

## INDIRECT SPECTROPHOTOMETRIC METHOD FOR MICRO DETERMINATION OF METHYLDOPA AND DOPAMINE HYDROCHLORIDE USING ORANGE G

A.J. Hasan<sup>1</sup>, O.A. Sheej Ahmad<sup>2,\*</sup><sup>1</sup>Ninawa University, Mosul, Iraq<sup>2</sup>Chemistry Department, College of Education for Pure Science, Mosul University, Mosul, Iraq  
email: [dr.omar1979@uomosul.edu.iq](mailto:dr.omar1979@uomosul.edu.iq)

Received 25.05.2024

Accepted 02.07.2024

**Abstract:** An accurate and simple indirect spectrophotometric method has been created for the micro-determination of Methyldopa (MDP) and Dopamine hydrochloride (DA) in acidic media. The method was based on the catalytic oxidation reaction between Orang-G and N-bromosuccinimide in an acidic medium which could be used to create an indirect spectrophotometric method for the micro determination of dopamine hydrochloride and methyldopa in pure form and pharmaceutical formulations. The molar absorptivity and Sandell index of MDP and DA were  $(4.9 \times 10^4, 3.7 \times 10^4)$   $\text{l.mol}^{-1}.\text{cm}^{-1}$  and (0.0048, 0.0051)  $\text{ng/cm}^2$  respectively. The correlation coefficient values were 0.9936, 0.9966 with a recovery rate of 99.5% and 99.93%. In addition, the limit of detection (LOD) and limit of quantification (LOQ) were 0.24, 0.1788, and 0.727, 0.5418  $\mu\text{g/mL}$ . The recommended method was effectively applied for the determination of Dopamine hydrochloride and methyldopa in commercial formulation. No interference was experimentally noticed from common pharmaceutical formulation adjuvants. Statistical evaluation of the results with the standard addition method showed an excellent agreement and showed no significant variance in terms of accuracy and precision.

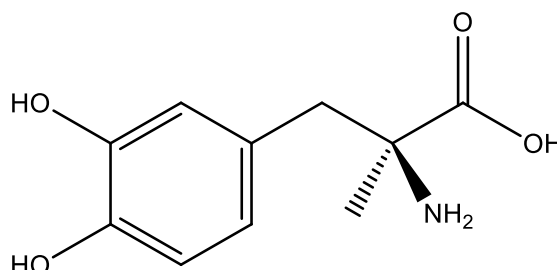
**Keywords:** Methyldopa, Dopamine hydrochloride, Spectrophotometric method, Spectrophotometric determination, Orange G, Pharmaceutical formulations.

**DOI:** 10.32737/2221-8688-2025-1-131-147

## 1. Introduction

Methyldopa (MDP) is a white or colorless, crystalline powder that is used to treat the symptoms of hypertension due to its ability to dilate blood arteries [1]. Chemically referred to as  $\alpha$ -methyl-3,4-dihydroxyphenylalanine, methyldopa (MDP) is a catechol derivative

(catecholamine) that is widely used as an antihypertensive therapy. MDP is an  $\alpha$ 2-adrenoreceptor agonist that acts centrally to lower sympathetic tone and lower blood pressure [2].



**Fig. 1.** Chemical structure of methyldopa

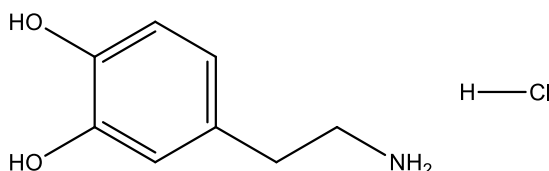
In order to facilitate easier blood flow throughout the body, it relaxes the blood vessels. It is considered one of the most famous medications used to treat the antihypertensive

medications [3]. The fact that methyldopa lowers renal vascular resistance is one of its primary benefits. This occurs because most peripheral tissues limit the decarboxylation of 5-

hydroxytryptophan (5-HTP) and dihydroxyphenylalanine (dopa), which is the precursor of norepinephrine. These studies, however, could not offer conclusive proof about the mode of action [1]. A chiral center can be found in methyldopa (S or R). However, the antihypertensive effect of methyldopa is caused by its S-isomer [4] (Fig.1).

Dopamine (DA), also known by its chemical name 4-(2-aminoethyl)-benzene-1,2-diol (Fig.2), is an essential catecholamine neurotransmitter that is extensively disseminated throughout the central nervous system to facilitate the transmission of [5]. Endogenous catecholamine (DA) functions as a sympathomimetic agent, primarily affecting dopaminergic and  $\beta$ 1-adrenergic pathways at low to moderate dosages, but also having  $\alpha$ -

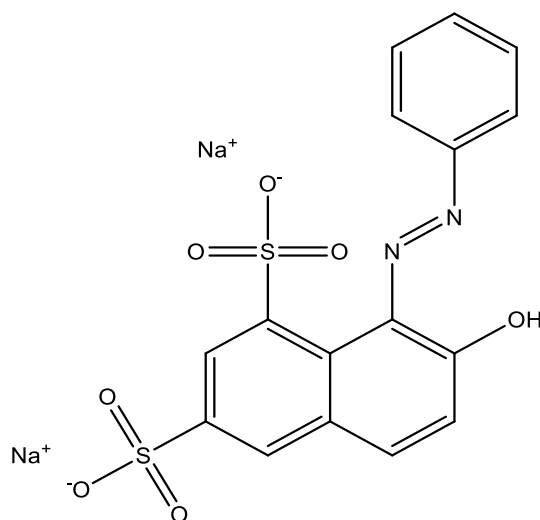
adrenergic effects at higher levels. Correction of hemodynamic abnormalities linked to shock episodes is its intended purpose [6]. High level of Dopamine can cause an invite neurological disease like schizophrenia and Parkinson's. Therefore, DA different important clinical prescriptions for Dopamine can also be prescribed as a treatment for the symptoms of many diseases, such as bronchial asthma, cardiovascular breakdown, hypertension and kidney problems associated with stun scenarios [7]. Other processes cause the kidneys to produce more urine and excrete more salt. It shields intestinal mucosa, lowers the activity of lymphocytes in the pancreas, the digestive system, and the immune system, respectively, and decreases both the generation of insulin and gastrointestinal motility [8].



**Fig. 2.** Chemical structure of Dopamine hydrochloride

Orange G is an organic sodium salt that is frequently mixed with other yellow dyes in an alcoholic solution to highlight the cells in the pituitary and pancreas. It serves as a dye for histology. Frequently, it's used to differentiate between tissue types and cellular structures

under a microscope. As well as, Orange G can also be used for dyeing silk, wool, leather, paper, wood stains, coloring inks and copying pencils. The chemical name of the dye is disodium 7-hydroxy-8-[(E)-phenyldiazenyl] naphthalene-1,3-disulfonate (Fig. 3).



**Fig. 3.** Chemical structure of Orange G dye

The literature on DA and MD shows numerous methods for their determination of

pharmaceuticals. Spectrophotometric methods based on use of reagent such as bromanil,

chloramine-T, 2,6-dichloroquinone-4-chloroimide and 3-aminopyridine [9-12]. As well as, oxidative coupling-based methods for the determination of DA and MD were published [1, 3, 13-15]. In addition, chromatographic method mainly HPLC [16-21] flow injection [22] fluorimetric, chemiluminescence [26, 27], and electrochemical methods were reported [28-34].

UV-Vis molecular absorption spectrometry is considered as simple practical analytical technique for drug quality control. It does not involve costly instrumentation, toxic

solvents like chromatography and solvent extraction.

Therefore, the simple and rapid analysis of Catechol drugs is still considered important. In keeping with our interest in the validation and development of straightforward, sensitive, and affordable spectrophotometric techniques for Catechol medications (dopamine hydrochloride, methyldopa), the current investigation was completed.

*The object of this work* is to enhance and improve analysis methods for the determination of (DA, MD) to be more rapid and sensitive.

## 2. Experimental part

**Drug solution.** DA and MD were freshly prepared at a concentration of (100 µg/mL) by weighing 0.010 g of the drug's compound and dissolving it in a distilled water then placed in an ultrasonic to complete the dissolution process. The drug solution was stored in refrigeration and remains stable for ten days.

**N-bromosuccinimide solution** was prepared at a concentration of  $5 \times 10^{-3}$  M by dissolving 0.0890 grams of the oxidizing agent in 100 mL of distilled water. NBS solution could react with light and cause decomposition, therefore it should be kept in an amber bottle to reduce the amount of light that the solution could exposed to. It remained stable for ten days by placing it in an opaque bottle and keeping it in the refrigerator.

**Orange-G dye solution** was prepared at a concentration of 200 µg/mL by dissolving 0.0200g in a small beaker. It was placed in an ultrasonic for 5 min to complete the dissolution of the substance. It was taken up with doubly distilled water and transferred to a 100 mL volumetric flask, diluting to volume. It remained stable for a week in the refrigerator.

**Hydrochloric acid solution.** It was prepared at a concentration of (1M) by diluting 8.3 ml of concentrated acid (12 M) with distilled

water in a volumetric flask of 100 mL.

**Methyldopa tablets.** Ten tablets (methyldopa tablet, accord UK) of the medicinal preparation were taken and ground well, then the equivalent of the weight of one tablet of the medicinal preparation was carefully weighed, and dissolved with water in a glass beaker, then transferred to a 50ml volumetric flask, and the volume was completed with distilled water to the mark, then filter the solution into a 100ml volumetric flask. After completing the filtration process, the filtrate is supplemented with distilled water up to the mark. From which a solution with a concentration of 100 µg/mL was prepared, and different concentration (4, 6, 10) µg/mL were taken, and the results were as shown in the Table 7.

**Dopamine hydrochloride injection.** A concentration of 10000 µg/mL was obtained by diluting The content of five dopamine Fresenius ampoules (Fresenius kabi Austria) (each ampoule contains 200 mg/5 mL of dopamine HCl) in a 100 mL volumetric flask. A solution with a concentration of 100 µg/mL was prepared, and different concentrations of (4.0,6.0,10) µg/mL were taken. The results are shown in the Table 7.

## 3. Results and discussion

To evaluate the rapidity of analysis and straightforwardness of determination of Drugs at low and high concentrations at affordable costs, primary experiments were performed to

demonstrate the opportunity of using Orange-G dye in drug determination. The spectrum of the Orange- G solution that was prepared in water was scanned between 350-700 nm as shown in

Figure 4. It was found that the dye gives a blank solution. maximum absorption at 478 nm versus the

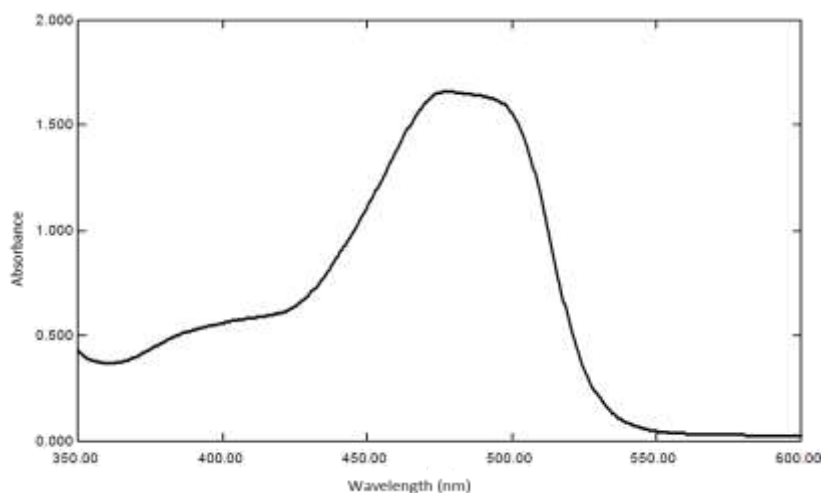


Fig. 4. Absorption spectrum of Orange-G dye

To determine the optimal amount of orange G dye from which the studied drug compounds can be estimated, which follows Beer's law, increasing volumes of the dye solution with a concentration of 200  $\mu\text{g/mL}$  was added to 10 mL volumetric flasks, and the volume was supplemented with distilled water to the mark. As can be seen in Figure 4, orange

G shows maximum absorbance at 478 nm. A linear calibration curve in the range (1-40  $\mu\text{g/mL}$ ) has been plotted to determine the highest possible concentration of the dye used. According to the calibration curve, as shown in Figure 5 the concentration was proven to be 40  $\mu\text{g/mL}$ .

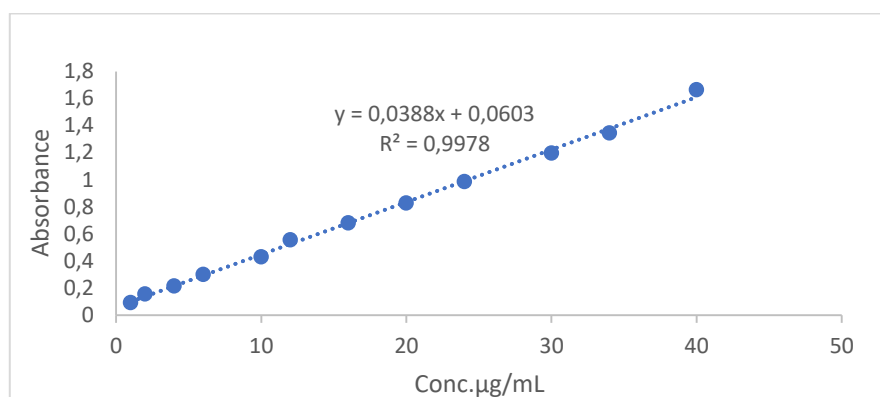
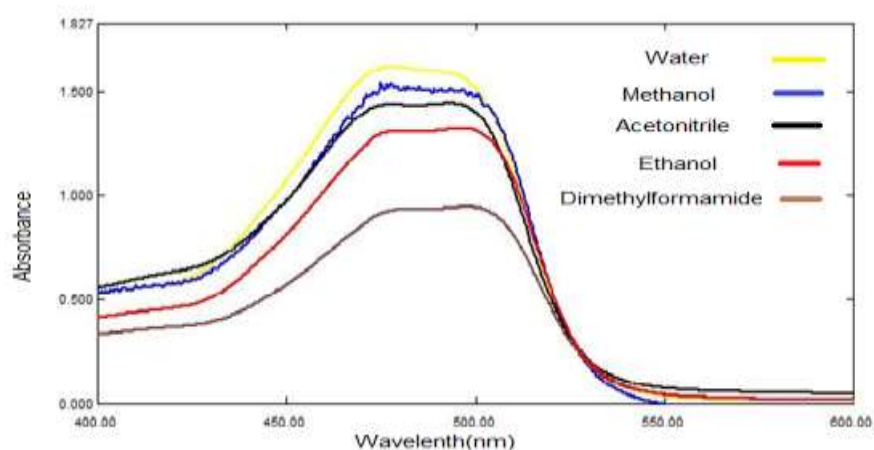


Fig. 5. Calibration curve of Orange-G dye

#### Setting the optimal conditions for conducting drug analysis.

**Study the effect of solvent on color intensity and wavelength.** Solvents have an important effect on the absorption spectra in the UV-Visible spectroscopy technique. Solvent polarity and polarizability could lead to shifts in the absorption bands and change the intensity of the absorption peak. To ensure accurate and

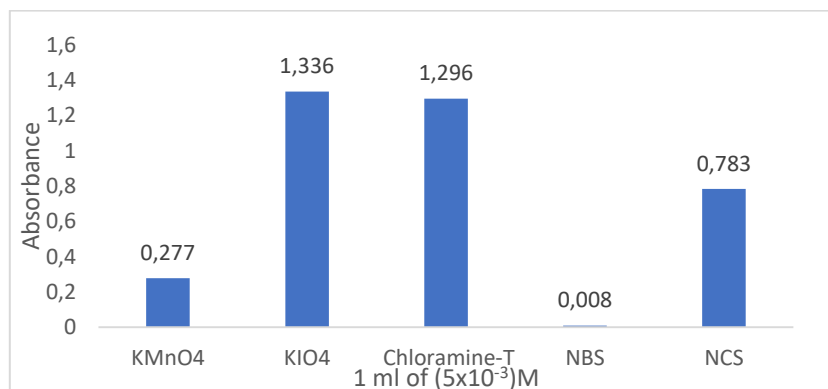
reliable measurements of the absorption spectra, a group of solvents were tested such as water, ethanol, methanol, acetonitrile, and dimethylformamide). In 10 mL volumetric flasks, the volume was completed with the mentioned solvents. The results in Figure 6 indicate that the water gives the highest intensity value.



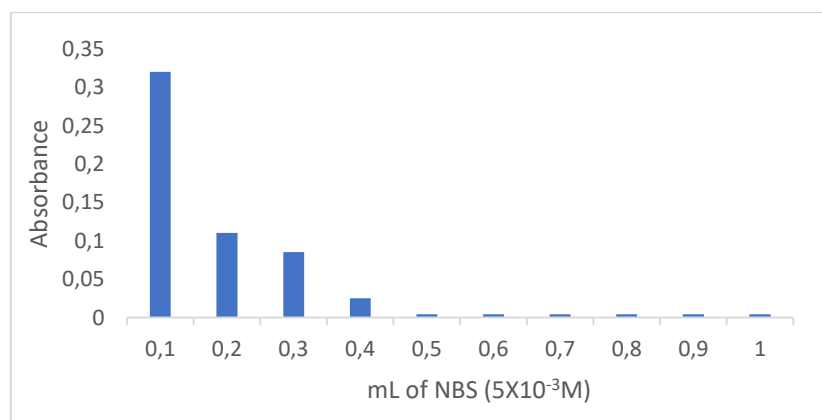
**Fig. 6.** Absorption spectrum of solvents

**Effect of oxidizing agent.** Choosing the appropriate oxidizing agent is an important factor for developing a spectrophotometric method. Therefore, different types of oxidizing agents such as (N-Bromosuccinimide (NBS), Chloramine-T, N-Chlorosuccinimide (NCS), Potassium permanganate, and Potassium periodate have been tested for bleaching dye. The optimal volume of dye was added then 1

mL of the concentration of ( $5 \times 10^{-3}$ )M of oxidizing agents in acidic medium of HCl was used. The reaction was waited to complete for five minutes, shaking then completing the volume with distilled water to the mark. The absorption and color intensity of the chemical reaction was increase when NBS used compare to other oxidizing agent as shown in Figure 7.



**Fig. 7.** Type of oxidant agent

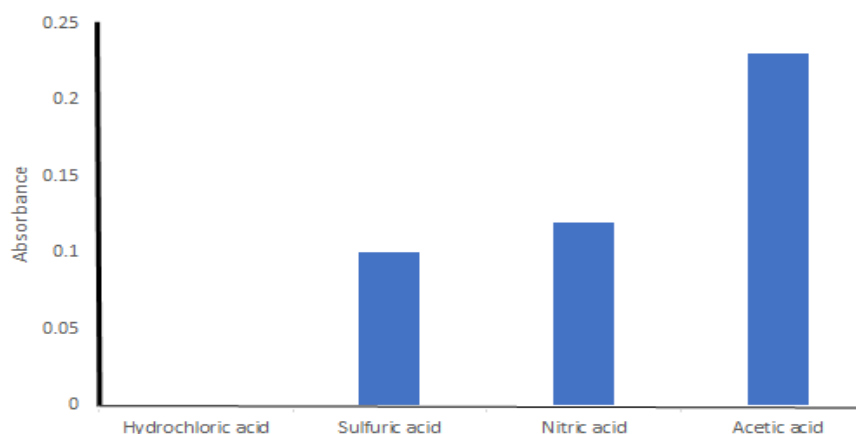


**Fig. 8.** Choose the amount of oxidizing agent

**The effect of the amount of oxidizing agent on the shortening of the dye Orange-G.** The fixed amount of Orange-G dye was added, followed by the addition of increasing volumes of the oxidizing agent N-Bromosuccinimide ( $5 \times 10^{-3}$  M) in the acidic medium HCl at a concentration of (1M) and a volume of (1 mL) Wait for five minutes, then supplement the volume with distilled water to the mark, with shaking, wait for five minutes, and measure the absorption. It is clear from Figure 8 that when adding 0.5 mL of the oxidizing agent, it gives the same shortness as 1 mL of the oxidizing agent, so it was approved.

#### Choose the most appropriate acidic

**medium.** Several acids were studied, namely HCl, H<sub>2</sub>SO<sub>4</sub>, CH<sub>3</sub>COOH, and HNO<sub>3</sub>, in order to determine the shortness of the dye in the optimal acidic medium. (1M) and (1mL) of acids were added individually in (10mL) volumetric flasks, followed by adding the fixed amount of (2mL) of the dye and the oxidizing agent. Wait for five minutes, then complete the volume with distilled water to the mark, shake, wait for five minutes, and measure the absorption. It turns out that the most suitable acid to shorten the Orange-G dye is hydrochloric acid, and the results are shown in Figure 9.



**Fig. 9.** Effect of different types of acids

**Study of the optimal amount of acid.** After selecting HCl as the best medium, the next step is to select the best volume of acid which

can gives lowest absorption value. Results in Table 1 indicated that 1 mL of 1M HCl is the best volume.

**Table 1.** Effect of Hydrochloric acid volume

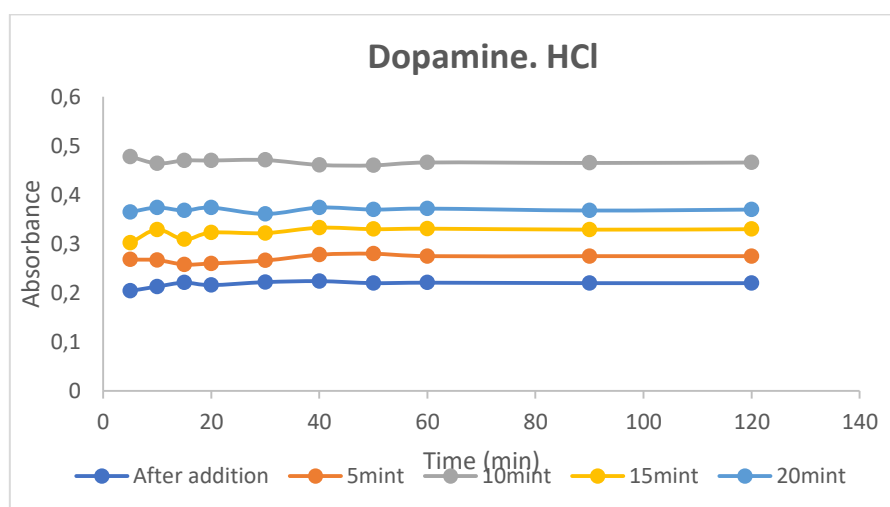
Volume mL of acid HCl (1M)	Absorbance
0.1	0.451
0.2	0.402
0.3	0.35
0.4	0.32
0.5	0.295
0.6	0.25
0.7	0.16
0.8	0.102
0.9	0.081
<b>1.0</b>	<b>0.000</b>
1.2	0.000
1.5	0.033

**The effect of the oxidation time of Drug compounds and Orange G dye.** The effect of the oxidation time is an important factor that can affect the absorbance. For this reason, the time for the oxidation of the two drugs (DA, MDP) was studied to reach the optimal time for the completion of the oxidation process for both

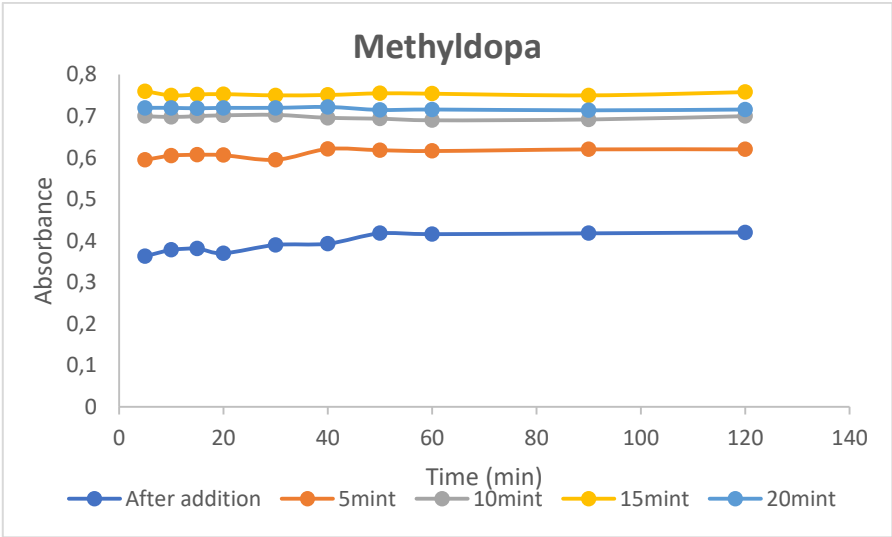
drugs. A 0.5 mL of NBS ( $5 \times 10^{-3} \text{M}$ ) was reacted with a fixed amount of each of the two compounds (MDP, DA)  $5 \mu\text{g/mL}$  in the presence of the acidic medium 1ml HCl of concentration 1M at different times. After setting optimal conditions, which can influence the oxidation time.

**Table 2. The effect of time on the oxidation of drugs and Orange-G dye**

Before adding Orange-G & dilution	Standing time (min)									
	After adding Orange G & dilution									
	5	10	15	20	30	40	50	60	90	120
<b>Dopamine. HCl</b>										
direct addition	0.204	0.213	0.221	0.216	0.222	0.224	0.220	0.221	0.220	0.220
5 min	0.268	0.267	0.258	0.260	0.266	0.278	0.280	0.275	0.275	0.275
10 min	0.478	0.470	0.470	0.470	0.471	0.467	0.460	0.466	0.465	0.466
15 min	0.302	0.329	0.309	0.323	0.322	0.333	0.330	0.331	0.329	0.333
20 min	0.365	0.374	0.368	0.374	0.361	0.374	0.370	0.372	0.368	0.370
<b>Methyldopa</b>										
direct addition	0.363	0.378	0.381	0.370	0.390	0.393	0.418	0.416	0.418	0.42
5 min	0.595	0.605	0.607	0.606	0.596	0.621	0.618	0.616	0.620	0.62
10 min	0.7	0.698	0.7	0.702	0.703	0.696	0.694	0.69	0.692	0.7
15 min	0.760	0.750	0.757	0.753	0.747	0.751	0.755	0.754	0.750	0.758
20 min	0.72	0.72	0.719	0.72	0.72	0.722	0.715	0.716	0.714	0.716





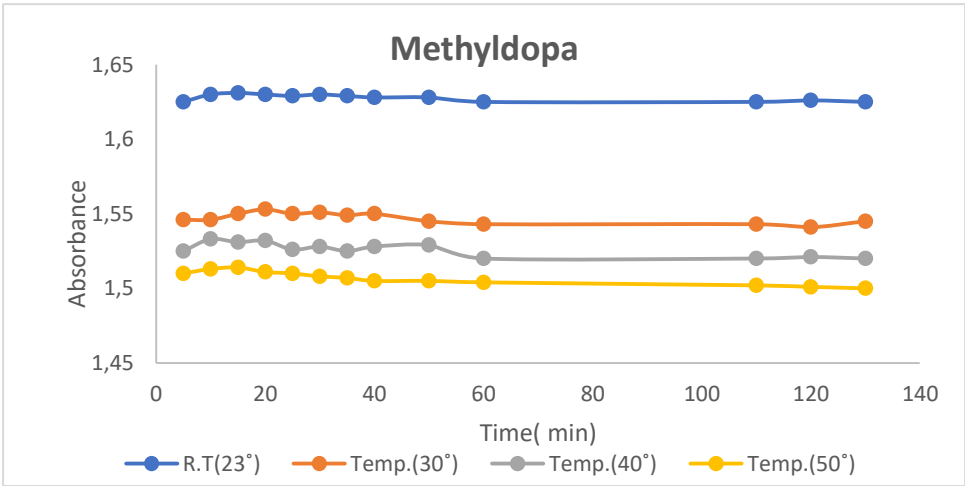


**Fig. 10.** The effect of oxidation time on the of pharmaceutical compounds and Orange-G dye

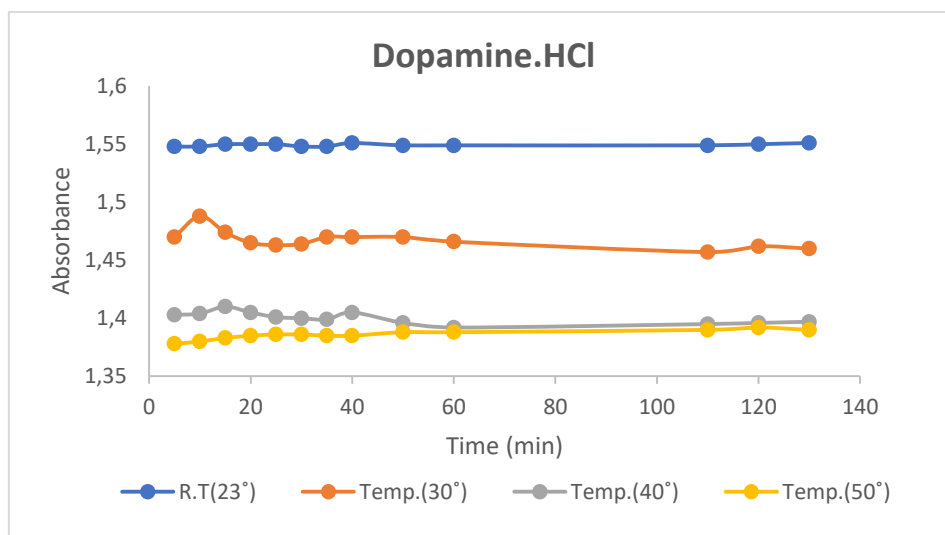
The results in Table 2 and Figure 10 indicated that the oxidation time before dilution is 10 minutes. For DA the oxidation time before dilution is 15 minutes for methyldopa. For this reason, the oxidation time was fixed for use in subsequent experiments.

**Table 3.** The effect of temperature on the oxidation reaction of Orange-G dye and its stability when determining the studied drugs

Temp (C°)	Absorbance/standing time (min)												
	5	10	15	20	25	30	35	40	50	60	110	120	130
Dopamine. HCl													
R.T	1.548	1.548	1.550	1.550	1.550	1.548	1.555	1.553	1.549	1.545	1.549	1.545	1.545
30	1.47	1.488	1.474	1.465	1.463	1.464	1.47	1.47	1.47	1.466	1.457	1.462	1.46
40	1.403	1.404	1.41	1.405	1.401	1.4	1.399	1.405	1.396	1.392	1.395	1.396	1.397
50	1.378	1.38	1.383	1.385	1.386	1.386	1.385	1.385	1.388	1.388	1.39	1.392	1.39
Methyldopa													
R.T	1.625	1.63	1.631	1.63	1.629	1.63	1.629	1.628	1.628	1.625	1.625	1.626	1.625
30	1.546	1.546	1.55	1.553	1.55	1.551	1.549	1.55	1.545	1.543	1.543	1.541	1.545
40	1.525	1.533	1.531	1.532	1.526	1.528	1.525	1.528	1.529	1.52	1.52	1.521	1.52
50	1.51	1.513	1.514	1.511	1.51	1.508	1.507	1.505	1.505	1.504	1.502	1.501	1.500







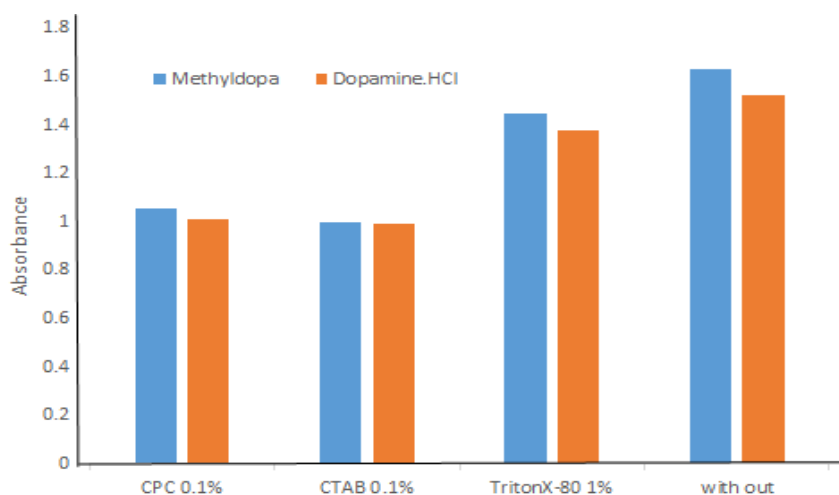
**Fig.11.** The effect of temperature on the oxidation of pharmaceutical compounds and dye

**The effect of temperature on the oxidation reaction and the stability of product.** The effect of different temperatures on color intensity and wavelength value under the conditions which previously optimized has been evaluated.

The results in the Table 3 and Figure 11 showed that the laboratory temperature (23°) is the best temperature for the oxidation reaction and the color and absorbance remain stable for

more than hour. It was also observed that absorption decreases as the temperature increases.

**Effect of surfactants.** Negative, positive, and neutral surfactants have been added to the reaction mixture to point out the effect of surfactants on absorption value. The results presented in Figure 12 show the inefficiency of these surfactants, and therefore they were not adopted in subsequent experiments.



**Fig. 12.** Effect of surfactants

**Effect of addition sequence.** A change in color intensity was observed when the sequence of addition between the drug, oxidizing agent,

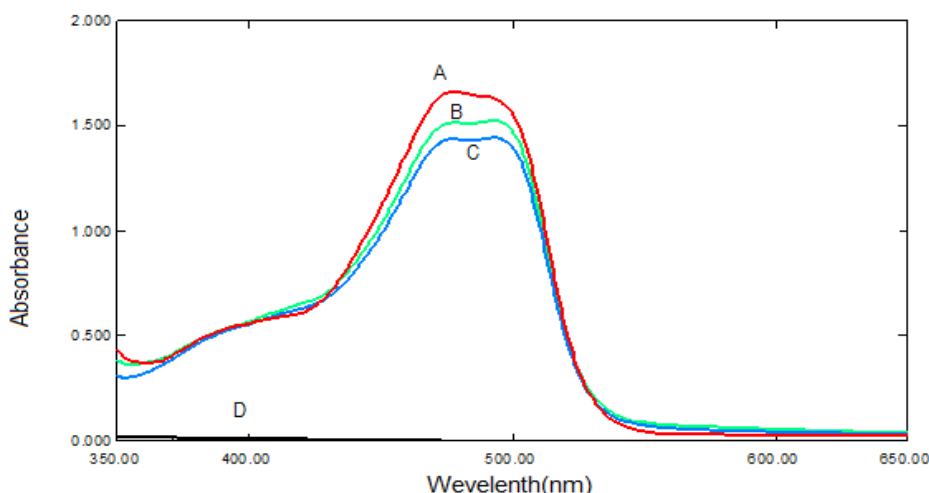
acid, and dye. As shown in Table 4, order No. 1 was the best order of addition which was already approved in previous experiments.

**Table 4.** Effect of the addition sequence of methyldopa

Order number	Reaction components	Absorbance
I	S+NBS+A+Dye	1.567
II	S+A+NBS+Dye	0.245

III	S+NBS+Dye+A	1.452
IV	Dye+A+S+NBS	0.083
V	NBS+S+A+Dye	1.101

**Final absorption spectrum.** Figure 13 shows that the Orange-G gives the highest absorption ( $\lambda_{\max}$ ) at 478nm. In addition, the blank has no peak or absorption value in the same region.



**Fig. 13.** Final absorption spectrum for the determination of methyl dopa and dopamine hydrochloride. A = Orange-G (40  $\mu\text{g/mL}$ ), B = dye in the presence of methyl dopa (9  $\mu\text{g/mL}$ ) and NBS, C = dye in the presence of dopamine hydrochloride (10  $\mu\text{g/mL}$ ) and NBS, D = blank

**Table 5.** A summary of the optimal conditions for both drugs

Parameter	Value	
	Methyl dopa	Dopamine. HCl
$\lambda_{\max}$ (nm)	478	478
HCl(1M) (mL)	1.0	1.0
N-Bromosuccinimide ( $5 \times 10^{-3}\text{M}$ ) (mL)	0.5	0.5
Temp. of the oxidation ( $^{\circ}\text{C}$ )	23	23
Orange-G( $\mu\text{g/mL}$ )	40	40
Oxidizing time(min)	15	10
Development time after dilution (min)	5	5
Stability period (min)	More than 120 min	

#### Calibration curve for determination of Dopamine hydrochloride and Methyl dopa.

Using the optimal conditions, calibration curve has been set it up to determine DA, MDP. Figure 14 and 15 show a linear correlation between absorbance and drug concentration.

The statistical results related to the standard curve are presented in the Table 6. Based on the molar absorptivity value and sandell index, results showed high sensitivity and the relationship between absorption and concentration was an excellent.

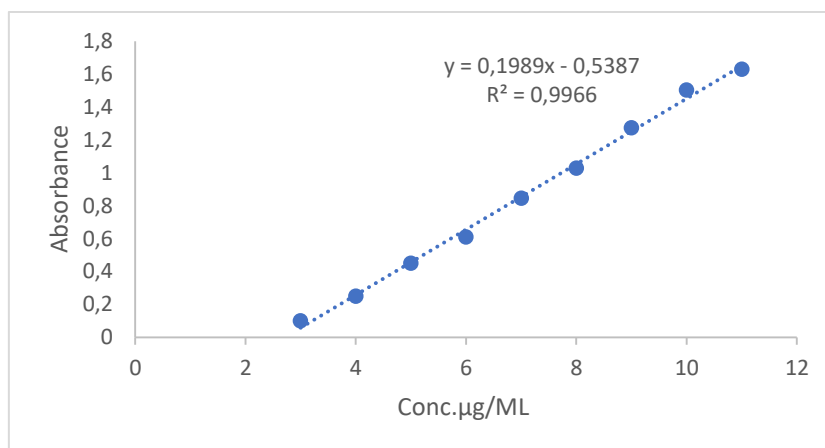


Fig. 14. Calibration curve of Dopamine. HCl

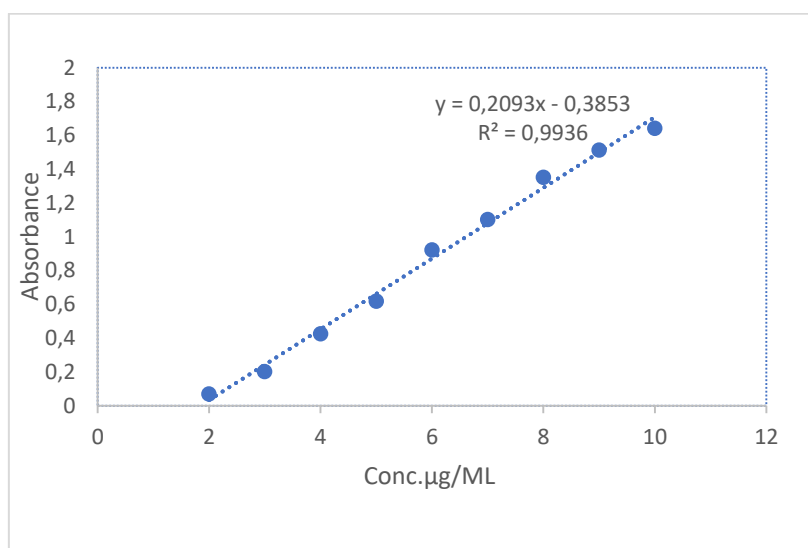


Fig. 15. Calibration curve of Methyldopa

Table 6. Summary of statistical values

Parameter	Methyldopa	Dopamine. HCl
Linearity range (µg/ml)	2.0-10	3.0-11
Molar absorptivity ( $\text{l.mol}^{-1}.\text{cm}^{-1}$ )	$4.9 \times 10^4$	$3.7 \times 10^4$
Sandell Sensitivity ( $\text{ng/cm}^2$ )	0.0048	0.0051
LOD (µg/ml)	0.24	0.1788
LOQ (µg/ml)	0.727	0.5418
Intercept	- 0.3853	- 0.5387
Slope	0.2093	0.1989
Determination coefficient ( $R^2$ )	0.9936	0.9966

**Accuracy and compatibility of the method.** The accuracy and compatibility of the method were evaluated by calculating recovery and the relative standard deviation for both drug compounds. The results indicated that the method has good accuracy and compatibility,

with an average recovery rate that ranged between 99.93% and 99.5%, with a relative standard deviation of less than 1.5% and 0.76% for the two drug compounds (dopamine hydrochloride and methyldopa), respectively.

**Table 7. Accuracy and precision of the method**

Drug	Conc. of drug (µg/mL)		Recovery* (%)	Average recovery (%)	RSD* (%)
	Added	Found			
Dopamine HCl	4	3.98	99.5	99.93	1.48
	6	5.85	97.5		0.45
	10	10.28	102.8		0.26
Methyldopa	4	3.9	97.5	99.5	0.75
	6	6.24	104.0		0.72
	10	9.7	97		0.23

\*Average of Six determinations.

#### Application of the developed method to pharmaceutical preparations:

The results illustrate in Table (8) indicated that the created method is considered successful in determining drug compounds in their

pharmaceutical formulations, as it agrees well with the original content of the two preparations, as the average recovery percentage reached between 99.2% and 101.5%.

**Table 8. Quantitation of dopamine hydrochloride and methyldopa in their pharmaceutical formulations using the proposed method**

Pharmaceutical preparation	Certified value	Amount present (µg/mL)	Drug content found* (mg)	Recovery* (%)	Average recovery (%)
Methyldopa tablet-accord-UK	250 mg	4	246.25	98.5	99.2
		6	255.75	102.3	
		10	242.5	97	
Dopamine Fresenius ampoule-Austria	200 mg	4	201	100.5	101.5
		6	204	102	
		10	204	102	

\*Average of six determinations.

**Evaluation of the proposed method.** To evaluate the proposed spectrophotometric method for estimating methyldopa in the pharmaceutical preparation, a statistical comparison was conducted against the potentiometric titration method outlined in the British Pharmacopoeia. The standard method involved titrating a solution of the dissolved

pharmaceutical preparation in glacial acetic acid and Perchloric acid to determine the endpoint. The accuracy and validity of the proposed method were assessed using t-tests and F-tests at a 95% confidence level, adhering to the statistical principles outlined below. The results of this comparison are presented in the Table 9.

$$\pm t = \frac{\bar{X}_1 - \bar{X}_2}{S_{pooled}} \sqrt{\frac{N_1 N_2}{N_1 + N_2}}, \quad S_{pooled} = \sqrt{\frac{\sum(X_1 - \bar{X}_1)^2 + \sum(X_2 - \bar{X}_2)^2}{N_1 + N_2 - 2}}, \quad F = \frac{S_1^2}{S_2^2}$$

$$S^2 = \frac{\sum(X_i - \bar{X}_i)^2}{N - 1}$$

**Table 9. Comparing the accuracy of the developed method for determining methyldopa in its pharmaceutical preparation with the standard method.**

Pharmaceutical preparation	Recovery (%)		t <sub>exp.</sub>	F <sub>test</sub>
	Present	Standard		

	method*	method		
Methyldopa tablet- accord - UK	98.5	100.91	0.568	16.13
	102.3	99.73		
	97	99.73		

\*Average of six determinations.

#### Application of the standard addition method.

To prove the efficiency of the proposed method and its success in determining the two drug compounds and their freedom from the interference of drug additives in pharmaceutical preparations. The standard pharmaceutical addition method for dopamine hydrochloride

and methyldopa was applied. From Figures 16,17 and Table (10) it can be inferred that the standard addition method is compatible with the proposed method well within the acceptable range of error ( $\pm 5\%$ ), which indicates that the method has satisfactory selectivity.

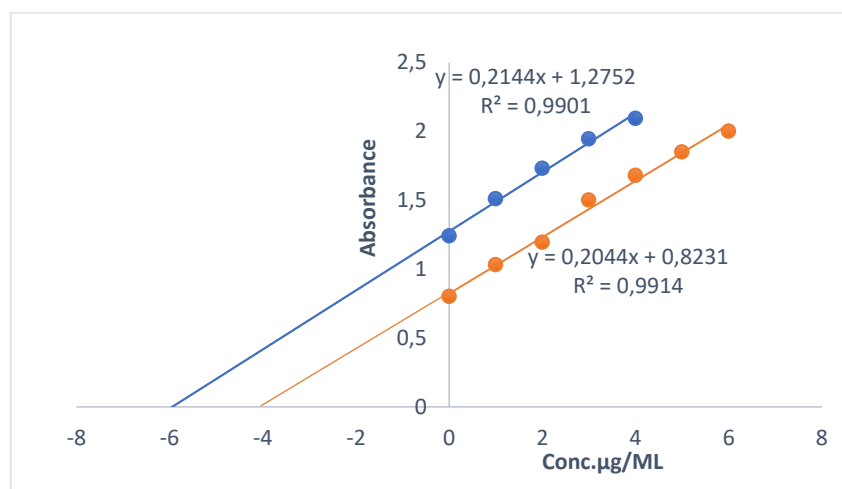


Fig. 16. Standard addition curve for DA in pharmaceutical preparation

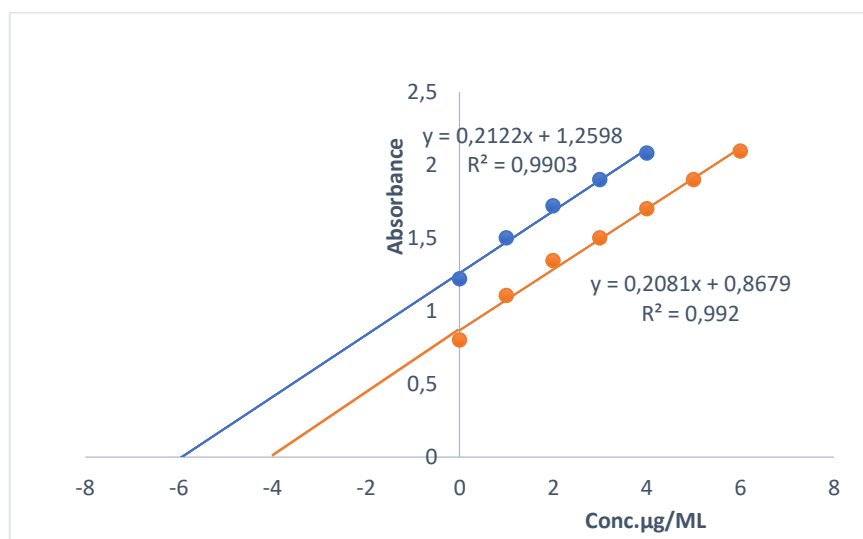


Fig. 17. Standard addition curve for MDH in a pharmaceutical preparation

Table 10. Standard addition method for determining the studied drug compounds

Pharmaceutical preparation	Certified value	Amount present (µg/mL)	Recovery (%)	Drug content found (mg)	
				Present method*	Standard addition method
Methyldopa					

Methyldopa-UK	250 mg	4	104.25	246.25	260.62
		6	98.95	255.75	247.37
Dopamine. HCl					
Dopamine Fresenius	200mg	4	100.67	201	201.34
		6	99.13	204	198.26

\*Average of Six determinations.

**Comparing the proposed method with other methods.** The proposed method has been compared with published methods. Table 11 revealed that the proposed method has high

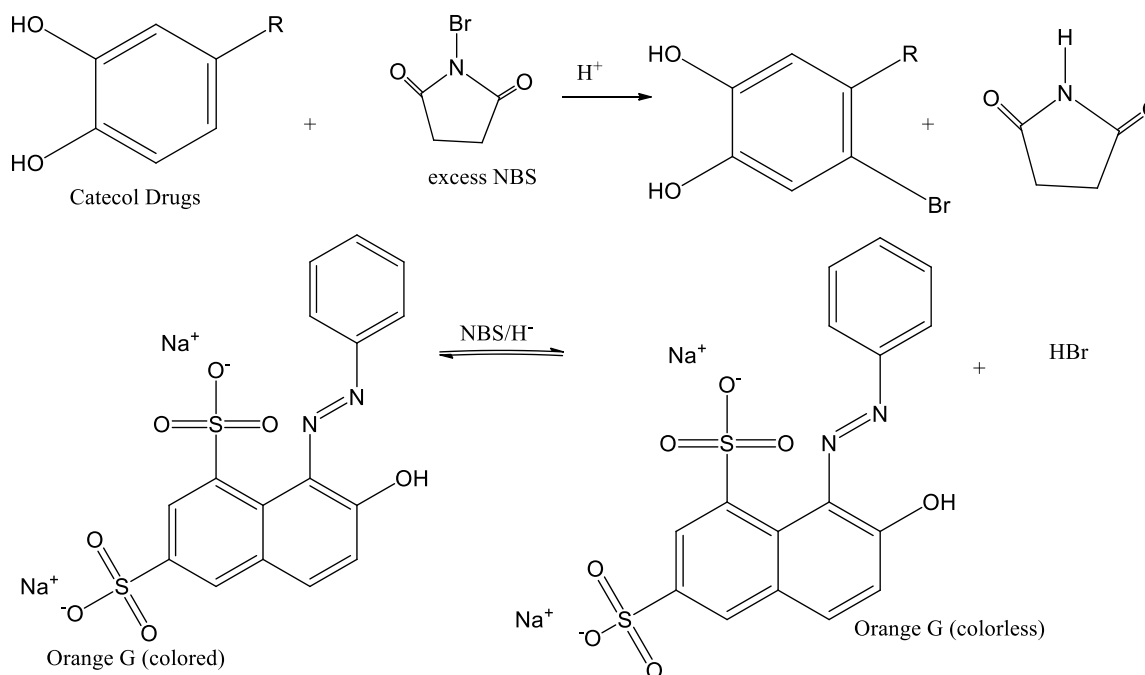
sensitivity in terms of molar absorptivity. In addition, it does not require the use of a buffered solution or temperature control.

**Table 11.** Comparing the proposed method with other methods

Analytical parameter	Present method		Literature method [3]	Literature method [34]
	Methyldopa	Dopamine.HCl	Methyldopa	Dopamine.HCl
$\lambda_{\text{max}}$ (nm)	478	478	481	5 52
Method	Oxidation	Oxidation	Oxidative Coupling	Oxidation
Linearity ( $\mu\text{g}/\text{mL}$ )	2.0-10	3.0-11	5.0-40	1.0-7
Molar absorptivity ( $\text{l.mol}^{-1}.\text{cm}^{-1}$ )	$4.9 \times 10^4$	$3.7 \times 10^4$	$6.082 \times 10^3$	$2.7213 \times 10^4$
Recovery (%)	99.5	99.93	101.02	99.69
RSD (%)	0.56	0.73	1.008	2.2

**The suggested mechanism of the chemical reaction.** Based on the literature and mechanics

of the oxidation process using N-bromosuccinimide (NBS).



**Fig. 18.** Proposed reaction mechanism

The NBS can be considered as an oxidation and bromination agent in chemical reactions at the same time. It is suggested that the bromination process can take place through the increase of the oxidizing agent N-Bromosuccinimide with the two medicinal compounds, then the

remaining amount of N-Bromosuccinimide works as a bleaching agent for the dye color ye, as the absorption of the dye is equal to the estimated two drugs, and therefore their concentration can be known indirectly as shown in Figure 18.

### Conclusion

Spectrophotometry is a superanalytical technique for the characterization and analysis of some chemical compounds in different fields including chemistry and pharmacy. This is working with different ideologies which are planned through numerous instrumentation techniques such as UV-Vis spectrophotometry. These analytical methods are simple, cost-effective, selective, reliable, and do not require any extraction steps or sample preparation, and offer detection limits at the sub-ppm level

concentrations simply and consecutively. The present method includes the determination of dopamine hydrochloride and methyldopa in water with high accuracy and precision enabling a successful application to determine drugs in pharmaceutical dosage. It is possible to apply the suggested method to evaluate many drugs and compounds that undergo oxidative processes using the orang G curve. Finally, the method is very inexpensive and has high sensitivity.

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