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DEVELOPMENT OF THE METHOD FOR QUANTITATIVE DETERMINATION OF PHENYLEPHRINE HYDROCHLORIDE IN THE COMBINED DROPS

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Key words: phenylephrine hydrochloride; validation; quantitative determination; spectrophotometry; combined nasal drops

The work is devoted to development of the method for quantitative determination of phenylephrine hydrochloride in the combined nasal drops Gripocitron Rinis. Combination of phenylephrine hydrochloride possessing the vasoconstrictor action with dimetindene maleate having the antihistaminic action formulated into drops decreases nasal discharge and helps to clear the nasal passages without disturbing physiological functions of the ciliated epithelium and mucous membrane. It has been found that the quantitative content of phenylephrine hydrochloride in the combined nasal drops can be determined by spectrophotometry in the ultraviolet spectrum at the wavelength of 296 nm. The method proposed demonstrates the possibility of phenylephrine hydrochloride determination in combined drops in the presence of another active pharmaceutical ingredient – dimetindene maleate, which has the absorption minimum in sodium hydroxide solution at this wavelength. The influence of the additional ingredient – benzalkonium chloride drops on the nature of the absorption spectrum of phenylephrine hydrochloride is recommended to eliminate by the action of potassium dichromate solution. The optimal amount and concentration of solutions of sodium hydroxide and potassium dichromate, the analytical band have been determined. Validation of the given method has been carried out according to the following validation characteristics: linearity (a = 4.34 ≤ maxa 5.10%, b = 1.04), accuracy (0.52 ≤ maxδ 1.02%), convergence (1.07% ≤ max Δas 3.20%) and the correlation coefficient r, which is 0.9997. It has been found that the method suggested for determination of phenylephrine hydrochloride in nasal drops is precise, accurate, reproducible and linear, and it allows recommending it for using in pharmaceutical analysis.

Phenylephrine hydrochloride is a known sympathomimetic providing a selective stimulating effect mainly on postsynaptic α-adrenergic receptors [4, 5]. Being a decongestant with a moderate vasoconstrictor action phenylephrine contains in a number of nasal drugs for local application, such as Nasol Baby (phenylephrine hydrochloride), Nasol Kids (phenylephrine hydrochloride with eucalyptol and benzalkonium chloride), Vibrocil and Gripocitron Rinis (phenylephrine hydrochloride with dimetindene maleate and benzalkonium chloride), which are used for symptomatic treatment of nasal congestion, acute and chronic rhinitis, allergic rhinitis, sinusitis, acute otitis.

The search of safe and effective monocomponent and combined decongestant nasal drops for local application with phenylephrine hydrochloride actualizes development and standardization of methods for quality control of this substance in the presence of other active pharmaceutical ingredients (API) [3, 6, 7, 9].

Materials and Methods

The object of the research was Gripocitron Rinis, nasal drops, 15 ml. The active substances were phenylephrine hydrochloride equivalent to 2.5 mg of phenylephrine, dimetindene maleate 0.25 mg; excipients were citric acid, monohydrate; anhydrous sodium hydrogen phosphate; sorbitol (E 420); benzalkonium chloride; peppermint oil; purified water as needed. The reference standard (RS) of the substance of phenylephrine hydrochloride (Unichem laboratories Ltd, India) is batch PPPPH/1104 from 01.10.10.

The analytical study was performed by the method of absorption spectroscopy on an Evolution 60S spectrophotometer v4.003. During the work "AXIS" electronic laboratory balances, measuring glassware of class A were used. Reagents and titrants used in tests met the requirements of the State Pharmacopoeia of Ukraine [3, 8].

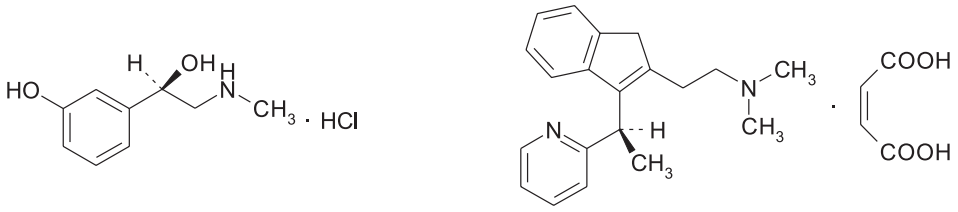
Experimental Part

Test solution. To the accurately weighed volume of the drug, which is equivalent to 2.5 mg of phenylephrine hydrochloride, add 0.25 ml of 5% solution of potassium dichromate, 5 ml 1 *M* solution of sodium hydroxide and dilute with water to the volume of 50.0 ml.

Reference solution (a). Dissolve 0.05 g of phenylephrine hydrochloride RS in 30 ml of 0.1 *M* solution of sodium hydroxide, dilute the volume of the solution with the same solvent to 50.0 ml. To 2.5 ml of the solution obtained add 0.25 ml of 5% solution of potassium dichromate, 5 ml of 1 *M* solution of sodium hydroxide and dilute with water to the volume of 50.0 ml.

Compensation solution. Dilute 0.25 ml of 5% solution of potassium dichromate and 5 ml of 1 *M* solution of sodium hydroxide with water to the volume of 50 ml.

The optical density of the test solution and the reference solution is measured at the wavelength of 296 nm with respect to the compensation solution.



I II

Scheme

Results and Discussion

To develop the method for quantitative determination of phenylephrine hydrochloride in Gripocitron Rinis drops the parameters of active ingredients solubility [3, 5] were studied, and their UV-spectra were investigated. Phenylephrine hydrochloride ((1*R*)-1-(3-hydroxyphenyl)-2-(methylamino)ethanol hydrochloride) (I) and dimetindene maleate (*N,N*-dimethyl-2-[3-[(*RS*)-1-(pyridine-2-yl)ethyl]-1*H*-indene-2-yl]ethanamine (*Z*)-butenedioate) (II) are readily soluble in diluted acids and alkalis, in alcohols their solubility is slightly different (Scheme).

At the same time it was found that when using 0.1 *M* solution of hydrochloric acid or alcohol as solvents the maxima of the UV-spectra of 0.005% solution of phenylephrine hydrochloride were observed at the wavelengths of 275 nm or 272 nm [1], and in 0.0005% solution of dimetindene maleate in the acidic medium at 260 nm and the alcoholic one – at 258 nm. Therefore, under these conditions dimetindene maleate has impact on the character of the ultraviolet spectrum of phenylephrine hydrochloride.

When replacing the solvent on 0.1 *M* solution of sodium hydroxide the absorption spectra of 0.005% solution of phenylephrine hydrochloride is characterized by the presence of two absorption maxima at the wavelengths of 239 nm and 292 nm (Fig. 1).

The UV-spectrum of 0.0005% alkaline solution of dimetindene maleate has the absorption maximum at the wavelength of 263 nm, which corresponds to the UV-spectrum minimum of phenylephrine hydrochloride, and at the wavelength of 292 nm the substance does not practically absorb. It indicates that dimetindene maleate does not interfere with detection of phenylephrine hydrochloride under such conditions. The alkaline solutions of Gripocitron Rinis drug and the test mixture containing the active substances in the same concentrations as in the dosage form were also studied. It has been found in the analysis of UV-spectra in the range from 220 to 320 nm that the spectrum of the test solution is characterized by the absorption maxima at the same wavelengths and practically overlaps with the absorption spectrum of phenylephrine hydrochloride solution. The maximum of the absorption spectrum of the solution prepared from the drops under research is also observed at the wavelength of 292 nm, but it is more intensive. Therefore, excipients containing in the drops influence on the character of the drug absorption spectrum (Fig. 1).

Our assumption has been made that the presence of benzalkonium chloride included into the composition of the drops as a preservative and antiseptic has an impact on the character of the UV absorption spectrum of the drops. From literature data it is known that the al-

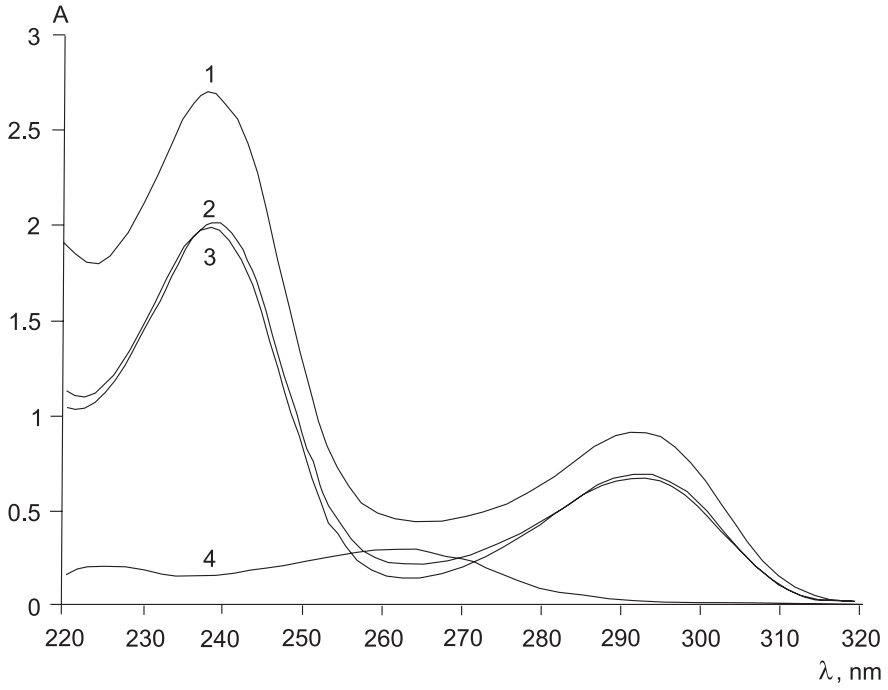


Fig. 1. The UV absorption spectra in 0.1*M* solution of sodium hydroxide: 1 –Gripocitron Rinis drug; 2 – test solution; 3 – 0.005% solution of phenylephrine hydrochloride; 4 – 0.0005% solution of dimetindene maleate.

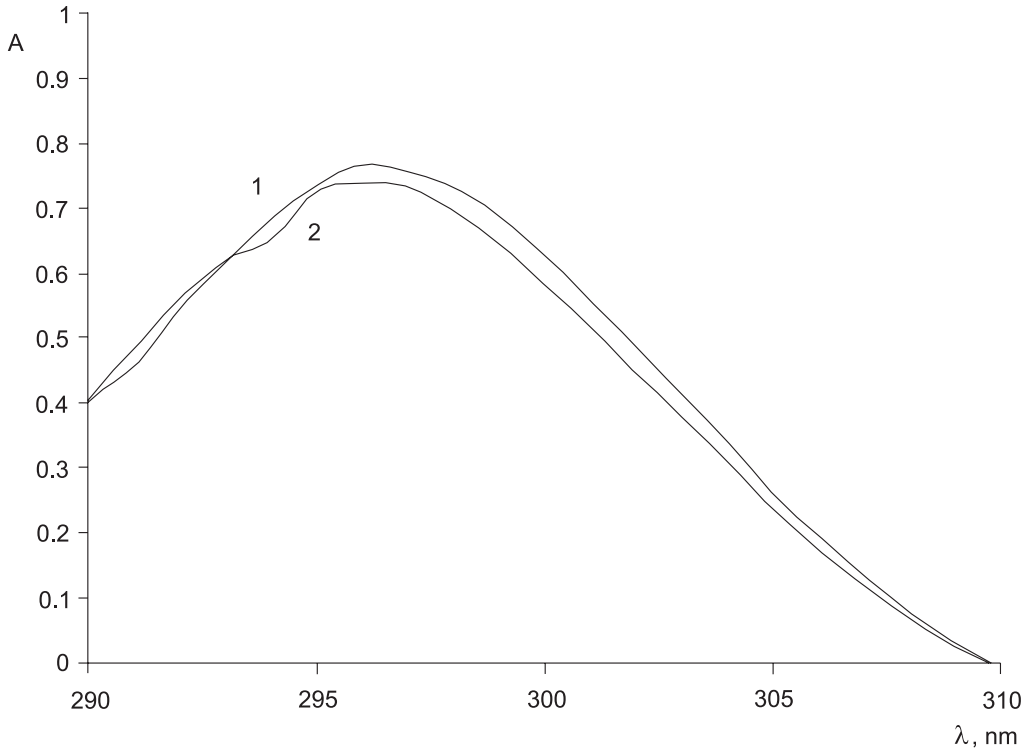


Fig. 2. The UV absorption spectra in 0.1*M* solution of sodium hydroxide with addition of 0.25 ml of 5% solution of potassium dichromate: 1 – Gripocitron Rinis drug; 2 – 0.005% of phenylephrine hydrochloride solution.

Table 1

Dependence of the optical density on the amount of potassium dichromate solution

Optical density	The amount of 5% solution of potassium dichromate, ml			
	0.15	0.20	0.25	0.30
A	0.716	0.845	0.706	0.938

coholic solution of benzalkonium chloride in the ultraviolet light has absorption maxima at 257, 263 and 269 nm [10]. To precipitate benzalkonium chloride its ability to interact with potassium dichromate was used. When studying the UV absorption spectra of Gripocitron Rinis drug in 0.1 *M* solution of sodium hydroxide with addition of 5% solution of potassium dichromate and phenylephrine hydrochloride solution under the same conditions in the region from 290 to 310 nm it has been found that their intensity is practically the same (Fig. 2).

Since the solution of potassium dichromate is coloured in an orange-yellow colour, the solution containing the same amount of potassium dichromate as in the test and reference solutions is used as a compensation solution in order to obtain more reliable data.

Table 2

The results of the linearity study

Parameters	Values	Values according to the SPhU
b	1.0425	
s_b	0.0049	
a	| -4.3372 |	max, a=4.80%
s_0	0.6585	max, S_0=1.69%
s_Y	13.0741	
r	0.9997	min, r= 0.99236

To determine the amount of potassium dichromate required for benzalkonium chloride binding the model solutions of drops were prepared; different amounts of 5% solution of potassium dichromate (Tab. 1) were added to them.

It has been found that to carry out the precipitation reaction with benzalkonium chloride 0.25 ml of 5% solution of potassium dichromate is required since with this amount of the solution the spectrum is the most similar to the spectrum of phenylephrine hydrochloride.

Thus, the research conducted allows to propose the quantitative determination of phenylephrine hydrochlo-

Table 3

The results of analysis for test solutions and their statistical processing

Mean, Z%	99.84
Relative standard deviation, Sz%	0.16
Relative confidence interval Δas%=t(95%.8)·Sz=1.860·Sz=	0.29
Critical value for convergence of results Δas%	3.20
Systematic error δ	0.05
Criterion of the systematic error insignificance 1) δ≤Δ/√9=0.05/3=0.016 2) if it is not satisfied 1), then δ≤0.75	satisfied
The overall conclusion of the method	correct

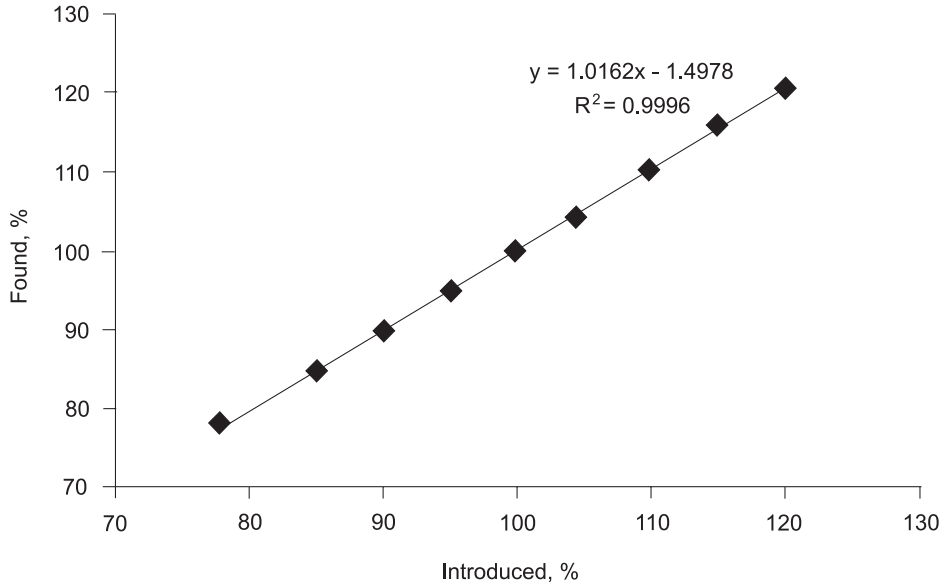


Fig. 3. The linear dependence of the optical density on the concentration of phenylephrine hydrochloride solutions in the normalized coordinates.

Table 4

The results of quantitative determination of phenylephrine hydrochloride in drops

No.	The volume of a dosage form, ml	A	A_o	Found phenylephrine hydrochloride, mg	Metrological characteristics
1	1.00	0.589	0.589	2.4975	$\bar{x}$ = 2.4939 S^2 = 0.0001 S = 0.0112 $\Delta\bar{x}$ = 0.0048 ε% = 0.47
2		0.590		2.5017	
3		0.586		2.4848	
4		0.585		2.4805	
5		0.592		2.5102	
6		0.587		2.4890	

ride in the combined nasal drops can be determined by spectrophotometry after preliminary transfer of dimetindene maleate into the base and precipitation of the excipient benzalkonium chloride with potassium dichromate solution.

To use the method for analysis of phenylephrine hydrochloride in the combined drops some validation characteristics such as linearity, precision, accuracy and convergence were studied (Tab. 2).

Linearity of the method was studied on the model solutions within the range of concentrations of 80-120%. It corresponds to the range of use relative to the nominal content of phenylephrine hydrochloride in nasal drops (Fig. 3, Tab. 2) [2].

The method of analysis is characterized by sufficient convergence and accuracy within the whole range of concentrations of 80-120%; it can be seen from the results obtained that are presented in Tab. 3.

The results of quantitative determination of phenylephrine hydrochloride in nasal drops by spectrophotometry are given in Tab. 4.

It has been found that relative uncertainty of the individual result is ±0.47%.

CONCLUSIONS

1. The method for quantitative determination of the active substance – phenylephrine hydrochloride in Gripocitron Rinis nasal drops has been developed by UV-spectroscopy.

2. The validation characteristics of the method proposed (accuracy and convergence, linearity, precision) have been studied. According to the results this method can be recommended for using in analysis of the given dosage form.

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РОЗРОБКА МЕТОДИКИ КІЛЬКІСНОГО ВИЗНАЧЕННЯ ФЕНІЛЕФРИНУ ГІДРОХЛОРИДУ В КОМБІНОВАНИХ КРАПЛЯХ

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Ключові слова: *фенілефрину гідрохлорид; валідація; кількісне визначення; спектрофотометрія; комбіновані назальні краплі*

Робота присвячена розробці методики кількісного визначення фенілефрину гідрохлориду у комбінованих назальних краплях Грипоцитрон Риніс. Поєднання у складі крапель фенілефрину гідрохлориду, який чинить судинозвужуючу дію, та диметиндену малеату, що має антигістамінну дію, зменшує виділення з носа і сприяє очищенню носових ходів, не порушуючи при цьому фізіологічних функцій миготливого епітелію та слизової оболонки. Встановлено, що кількісний вміст фенілефрину гідрохлориду в комбінованих назальних краплях можна визначати методом спектрофотометрії в ультрафіолетовій області спектра за довжини хвилі 296 нм. Запропонована методика доводить можливість визначення фенілефрину гідрохлориду в складних краплях у присутності іншого активного фармацевтичного інгредієнта диметиндену малеату, який у розчині натрію гідроксиду за цієї довжини хвилі має мінімум поглинання. Вплив допоміжного компонента крапель – бензалконію хлориду на характер спектра поглинання фенілефрину гідрохлориду рекомендовано усунути дією розчину калію дихромату. Встановлені оптимальна кількість та концентрація розчинів натрію гідроксиду та калію дихромату, аналітична хвиля дослідження. Проведена валідація зазначеної методики за такими валідаційними характеристиками: лінійність (a = 4.34 ≤ maxa 5,10%, b = 1,04), правильність (0,52 ≤ maxδ 1,02%), збіжність (1.07% ≤ max Δas 3,20%) та коефіцієнт кореляції r, який становить 0.9997. Встановлено, що запропонована методика визначення фенілефрину гідрохлориду в назальних краплях є точною, правильною, відтворюваною і лінійною, що дозволяє рекомендувати її для використання у фармацевтичному аналізі.

РАЗРАБОТКА МЕТОДИКИ КОЛИЧЕСТВЕННОГО ОПРЕДЕЛЕНИЯ ФЕНИЛЭФРИНА ГИДРОХЛОРИДА В КОМБИНИРОВАННЫХ КАПЛЯХ

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Ключевые слова: *фенилэфрина гидрохлорид; валидация; количественное определение; спектрофотометрия; комбинированные назальные капли*

Работа посвящена разработке спектрофотометрической методики количественного определения фенилэфрина гидрохлорида в комбинированных назальных каплях Гриппоцитрон Ринос. Сочетание в составе капель фенилэфрина гидрохлорида, обладающего сосудосуживающим действием, и диметиндена малеата с антигистаминным эффектом уменьшает выделения из носа и способствует очищению носовых ходов, не нарушая при этом физиологических функций мерцательного эпителия и слизистой оболочки. Установлено, что количественное содержание фенилэфрина гидрохлорида в комбинированных назальных каплях можно определять методом спектрофотометрии в ультрафиолетовой области спектра при длине волны 296 нм. Предложенная методика доказывает возможность определения фенилэфрина гидрохлорида в сложных каплях в присутствии другого активного фармацевтического ингредиента диметиндена малеата, который в растворе натрия гидроксида при указанной длине волны имеет минимум поглощения. Влияние вспомогательного компонента капель бензалкония хлорида на характер спектра поглощения фенилэфрина гидрохлорида рекомендовано устранять действием раствора калия дихромата. Установлены оптимальное количество и концентрация растворов натрия гидроксида и калия дихромата. Проведена валидация данной методики по следующим валидационным характеристикам: линейность (a = 4.34 ≤ maxa 5,10%, b = 1,04), правильность(0,52 ≤ maxδ 1,02%), сходимость(1,07% ≤ max Δas 3,20%) и коэффициент корреляции r, равный 0,9997. Установлено, что предлагаемая методика определения фенилэфрина гидрохлорида в назальных каплях является точной, правильной, воспроизводимой и линейной, что позволяет рекомендовать ее для использования в фармацевтическом анализе.