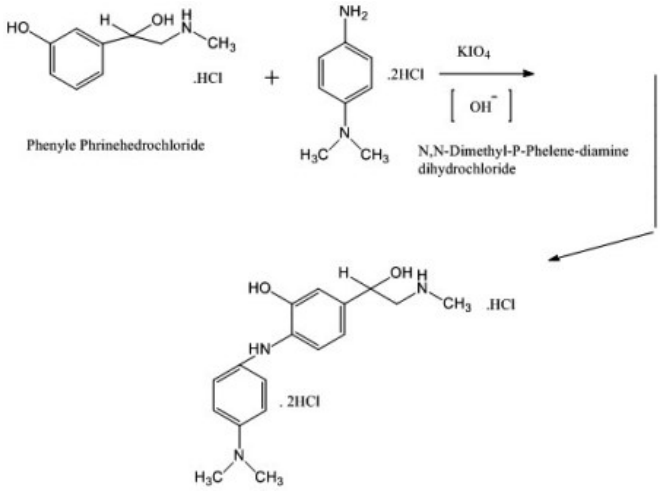

Figure 5: Proposed equation of interaction

Direct Method

Three different concentrations of Tussiram solution with a concentration of 100 µg/mL were prepared. Their absorbance at 680 nm wavelength was studied. The solutions were treated with the same topics as the calibration curve. Additionally, the outcomes demonstrate that the adopted method works successfully to anticipate the drug in its syrup (Table 12).

Statistical Calculations of the Results of NNDPH

For precision with the standard method, the t test method was used to evaluate the validity of the proposed method.[5] The evaluation was carried based on potentiometric titration of pure drug dissolved in anhydrous acetic acid with per-chloric acid. It is worth mentioning, no method was mentioned in British Pharmacopeia for determining PEH in syrup preparations. Accordingly, Table 13 lists the comparison between NNDPH

with other reposted methods. The obtained result was less than the tabular value for F, t; F = 3.22, t = 1.24 (which is required). While, the tabular value for F, t was F = 6.36, t = 2.776 at confidence limit 95%, and for four degrees of freedom. Consequently, these values demonstrate the success of the proposed method.

Comparison between the Two Methods

The success of NNDPH was also proved by comparing it with a method[10] (Table 14).

CONCLUSIONS

PEH is a nasal decongestant that helps to relieve a blocked nose. It reduces the size of the blood vessels in the nose and sinuses, helping you to breathe more easily. However, many reagents were used as an oxidative coupling for this drug. Many methods were used to determine this drug and its properties. This paper proposes an oxidative coupling reaction of phenylephrine with N, N dimethyl-p-phenylenediamine dihydrochloride with ferric chloride in basic media to form a green-blue soluble dye product has been used. According to the findings, the current method is suitable for the routine analysis of this drug in its pharmaceutical preparations and its pure form. This method has also been characterized by linearity, accuracy, and high compatibility, and the procedure does not require special conditions, like temperature or pH limit.

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