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### COOMASSIE BRILLIANT BLUE STAINING USED IN SPECTROPHOTOMETRIC ASSAY FOR DOPAMINE HYDROCHLORIDE AND METHYLDOPA DETERMINATION

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**Abstract:** The catalytic oxidation reaction between Coomassie Brilliant Blue and N-bromosuccinimide was used in an acidic medium to develop a spectrophotometric method for the estimation of dopamine and methyldopa in their pure forms and in pharmaceutical preparations. The developed method exhibited high sensitivity, with molar absorptivity values of  $1.77 \times 10^4$  and  $2.97 \times 10^4$  L mol<sup>-1</sup> cm<sup>-1</sup>, and Sandell's sensitivity values of 0.0080 and 0.0107 mg cm<sup>-2</sup>, respectively. The estimated quantities of dopamine hydrochloride and methyldopa were within the range of  $0.5-18 \mu \text{g/mL}$  and  $0.5-13 \mu \text{g/mL}$ , respectively. The developed method was successfully applied to tablet and injection formulations, and the results were in accord with the original drug content. The developed method was compared with standard addition and standard method in the British Pharmacopoeia, and no significant differences were revealed thus confirming the success of the developed method in the estimation of the studied drugs. Other statistical values were also studied . **Keywords:** catalysed oxidation, coomassie brilliant blue, dopamine hydrochloride, methyldopa, and spectrophotometric method.

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#### Introduction

Dopamine is a neurotransmitter that plays a crucial role in large physiological processes, including movement control, motivation, reward, and mood regulation [1]. It is synthesized from the amino acid tyrosine, and it is found throughout the central nervous system. The chemical composition of dopamine includes a catechol (a benzene ring with two hydroxyl groups) attached to an amine group [2] as shown in Fig.1.



Fig. 1. Chemical formula of dopamine hydrochloride

This structure allows dopamine to interact with specific receptors in the brain and elicit various physiological responses. Pharmacologically, dopamine acts as both a neurotransmitter and a hormone. It binds to specific receptors on target cells and triggers intracellular signalling pathways that lead to changes in cellular function. Dopamine agonists are used clinically for their ability to stimulate dopamine receptors and treat conditions such as Parkinson's disease and restless leg syndrome. Biologically, dopamine involves in many important processes, including movement control, motivation, reward. and mood regulation. Dysregulation of the dopamine been implicated in system has various

psychiatric disorders, including schizophrenia and addiction [3]. It is used to treat preeclampsia and gestational hypertension during pregnancy, as well as hypertensionrelated emergencies such as hypertensive encephalopathy and acute heart failure. Methyldopa is also used as off-label for conditions such as migraines and Tourette's syndrome. From a biological point of view, methyldopa is metabolized in the liver before being excreted by the kidneys. The chemical structure of methyldopa consists of a benzene ring with two hydroxyl groups (-OH) and a carboxyl group (-COOH) attached to it as shown in Fig.(2).



Fig. 2. Chemical formula of methyldopa

The active metabolite of methyldopa is alpha-methyl norepinephrine, which acts on alpha-adrenergic receptors to lower blood pressure. Methyldopa has a half-life of 2-3 hours and is usually administered orally in tablet form. In summary, methyldopa is a synthetic amino acid that acts as a centrally acting antihypertensive drug. It stimulates alphaadrenergic receptors in the brainstem to reduce sympathetic nervous system activity and lower blood pressure. Methyldopa has several pharmacological uses in addition to treating hypertension, including preeclampsia, hypertensive emergencies, and off-label use in migraine and Tourette's syndrome. Methyldopa is extensively metabolized in the liver before being excreted by the kidneys [4].

Chromatographic [5-8], electrophoretic [9-11], polarographic [12,13], fluorometric [14-17], and spectrophotometric [18-21] methods are all viable options for evaluating dopamine hydrochloride and methyldopa.

### **Materials and Methods**

**Apparatus:** Absorbance and spectral measurements were taken with a 1.0 cm thick quartz cell and a PG Instruments-England T92+ UV spectrophotometer.

**Reagents.** Drug solutions were prepared by dissolving 0.01 g of these pure substances in a small amount of distilled water and then filling a 100 mL volumetric flask to the mark.

**Coomassie Brilliant Blue 400µ/mL dye solution.** Prepared by dissolving 0.04 grams of the pure substance in a small beaker and then diluting it with distilled water in a 100 mL volumetric flask to the mark. The solution was then transferred to an ultrasonic bath to complete the dissolution process before being placed in a dark container.

Solutions of oxidizing agents. N-Bromosuccinimide  $2 \times 10^{-3}$  M: 0.0355 g of the pure crystalline substance was dissolved in a 100 mL volumetric flask filled with distilled water up to the mark. The flask was subjected to ultrasound to ensure complete dissolution and subsequent dilute solutions were made from it. Finally, all solutions were transferred to an opaque bottle, while the concentrations of the other oxidizing agents were prepared after being added in their optimal amounts.

Procedure. Increasing volumes of (0.05-1.8) and (0.05-1.3) mL of dopamine and methyldopa were added, respectively, at a concentration of 100 µg/ml two separate sets of 10-mLvolumetric flasks to cover the concentration ranges shown in Table 6 and the presence of optimal quantities. 1 mL of acetic acid at a concentration of 3 mol/L, followed by the addition of 1.2 mL of the oxidizing agent N-Bromosuccinimide at a concentration  $(2 \times 10^{-3})$ mol/L), and the solutions were left for 10 minutes with continuous shaking to complete their oxidation, followed by the addition of the optimal amount of Coomassie Brilliant Blue dye (100 µg/mL) and waiting for 10 minutes before diluting with distilled water and completing the volume to the mark. The absorbance of the solutions was measured at 550 nm against their blank solution.

### Pharmaceuticals preparations.

Analysis of Dopamine Hydrochloride Injection: A concentration of 2000  $\mu$ g/mL was obtained by diluting one injection containing 200 mg/5 mL with distilled water in a 100 mL volumetric flask. By further diluting with distilled water, a 100  $\mu$ g/mL solution was prepared. The calibration curve was then constructed using different volumes of this solution.

Analysis of the two types of methyldopa tablets is as follows: five tablets were weighed and then crushed and ground. From the resulting mixture, a single tablet weighing 0.250 g was taken. This tablet was dissolved in distilled water and the resulting solution was filtered. Next, the filtered solution was transferred to a 250 mL volumetric flask and diluted with distilled water to reach a concentration of 1000  $\mu$ g/mL. Subsequently, the solution was further diluted multiple times to obtain solutions within the range of a pure standard curve. Finally, these solutions were treated under the same conditions as the parent drug compound.

### **Results and Discussion**

Preliminary test: Coomassie Brilliant Blue was used to estimate two pharmacological compounds (dopamine hydrochloric and methyldopa) using an indirect spectroscopic method. The method was investigated in two stages. First, the dye's linear properties were investigated to determine the optimal amount. As a result, increasing of Coomassie Brilliant Blue ranging from (0.01-2.5) mL was added at a concentration of 400  $\mu$ /mL to a series of volumetric flasks with a capacity of 10 mL, as shown by the standard curve.



Fig. 3. Peak of 100 µg/mL of Coomassie Brilliant Blue Vs distilled water.

The optimal amount provides the highest absorption value at 550 nm within the linear range  $(0.4-100) \mu$  /mL for the dye in Fig.(3) and Fig (4), so a concentration of 100  $\mu$ /mL was adopted as the highest value subjected to Beer's law and used in the subsequent experiments. While the second stage included several experiments to examine the possibility of bleaching the dye in an acidic or basic medium, each separately, by adding a fixed amount of 1 mL of hydrochloric acid at a concentration of 1

M and 1 mL of sodium hydroxide at a concentration of 1 M, followed by adding a fixed amount of oxidizing agents, then adding the optimal amount of the dye, as the experiments showed that a dropping of the dye occurred with the increase in the amount of the oxidizing agent in the acidic medium, while in the alkaline medium was non-quantitative. Practical experiments show that there is an increase in the absorption of Coomassie Brilliant Blue with an increase in the

concentration of (dopamine hydrochloride and methyldopa), as the amount of the added oxidizing agent decreases in a calculated manner with the linear increase of the two medicinal compounds, which suggests the possibility of using Coomassie Brilliant Blue dye as a chromogenic reagent to estimate dopamine HCl and methyldopa.



Fig. 4. Calibration plot of Coomassie Brilliant Blue.

Table 1.	Effect of solv	ents.
Type of solvent	wavelength	Absorbance
Ethanol	592	1.047
Methanol	590	1.065
Acetone	594	1.077
Chloroform	-	turbid
Acetonitrile	592	1.036
Water	550	1.62



Fig. 5. Peaks of diffirent solvents.

### **Optimum Conditions:**

**Effect of solvents.** The effect of different polar solvents on dye absorbency was studied. An optimal volume of dye (2.5 mL) and 1 mL of hydrochloric acid at a concentration of 1 M were added to each flask. The absorbance was measured after supplementing the volume with different solvents as shown in Table 1. The water that showed the highest absorbance was adopted in subsequent studies.- Fig. (5).

**Proper oxidizing agent.** To figure out the proper oxidant for bleaching Coomassie Brilliant Blue dye, 2.5 mL of CBB (400 µg/mL)

was placed in a set of 10 mL volumetric flasks containing 1 mL of 1 M HCl. Then, 1 mL of 2 x  $10^{-3}$ Μ N-Bromosuccinimide, N-Chlorosuccinimide, chloramine T. sodium periodate, potassium permanganate, and ferric ammonium sulfate were added to each flask. The solutions were stirred for 10 minutes, and the absorbance at 550 nm was measured. The oxidizing agent N-Bromosuccinimide was found to produce the greatest bleaching of Coomassie Brilliant Blue Table 2 and was therefore selected for further use.

Table 2. Effect of oxid	kidizing agent type.		
Oxidizing agent	Absorbance		
$2 \times 10^{-3} \text{ M}$			
NBS	0.300		
NCS	1.319		
Chloramine T	1.438		
NaIO <sub>4</sub>	1.368		
KMnO <sub>4</sub>	0.510		
$(NH_4)Fe(SO_4)_2$	1.232		
Without	1.618		

Effect of the amount of oxidizing agent. To find the optimal amount of the oxidizing agent N-Bromosuccinimide, which gives the highest bleaching of Coomassie Brilliant Blue dye, growing volumes of the oxidizing agent NBS within the range (0.1-1.2) ml at a concentration of  $2 \times 10^{-3}$  M were added to a number of volumetric vials of 10 ml capacity and added to it 1 ml of hydrochloric acid at a concentration of 1 mol/L, followed by adding the optimal

amount of dye 2.5 ml at a concentration of 400  $\mu$ g/ml and measuring the absorption at 550 nm after 10 minutes of shaking, stirring, diluting with distilled water and completing the volume to the mark. It found that 1.2 ml of NBS oxidizing agent at a concentration of 2 × 10<sup>-3</sup> M is the best value for shortening the colour of the dye, so it was adopted in future studies. Fig. (6) Shows the optimal amount needed for Coomassie Brilliant Blue.



Fig. 6. Effect of the amount of oxidizing agent.

Effect of acid type. Based on preliminary experiments showing that the oxidation reaction between the NBS and Coomassie Brilliant Blue takes place in an acidic medium, the effect of adding different types of strong and weak acids to select the best acid was investigated to interact with the two drug compounds and the dye, 6  $\mu$ g/ml of both compounds was added.in two separate sets of mL volumetric flasks, then 1 mL of each acid was added at a concentration of 1 M, followed by the addition of the optimal amount of N-Bromosuccinimide (1.2 ml at a concentration of  $2 \times 10^{-3}$ ) M, then 2.5 ml of the dye was added and the absorbance at 550 nm versus blank was measured after dilution with distilled water and complement to10 ml. Table 3 shows that acetic acid is the best acidic medium in which the reaction takes place and was therefore used in subsequent studies.

Table	3. Effect of acid	d type
Type of acid 1M	Methyldopa	Dopamine. HCl
HCl	0.308	0.152
$H_2SO_4$	0.273	0.035
HNO <sub>3</sub>	0.188	0.027
CH <sub>3</sub> COOH	0.584	0.397

Effect of concentration and volume of acetic acid. To determine the concentration and volume of acetic acid, various concentrations of acetic acid in the range (1-5) mol/L were placed in two sets of 10 mL volumetric flasks containing 6  $\mu$ g/mL of both drug components, followed by the addition of N -Bromosuccinimide and in the presence of 2.5 ml Coomassie Brilliant Blue, was measured for

absorption after filling the volume with distilled water to the mark. The results showed in Fig. 7 that 3 M was most appropriate and was therefore adopted in subsequent studies. Then it was studied the effect of different volumes of 3 M acetic acid in the range of (0.5-3) mL. Fig. 8 which showed that 1 mL is an appropriate volume for both compounds.



Fig. 7. Effect of concentration of acetic acid.

Effect of time on reaction stability. In order to find sufficient time for the oxidation of two medicinal compounds, 6  $\mu$ g/ml of them were added with an optimal volume (1.2 ml N-Bromosuccinimide) at a concentration of (2x10<sup>-3</sup>M) and in the presence of 1 ml of acetic acid at

a concentration of 3M, shake frequently at different time intervals, then add 2.5 ml Coomassie Brilliant Blue, the reaction was monitored over time, once more after dilution with distilled water to 10 mL and at a laboratory temperature of 24°C and for both drugs.



Fig. 8. Effect of volume of acetic acid.

The results showed that a period of 10 minutes proved to be sufficient for the oxidation of (dopamine hydrochloride and methyldopa). It found that a period of 10 minutes was sufficient

to complete the interaction with the two drugs, with a period of less than 120 minutes for both drug compounds as shown in Table 4.

				01 011110	0111040	nom otho				
Standing time		Absorbance / standing time (min.)								
before dilution	5	10	15	20	30	40	50	60	120	130
(min.)										
			Do	pamine						
After addition	0.104	0.103	0.106	0.108	0.113	0.117	0.120	0.123	0.130	0.130
5	0.390	0.340	0.352	0.352	0.352	0.355	0.352	0.336	0.360	0.350
10	0.538	0.540	0.540	0.540	0.540	0.540	0.540	0.540	0.540	0.535
15	0.457	0.456	0.454	0.450	0.447	0.446	0.447	0.448	0.442	0.437
			Met	hyl dopa						
After addition	0.128	0.126	0.129	0.129	0.136	0.136	0.139	0.140	0.140	0.139
5	0.492	0.520	0.523	0.522	0.522	0.522	0.523	0.523	0.520	0.500
10	0.717	0.720	0.720	0.720	0.720	0.720	0.720	0.720	0.715	0.715
15	0.606	0.609	0.610	0.609	0.606	0.605	0.605	0.604	0.605	0.598

Table 4. Effect of time on reaction stability



Fig. 9. Effect of temperature on methyldopa.

Effect of temperature on reaction stability. The effect of temperatures (20–40) °C on the oxidation of two medicinal compounds was studied, as shown in Fig. (9). It revealed that the laboratory temperature of 24°C is optimal for methyldopa or dopamine after adding the remaining optimal amounts to several 10mL

graduated bottles and waiting 10 minutes. Then 100  $\mu$ g/ml Coomassie Brilliant Blue was added and, after dilution, another 10 minutes, till finally absorbance measurement at 550 nm. The measurements were also carried out at 0 ° C, and the results were not useful because no oxidation reaction occurred.



Fig. 10. Effect of temperature on dopamine.

**Effect of surfactant.** The effect of a range of surfactants on the absorption response was tested by adding 1 ml of each surfactant and the two therapeutic compounds. Based on the

results shown in Fig. 11, it was shown to have a negative impact on absorption, for this reason it was excluded from further studies.



Fig. 11. Effect of serfactant.

**Study into effect of sequence addition.** Fig. 12 is indicative that the order of addition of the reaction components used in the previous runs is correct and that any changes in the order of addition have a negative impact on the

absorption or the reaction. These two medicinal compounds are represented by the symbol (D) and the oxidizing agent (NBS), while the acidic medium (A) and the dye (B.b).



Fig. 12. Effect of sequence addition

**Final Absorption Spectrum:** Fig. 13 below shows the final absorption spectra for the two drugs in optimal conditions. Table 5 presents a

summary of the best conditions achieved during the evaluation of the two drugs.



Fig. 13. Absorbance of tested substances where (A) Absorption spectrum of 100 μg/mL of Brilliant Blue against blank solution; (B) Absorption spectrum of 6 μg/mL of methyldopa against blank solution; (C) Absorption spectrum of 6 μg/mL dopamine HCl against blank solution; (D) Absorption spectrum of mock solution against distilled water.

<b>Tuble 5.</b> Summary of the optimal conditions:				
Experimental condition	Dopamine. HCl, Methyldopa			
Wavelength (nm)	550			
Color	Blue			
Conc. of CH <sub>3</sub> COOH M	3			
Volume of CH <sub>3</sub> COOH(mL)	1			
N-Bromosuccinimide $(2 \times 10^{-3})$ M amount(mL)	1.2			
Coomassie Brilliant blue	100			
Oxidizing time(min)	10			
Development time after dilution(min)	10			
Stability period (min)	120			

**Table 5.** summary of the optimal conditions.

The calibration plot is as follows: increasing volumes of (0.05-1.8) and (0.05-1.3) mL of dopamine and methyldopa were placed in two sets of separate 10 mL flasks to and cover the concentration ranges shown in Table 6. After adding the remaining ingredients, absorbance was measured at 550 nm against a blank. Fig. 14 shows that the method follows Beer's law within the concentration ranges of  $(0.05 - 18) \mu g/ml$  for dopamine hydrochloride  $(0.05 - 13) \mu g/ml$  for methyldopa. However, a negative bias is observed above the upper limit of these ranges. The limit of detection (LOD) and the limit of quantification (LOQ) were calculated using six replicates of the lowest concentration of the drug compounds. To evaluate the sensitivity of the method, the molar absorbance values and Sandell's sensitivity were calculated, with high values showing that the method exhibits high sensitivity. The linear correlation coefficient values of 0.997 and 0.9995 for dopamine and methyldopa indicate excellent linearity of the standard curves.



Fig. 14. Standard curves for estimation of methyldopa and dopamine.

Measured values	Variable of dopamine. HCl	Variable of methyldopa
Beers law limits (µg/mL)	0.5-18 (µg/mL)	0.5-13(µg/mL)
Molar absorptivity (L.mol <sup>-1</sup> .cm <sup>-1</sup> )	$1.74 \times 10^{4}$	$2.97 \times 10^{4}$
LOD(µg/ml)	0.0514	0.0377
LOQ(µg/mol)	0.155	0.114
Shandell's Sensitivity mg.cm <sup>-2</sup>	0.0108	0.0080
Determination Coefficient(R <sup>2</sup>	0.9970	0.9995
Slope	0.0918	0.125
Intercept	-0.0135	-0.0311

Table 6.	Summary	of	statistical	values.
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Accuracy and precision. The results obtained demonstrate that the method exhibits good accuracy and precision. The recovery rates for dopamine hydrochloric acid and methyldopa were found to be 99.96% and 101.48%, respectively. These values indicate that the method is capable of accurately quantifying the

target compounds. Furthermore, the mean relative standard deviation (RSD) was calculated and found to be less than 1.50%. This low RSD value suggests a high level of indicating precision, that the method consistently yields reliable and reproducible results (Table 7).

Table 7. Accuracy and precision

	1 401	e mineeurue y t	ind precision.		
Drug	Amount	Amount	Recovery	average	RSD
	added	found	%	recovery	%
				%	

### **COOMASSIE BRILLIANT BLUE STAINING**

	1	0.98	98.42		3.66
Dopamine	6	6.07	101.16	101.48	0.823
	13	13.63	104.87		0.807
	2	1.94	97.3		2.83
Methyldopa	8	8.07	100.8	99.99	0.371
	11	11.19	101.8		0.178

method Application of the to pharmaceutical preparations. The developed method was utilized to quantify the two medicinal compounds different in pharmaceutical preparations, specifically dopamine hydrochloric injection and methyldopa tablets. The application of the

method followed the procedure outlined in Table 5. The obtained results, summarized in the Table 8 below, demonstrate that the developed method is suitable for directly analysing the mentioned drugs in their respective formulations.

Table 8. Estimati	on of dopami	ne and methy	yldopa in pharm	naceutical p	reparations.
Dhammaaautiaal	Certified	Amount	Decovery*	Average	Drug

Pharmaceutical preparation	Certified Value	Amount present μg/mL	Recovery* (%)	Average recovery (%)	Content found* (mg)
Donamine		1	100.97		
ampoule	200	3	103.33	101 25	202.5
(Turkey)	mg/5mL	6	100.5	101.25	202.5
(Turkey)		13	100.2		
Donomino		1	96.29	98.48	196.96
Dopannie	200 mg/5mL	2	96.94		
(Turkov)		6	100.5		
(Turkey)		13	100.2		
	250 mg	2	98.13		248.02
Methyldopa		4	98.94	00.21	
tablet-(Iraq)		8	100.4	99.21	
		11	99.4		
		2	96.15		244.07
Methyldopa	250	4	96.84	07.62	
tablet-(Egypt)	250 mg	8	98.98	97.03	
		11	98.55		

**Evaluation of the proposed method.** To demonstrate the efficacy of the proposed spectrophotometric method and its ability to accurately estimate the studied pharmaceutical compounds, while ensuring the absence of interference from additives in their pharmaceutical formulations, the standard method approved in the British Pharmacopoeia was applied. This method involved dissolving the pharmaceutical formulation of methyldopa tablets in glacial acetic acid with perchloric acid, followed by potentiometric titration to specify the endpoint. A statistical comparison and evaluation were conducted between the proposed spectrophotometric method for estimating methyldopa in tablet formulations and the standard method using four degrees of freedom. This was undertaken to assess the accuracy and validity of the analytical application of the proposed method, using t-test and F-test at a confidence level of 95%. Results of this assessment are shown in Table 9.

Table 9. Evaluation of the proposed method

PharmaceuticalRecovery % $t_{exp}$ $F_{tc}$ Preparation
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	Present method	Standard method		
Mathuldona Diala Irag	98.13	98		
Methyldopa Diala- Iraq	98.9	98.5	1.155	1.96
	99.4	98		
	100.4	99		
	98.9	98		
Methyldopa Cairo- Egypt	98.5	98	0.369	7.29
	96.1	98.5		
	96.8	99		

Application of the standard addition method. The standard addition method was used to prove that the proposed reaction for the determination of dopamine hydrochloride free from interference from additives in pharmaceutical formulations, as shown in Fig. 15,16 and Table 10 below show that the standard addition method agrees well with the proposed method, indicating a satisfactory selectivity of the method.



Fig. 15. Standard addition methods for  $1,2 \mu g/ml$  of dopamine.



Fig. 16. Standard addition methods for  $3,6 \mu g/ml$  of dopamine.

Table 10. Summary of standard addition methods for dopamine hydrochloride.

Pharmaceutical Preparation	Certified value	Amount Present (µg/mL)	Recovery %	Drug content (200mg/5mL)
Donominahovan	200mm ~/5mm I	1	97.13	194.26
Dopamine haver	200mg/5mL	2	96.39	192.78

Dopamine feresenius	3	98.18	196.36
	6	97.68	194.36

**Proposed chemical reaction mechanism.** It is suggested that a bromination process take place through the calculated increase of the oxidizing agent N-Bromosuccinimide with the two medicinal compounds, then the remaining amount of it works to bleach the colour of dye, as the absorption of the dye is equal to the estimated two drugs, and therefore their concentration can be known indirectly (Fig.17):



R= CH<sub>2</sub>CH<sub>2</sub>NH<sub>2</sub> = Dopamine

R = CH<sub>2</sub>CHNH<sub>2</sub>COOH = Methyldopa

Fig. 17. Proposed chemical reaction mechanism

#### Conclusion

The present method performs the assessment of dopamine and methyldopa in water with high precision and efficiency in short steps and enables the successful application of the method in pharmaceutical formulation. It is also possible to evaluate many drugs and compounds that undergo oxidative processes using a standard Coomassie Brilliant Blue curve. Finally, the method is very inexpensive and has high sensitivity.

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**Author Contributions**: Each of the individuals named in the study contributed equally to the work and to the writing.

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# SPEKTROFOTOMETRİK ANALİZDƏ DOPAMİN HİDROXLORİD VƏ METİLDOPANI MÜƏYYƏN ETMƏK ÜÇÜN İSTİFADƏ EDİLƏN KUMASSİ BRİLLİANT MAVİSİ İLƏ BOYAMA

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**Xülasə:** Kumassi Brilliant Mavisi və N-bromosuksinimidin turş mühitdə katalitik oksidləşmə reaksiyası əsasında, təmiz formalarda və əczaçılıq preparatlarında dopamin və metildopanın təyini üçün spektrofotometrik üsul işlənib hazırlanmışdır. Hazırlanmış üsul yüksək həssaslıq göstərmiş: molyar udma göstəriciləri müvafiq olaraq  $1.77 \times 10^4$  və  $2.97 \times 10^4$  L·mol<sup>-1</sup> ·cm<sup>-1</sup>, Sandell həssaslığı isə müvafiq olaraq 0.0080 və 0.0107 mq·sm<sup>-2</sup> olmuşdur. Dopamin hidroxlorid və metildopanın təxmini miqdarı müvafiq olaraq 0.5-18 mkq/ml və 0.5-13 mkq/ml diapazonunda dəyişmişdir. Hazırlanmış üsul həb və inyeksiya şəklində olan dərmanlara uğurla tətbiq edilmiş, əldə edilən nəticələr orijinal dərmanların tərkibinə uyğun olmuşdur. Hazırlanmış metodika standart əlavə və Britaniya Farmakopeyası standart metodları ilə müqayisə edilmişdir. Tədqiqat dərmanlarının qiymətləndirilməsində işlənib hazırlanmış metodun uğurunu təsdiqləyən əhəmiyyətli fərqlər aşkar edilməmişdir. Eyni zamanda digər statistik məlumatlar da araşdırılmışdır.

Açar sözlər: katalitik oksidləşmə, Kumasi brilliant mavisi, dopamin hidroxlorid, metildopa, spektrofotometrik üsul.

### ОКРАШИВАНИЕ КУМАССИ БРИЛЛИАНТОВЫМ СИНИМ, ИСПОЛЬЗУЕМОЕ В СПЕКТРОФОТОМЕТРИЧЕСКОМ АНАЛИЗЕ ДЛЯ ОПРЕДЕЛЕНИЯ ДОФАМИНА ГИДРОХЛОРИДА И МЕТИЛДОПЫ

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Аннотация: На основе реакции каталитического окисления Coomassie Brilliant Blue (кумасси бриллиантовый синий) и N-бромсукцинимида в кислой среде разработан спектрофотометрический метод определения дофамина и метилдопы в чистых формах и в фармацевтических препаратах. Разработанный метод показал высокую чувствительность: значения молярной абсорбции составили  $1.77 \times 104$  и  $2.97 \times 104$  л· моль<sup>-1</sup> ·см<sup>-1</sup>, а чувствительность Санделла – 0.0080 и 0.0107 мг·см<sup>-2</sup>, соответственно. Расчетные количества дофамина гидрохлорида и метилдопы находились в диапазоне 0.5-18 мкг/мл и 0.5-13 мкг/мл соответственно. Разработанный метод был успешно применен к таблетированным и инъекционным препаратам, и полученные результаты соответствовали исходному содержанию препарата. Разработанный метод сравнивался со стандартным добавлением и стандартным методом Британской фармакопеи, и существенных различий выявлено не было, что подтверждает успешность разработанного метода в оценке исследуемых препаратов. Также были изучены другие статистические показатели.

**Ключевые слова:** каталитическое окисление, кумасси бриллиантовый синий, гидрохлорид дофамина, метилдопа, спектрофотометрический метод.