Journal of Engineering Science and Technology
Vol. 15, No. 3 (2020) 1824- 1839

SPECTROPHOTOMETRIC DETERMINATION OF METHYLDOPA WITH 2, 6-DIAMINOPYRIDINE REAGENT USING OXIDATIVE COUPLING REACTION

ISRAA TALIB HUMEIDY[1,*],
SARHAN ALI SALMAN[1], KADHIM K. HASHIM[2]

[1]Department of Chemistry, College of Science, Tikrit University, Tikrit, Iraq
[2] College of Environment, Al-Qasim Babylon Green University, Al-Qasim, Babylon, Iraq
*Corresponding Author: abudlayla@gmail.com

Abstract

The available techniques for methyldopa (MDP) measurement in both pure and solid pharmaceutical formulations are relatively slow, require pre-treatment procedures, and require expensive instruments. The current study investigates the applicability of a fast, affordable, and accurate spectrophotometric technique to determine methyldopa (MDP) concentration in both pure forms and solid pharmaceutical formulations. This method depends on the oxidative coupling reaction with 2, 6-diaminopyridine reagent in the presence of the oxidative agent of potassium periodate, in neutral medium, to yield an orange color product, which in turn is detectable by spectrophotometric methods. The orange color product is stable and soluble in water and it has the highest absorption at the wavelength of 478 nm. This work included the study of the oxidation time, quantity of an oxidant agent, quantity of the coupling reagent, effect of temperature and stoichiometry between MDP and 2,6- diaminopyridine and found to be 1:1, effect of interferences and calibration curve. The outcomes of this study indicate that the absorbance of the studied concentrations of MDP (2.0 to 24 $\mu g.ml^{-1}$) follows a linear equation with correlation coefficient of 0.99. Additionally, it has been found that the values of the molar absorptivity and Sandell's sensitivity index were 1.0624×104 L.mol-1.cm-1 and 0.0224 $\mu g.cm^{-2}$, respectively. While the value of relative standard deviation (RSD) was ranging between 1.340 and 1.793% depending on the studied concentration. These results confirm the applicability of this new technique to determine MDP concentration in both pure forms and solid pharmaceutical formulations.

Keywords: 2, 6-diaminopyridine, Methyldopa, Solid pharmaceutical formulations, Spectrophotometric.

1. Introduction

A number of previous studies indicated that the oxidative coupling reactions could be considered as an alternative to the cross-coupling reactions [1]. In the oxidative coupling reactions, two electrons will be emitted from a reactant and received by an oxidant. Although there is a wide body of literature focused on the mechanism of these reactions, the mechanism is only starting to emerge. Oxidative-coupling reactions are important organic reactions with wide applications in analytical chemistry as they are easy and highly sensitive methods. These reactions usually involve the association of two or more organic matter with the presence of an oxidizing agent where oxidation of these substances leads to the formation of effective intermediates that are chromogenic reagents [2], or fluorescent that interact with each other to form a spectrally measured product [3]. Though the oxidative coupling processes are usually classified according to both the catalyst and the oxidant [4], in the current study; they will be classified according to the oxidant used [1].

Methyldopa (MDP) is characterized as a white or colourless crystalline powder, which is used as a treatment for hypertension due to its ability to widen blood vessels [5]. Methyldopa is an agent reduce sympathetic outflow from vasomotor centres in the brain stem but allow these centres to retain or even increase their sensitivity to baroreceptor control. Methydopa is antihypertensive action appears to be due to stimulation of central α- adrenoceptors by α- methylnorepinephrine or α-methyldopamine. It lowers blood pressure chiefly by reducing peripheral vascular resistance, with variable reduction in heart rate and cardiac output [6]. Most cardiovascular reflexes remain intact after administration of methyldopa, and blood pressure reduction is not markedly dependent on posture. Postutal (orthostatic) hypotension sometimes occurs, particularly in volume-depleted patients. One potential advantage of methyldopa is that it causes reduction in renal vascular resistance [6]. This happens as a result of inhibition of the decarboxylation of dihydroxyphenylalanine (dopa), the precursor of norepinephrine, and of 5-hydroxytryptophan (5-HTP) in most peripheral tissues [7, 8]. However, these studies did not provide a rigid evidence about the mechanism of action. Various analytical methods were used to measure the concentration of MDP in pure and pharmaceutical preparations. For example, high performance liquid chromatography has been practiced to measure the concentration of MDP and its related substances [9].

Reversed-phase-HPLC (RP-HPLC) has also been practiced to instantaneously measurement of MDP concentration in pharmaceutical forms [10]. Some researchers applied the liquid chromatographic technique, aided by 4-dimethylamino- benzalaldehyde as a derivatizing reagent, to measure the concentration of MDP in pharmaceutical formulations [11]. Flow-injection spectrophotometric, which that depends on the production of open-chain o-benzoquinones by oxidizing of the targeted chemicals under the influence of the hexa-chloroiridate (IV) or permanganate ions, was also used to measure the concentrations of MDP. It is noteworthy to mention that this technique has been successfully used to determine catecholamines in the pure forms and in the pharmaceutical formulation [12]. Additionally, the performance of a new electrode, made from graphen (G) modified with 2, 7-bis (ferrocenyl ethyl) fluoren-9-one (2, 7-BFGPE), was evaluated by applying it to determine MDP in different solutions [13]. A new blue fluorescer technique was also applied to determine, indirectly, catecholamines in pharmaceutical formulations. The new blue fluorescer technique bases on chemiluminescence.

Reflectance spectroscopy is another technique that has been used to determine MDP in pharmaceutical products. The reflectance spectroscopy technique depends on the products of the reaction between MDP and molybdate ions, where the latter yields a yellow stable complex on the filter paper [14]. Spectrophotometric methods are depend on use of different reagents to estimate the concentration of MDP, including sodium nitrite in an acid environment, periodate sodium, 4-aminoacetophenone with sodium nitrite in acid medium, paraphenylenediamine with potassium periodate, and ortho-tolidine with potassium periodate in neutral environment [15-23]. Additionally, it includes ferric chloride and nitroso-R-salt (NRS), 2,6-dichloroquinone-4-chlorimide (DCQ), tetrachloro-p-benzoquinone (p-chloranil) and MDP, accelerated by hydrogen peroxide (H_2O_2), vanadium (V) which is reduced to vanadium (IV) and form complex with eriochrome cyanine (R) [15, 24-35]. A ready-made low-cost carbon paste electrode (CPE), made from graphite powder modified with nanostructured TiO_2 (TiO_2@CPE), has been used in the current study for the detection of methyldopa in pharmaceutical samples. It is noteworthy to mention that the TiO_2-modified graphite powder was developed and characterized by Kaushik, *et al.* [36].

According to the relevant literature, the available techniques for methyldopa (MDP) measurement in both pure and solid pharmaceutical formulations are relatively slow, require pre-treatment procedures, and require expensive instruments. Therefore, the current study investigates the applicability of fast, affordable, and accurate spectrophotometric technique to determine methyldopa (MDP) concentration in both pure forms and solid pharmaceutical formulations by oxidative coupling with 2,6-diaminopyridine reagent in the existence of the oxidative agent of potassium periodate.

2. Materials and Methods

2.1. Apparatus and analytical reagents

Spectral measurements were performed using the Shimadzu UV-Visible spectrophotometer (model: UV-160, Japan), Sartorius (model: BL210 S AG, Germany), magnetic stirrer (model: BIOSANMSH 300), and ultrasonic bath (model: 410 Lab tech, Korea). MDP containing pure forms and pharmaceutical formulations were obtained from Samarra Company (SDI) for pharmaceuticals and medical supplies, Iraq. All analytical reagents used in the current study were of high purity and used as supplied.

2.2. Preparation of the standard MDP solution

To prepare 1000 $\mu g.ml^{-1}$ MDP solution, 100 mg of methyldopa was dissolved in 100 ml of distilled water (DW) in a volumetric flask. 25 ml of this concentrated solution (1000 $\mu g.ml^{-1}$) was diluted with 100 ml with distilled water to get a concentration of 250 $\mu g.ml^{-1}$. The diluted solution was stored in a dark bottle to be used latter (within the next two week).

2.3. Solution of 2,6-diaminopyridine reagent

This solution was prepared by dissolving 0.1091g of 2,6-diaminopyridine reagent in suitable quantity of distilled water DW in a flask (100 ml in capacity), and then the solution was completed to the mark with DW to get a concentration of (1×10^{-2} M).

2.4. Solution of methyldopa tablets formulation

The solution of methyldopa tablets was prepared by grinding 10 Aldosam tablets (each one tablets contains 250 mg of MDP) to produce 4.9378 g of Aldosam tablets powder. A measured weight, 0.0395 g, of this powder was dissolved in measured volume of distilled water. Then, this solution was filtered using filter paper (604, RUNFILTER, Q240 mm). The filtrate was completed to 100 ml using DW to produce a solution with concentration of 250 $\mu g.ml^{-1}$.

3. Material and methods

3.1. Preliminary investigations

2.0 ml of 250 $\mu g.ml^{-1}$ of MDP solution was transferred into a 25 ml flask, then 2.0 ml of 1×10^{-2} M 2,6-diaminopyridine was added to this solution followed by addition of 1.0 ml of 5×10^{-2} M KIO_4 in neutral medium. The volume was completed to the required level with DW, to form an orange coloured product which can be measured at 478 nm against reagent blank [37, 38].

3.2. Calibration curve

Different volumes of 250 $\mu g.ml^{-1}$ MDP solution (ranging between 0.25 to 5.5 ml) were placed in different flasks (with net capacity of 25 ml). Then, 1.5 ml of KIO_4 (5×10^{-2} M) and 2.5 ml of 2,6-diaminopyridine reagent (1×10^{-2} M) were added to these flasks at temperature of 25°C. Then, these solutions were left, at room temperature, for 20 minutes to complete the reaction. The required volume of DW was added to each flask to complete the solution volume to the mark. The absorbance of the prepared solutions was then measured at 478 nm. The obtained calibration carve showed a linear trend for concentrations between 2.5 and 55 $\mu g.ml^{-1}$. While at concentrations more than 55 $\mu g.ml^{-1}$, the calibration curve showed a negative deviation from Beer's law. The value of the molar absorptivity was found to be 1.0481×10^{4} $L.mol^{-1}.cm^{-1}$, while the Sandell's sensitivity index was 0.0227 $\mu g.\ cm^{-2}$.

3.3. Stoichiometric ratio of formed product

The stoichiometry reaction between the MDP and 2,6-diaminopyridine have been studied spectrophotometrically by applying molar ratio and continuous variation methods [37]. In both methods, equal concentrations of MDP and 2,6-diaminopyridine (1×10^{-2} M) were used. In Job's method, different volumes of both drug solutions (1-9 ml) and reagent solutions (9-1 ml) were mixed in volumetric flasks with net capacity of 25 ml. Then, 1.5 ml of potassium periodate (5×10^{-2} M) was added to each flask; and then the volume was completed to the mark using DW. The absorptivity of the prepared solutions was measured at 478 nm [38]. In the molar ratio method, a measured volume of the standard drug solution (2 ml) was firstly placed in a volumetric flask (25 ml in capacity). Then, the required volumes of 2,6-diaminopyridine reagent solution (0.3 to 4.0 ml) and KIO_4 (1.5 ml) were added to the flasks. The volume of each solution was completed, by adding the required volume of DW, to the mark. The absorptivity of the prepared solutions was measured at 478 nm. The results indicated that the molar ratio was 1:1 that is highly agree with the results of Job's method [38].

4. Results and discussion

The results of the UV absorption tests show that MDP has weak absorption in the range of 250–300 nm (λmax is 280 nm in water and the molar absorptivity (ε) is 2789.4), which is not favorable as it decreases the accuracy of the spectrophotometric determination of MDP [17]. Thus, to enhance the light absorbing derivative, a suitable chromogen must be added to the MDP solution. The principle of the study is the MDP solution reacts with 2,6-diamino pyridine reagent in the presence of the oxidative agent of potassium periodate, in neutral medium, to form an orange coloured product that can be detected at 478 nm. The effects of key factors, such as contact time and volume of reagents, on the absorption were studied using 2 ml of MDP solution (250 μg.ml^{-1}) in 25 ml volumetric flask (20 μg.ml^{-1}). The absorbance was measured at 478 nm versus the blank reagent solution, Figs. 1 and 2 [39].

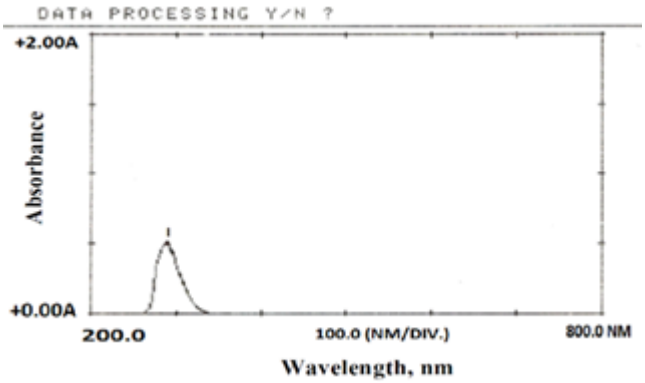


Fig. 1. Absorption spectrum of methyldopa at 280 nm before oxidation.

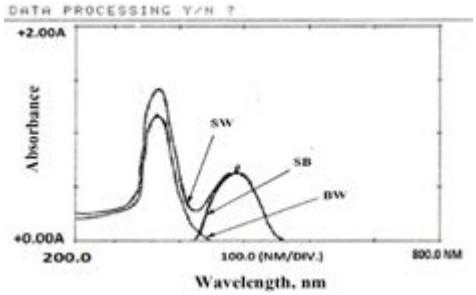


Fig. 2. Absorption spectra of coloured product against blank (SB), and distilled water (SW), and blank against distilled water (BW), where S: sample of coloured product, B: blank reagent, W: distilled water.

The time required to complete the oxidation process was determined as the maximum intensity of the coloured product that resulted from the reaction between MDP, potassium periodate, and 2,6-diaminopyridine. Experimentally, it has been noticed that the maximum intensity of the coloured product attained after 20 minutes, so 20 min is considered the optimal time for oxidation. The absorption level of the coloured product remains constant for 90 minutes at least, as shown in Fig. 3 [40].

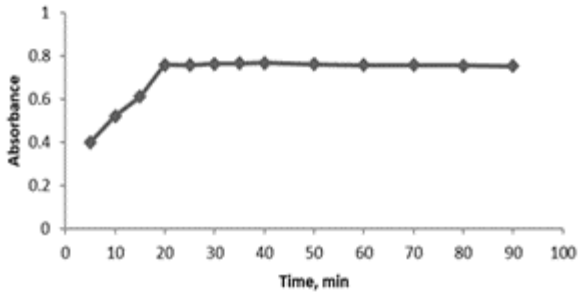


Fig. 3. Effect of oxidation time on the absorption of coloured product.

The influence of the quantity of the oxidant agent was examined by adding various volumes (0.3-2.5 ml) of the oxidant agent (KIO_4) to flasks (25 ml) containing 250 $\mu g.ml^{-1}$ of MDP and 1×10^{-2} M of 2,6-diaminopyridine reagent solution. Then, the solutions were left for 20 min for oxidation and the volumes were completed to the required level with DW. Figure 4 shows that 1.5 ml of KIO_4 gives the highest absorption value, therefore it was chosen in subsequent experiments [40].

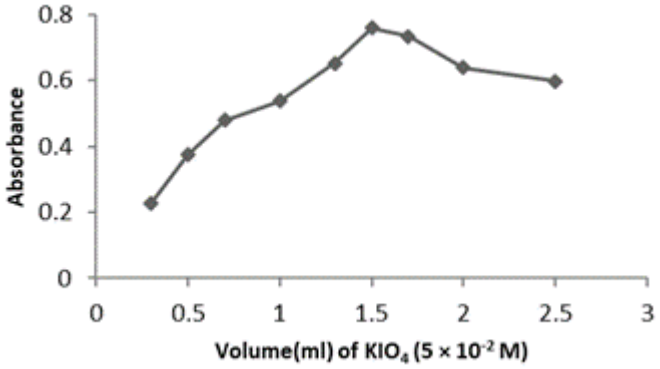


Fig. 4. Effect of an oxidant agent quantity on the absorbance.

The effect of different amounts of 2,6-diaminopyridine reagent on the colour intensity of the product were studied. From Fig. 5, it is observed that 2.5 ml of 2,6-diaminopridine reagent is the optimal quantity which gives higher absorption, therefore it was used in subsequent studies [39].

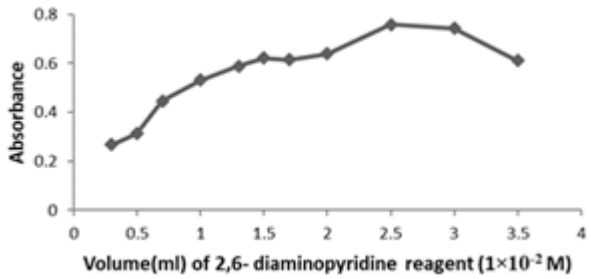


Fig. 5. Effect of 2,6-diaminopyridine reagent quantity.

The effect of various temperatures on MDP reaction with 2,6-diaminopyridine reagent was measured at different values (5 to 60°C) and neutral environment. It has been found that the absorption of the coloured product was remained stable at 25°C for more than 60 minutes, Fig. 6.

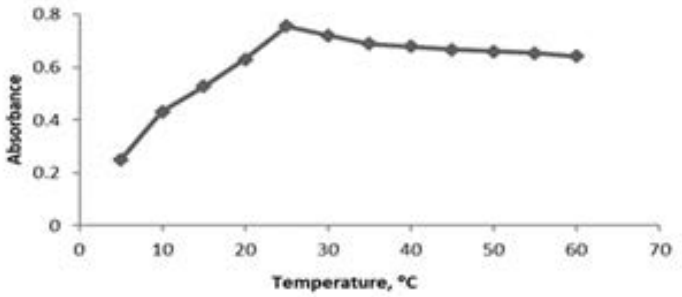


Fig. 6. Effect of temperature on the absorption of coloured product.

Using the optimal conditions from the previous experiments, the calibration curve was developed. The results of Fig. 7 show good linearity at ranges 2.5 to 50.0 µg.ml^{-1}. The obtained values of the molar absorptivity and Sandell's sensitivity index were 1.0481 x 10^4 L.mol.cm, and 2.27 x 10^{-2} µg.cm^{-2}. To ensure the accuracy of the tests, the absorption was measured, at 478 nm, for two different concentrations of the MDP (these concentrations were within the limits of Beer's law). The results indicated that the average recovery was 100.17 %, and the value of the relative standard deviation was ≤ 0.86%. These results confirm that this method has a very good accuracy.

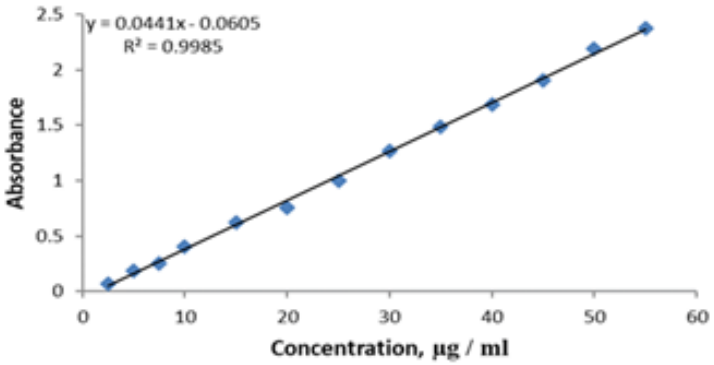


Fig. 7. Calibration curve for MDP- 2,6-diaminopyridine.

Job's and mole ratio methods have been applied to know the stoichiometry between MDP and 2,6- diaminopyridine. Figure 8 shows Job's methods (A) and mole ratio method (B) of formed product MDP- 2,6-diaminopyridine (the ratio of MDP: 2,6- diaminopyridine is 1:1).

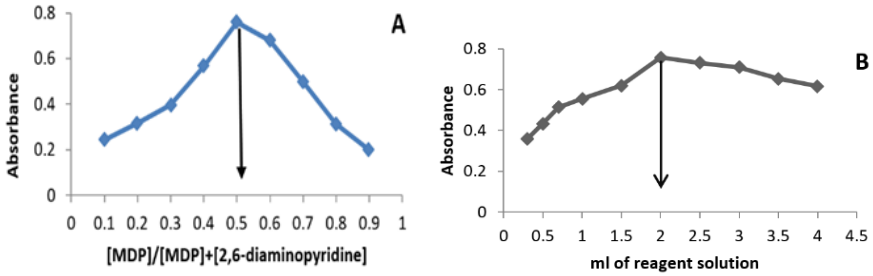
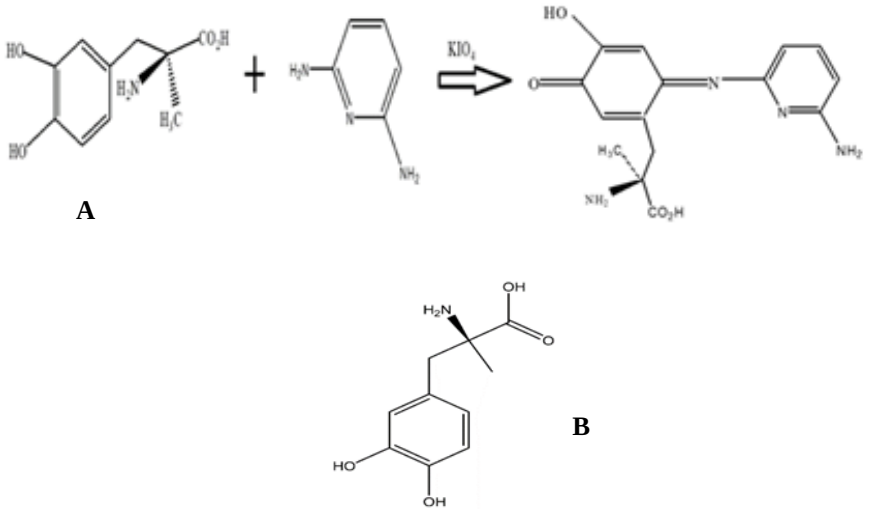


Fig. 8. Job's methods (A) and mole ratio method (B) of formed product MDP- 2,6-diaminopyridine.

Reaction of MDP with the 2,6 diaminopyridine reagent, in the presence of an oxidizing agent, leads to the formation of an oxidative coupling product, as described in the proposed question in Scheme 1.



Scheme 1. A)The proposed question of an oxidative coupling product [41], B) Chemical structure for Methyldopa [42].

To evaluate the efficiency and selectivity of this study, the influence of some foreign substances (sodium chloride, glucose, starch, maltose and talc), which are commonly found in solid pharmaceuticals, has been studied by adding different amounts (2.5, 5.0, 7.5) ml of foreign substances (1000 $\mu g.ml^{-1}$) to 20 $\mu g.ml^{-1}$ of MDP in a volumetric flasks (25 ml). The optimum conditions were applied and the amount has been completed to the mark with DW. The absorbance is measured at 478 nm versus blank and recovery was computed. The results appeared that the studied foreign substances did not interfere with the current method (Table 1) [39].

Table 1. Effect of interferences on 250 µg.ml^{-1} of MDP.

Foreign Compound	Recovery (%) of 20 µg.ml^{-1} of MDP per µg.ml^{-1} foreign compound added		
	100	**200**	**300**
Sodium chloride	100.65	99.78	100.45
Glucose	100.80	100.63	99.15
Starch	99.52	100.14	99.74
Maltose	100.03	99.49	98.31
Talc	99.87	100.25	100.50

The suggested method showed a successful performance, using the standard additions method (SAM), in terms of determination of methyldopa in aldosam tablets, Fig. 9 and Table 2.

Analysis results shown in the Table 2 confirm that standard additions method are well agreement with the current method for determining of methyldopa in the aldosam tablets and the current method is free of interferences [14].

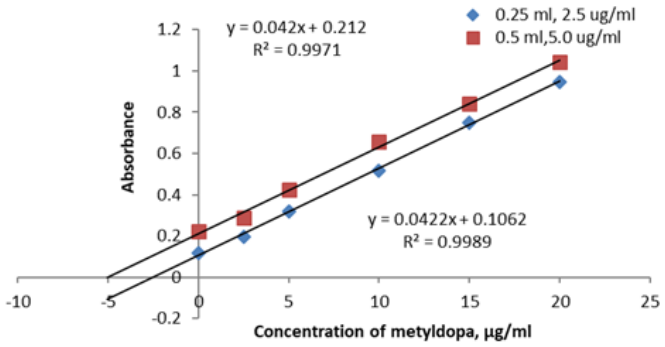


Fig. 9. Standard additions method of MDP in Aldosam tablets.

Table 2. Analysis results of Aldosam tablets by SAM.

Drug	Standard MDP concentration µg/ml	MDP measured µg/ml	RE%	Recovery, (%)
Aldosam	2.0	2.01	0.50	100.50
Tablets	4.0	3.99	-0.25	99.75

5. Residual MDP in medical wastewater

MDP has many useful medical applications, such as treatment for hypertension due to its ability to widen blood vessels. However, the residual MDP, like any other chemical elements, in the medical wastewater causes many environmental problems [43]. Therefore, different treatment methods have been practiced to remove MDP, and other medical pollutants, from medical wastewater before it discharged to the surrounding environment, such as nano-filtration units, chemical coagulation, and electrochemical methods [43-48]. In this study, the authors recommended the application of electrochemical units to remove MDP from wastewater as this method does not required chemical additives, it has the ability to achieve a simultaneous

removal of different pollutants, it has low operational cost, and it could be easily integrated with other treatment units [49-56]. Additionally, in comparison with other treatment units, electrochemical method generates less quantities of sludge that significantly enhances its cost-effectiveness because treatment and handling of solid wastes is an expensive process [57-59]. Additionally, according to the recent developments in the field of sensing and communication [60-63], it is recommended to use advanced sensing unit to monitor the concentration of MDP and other chemicals in the medical wastewater, which enables the operators of the treatment units to control the discharged concentration of such pollutants.

It is noteworthy to mention that difference between this study and previous studies is the use of 2,6-diaminopyridine as a new coupling reagent with the drug using an oxidizing agent and the formation of a coloured product absorbed at wavelength 478 nm. This reagent was not previously used as a coupling reagent with this drug. This method gave good results when applied to solid pharmaceuticals.

6. Conclusion

Some concluding observations from the study are listed below:

- The obtained results from the analysis process confirmed that the suggested method is easy, fast, appropriate and well-sensitivity for measurement of MDP in pure and pharmaceutical formulations.
- The outcomes of this study indicate that the absorbance of the studied concentrations of MDP (2 - 24 $\mu g.ml^{-1}$) follows Beer's law with correlation coefficient of 0.99, the molar absorptivity value and Sandell's sensitivity index were 1.0624×10^{4} $L.mol^{-1}.cm^{-1}$ and 0.0224 $\mu g.cm^{-2}$, respectively. Additionally, it has been found that the value of relative standard deviation (RSD) was ranging between 1.340 and 1.793% depending on the studied concentration.
- The current method meets all the main requirements of routine analysis because it is robust, does not include any pre-treatment of the sample and does not require certain working conditions (the use of organic solvents or temperature control) compared with most methylopa assay methods that require expensive instruments or require procedures with strict control in experimental conditions.
- This method is easy to perform as the final product is stable and soluble in water and gives a clear orange colour.

Nomenclatures

5-HTP	5-hydroxytryptophan
DCQ	2,6-dichloroquinone-4-chlorimide
p-chloranil	Tetrachloro-p-benzoquinone
R	Eriochrome cyanine
CPE	Carbon paste electrode
DW	Distilled water
G	Graphen
MDP	Methyldopa
NRS	Nitroso-R-salt
RP-HPLC	Reversed-phase-high-performance liquid chromatography
RSD	Relative standard deviation

References

1. Funes-Ardoiz, I.; and Maseras, F. (2018). Oxidative coupling mechanisms: current state of understanding. *ACS Catalysis*, 8(2), 1161-1172.
2. Gorumutchu, G.P.; Ratnakaram, V.N.; and Malladi, S. (2019). Determination of riociguat by oxidative coupling using visible spectrophotometry. *Oriental Journal of Chemistry*, 35(Special Issue 1), 48-53.
3. Temesi, J.; Rooney, K.; Raymond, J.; and o'connor, H. (2010). Effect of carbohydrate ingestion on exercise performance and carbohydrate metabolism in persons with spinal cord injury. *European journal of applied physiology*, 108(1), 131-138.
4. Hutchings, G.; Scurrell, M.; and Woodhouse, J. (1989). Oxidative coupling of methane using oxide catalysts. *Chemical Society Reviews*, 18(251-283.
5. Chobanian, A.V.; Bakris, G.L.; Black, H.R.; Cushman, W.C.; Green, L.A.; Izzo, J.L.; Jones, D.W.; Materson, B.J.; Oparil, S.; and Wright, J.T. (2003). Seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure. *Hypertension*, 42(6), 1206-1252.
6. Tveden‐Nyborg, P.; Bergmann, T.K.; and Lykkesfeldt, J. (2018). Basic & clinical pharmacology & toxicology policy for experimental and clinical studies. *Basic and Clinical Pharmacology and Toxicology*, 123(3), 233-235.
7. Modigh, K. (1973). Effects of chlorimipramine and protriptyline on the hyperactivity induced by 5-hydroxytryptophan after peripheral decarboxylase inhibition in mice. *Journal of Neural Transmission*, 34(2), 101-109.
8. Ren, L.Q.; Wienecke, J.; Hultborn, H.; and Zhang, M. (2016). Production of dopamine by aromatic L-amino acid decarboxylase cells after spinal cord injury. *Journal of Neurotrauma*, 33(12), 1150-1160.
9. Zheng, J.Q.; Jin, J.Z.; Chen, L.P.; and Li, H.L. (2010). HPLC determination of content of methyldopa and its related substances. *Chinese Journal of Pharmaceutical Analysis* , 30(8), 1440-1444.
10. Sahithi, M.L.; Perumal, V.; Sekhar, C.K.; and Babu, P.V. (2013). Development and validation of RP-HPLC method for simultaneous estimation of methyldopa and hydrochlorothiazide in pharmaceutical dosage form. *Journal of Advanced Pharmacy Education and Research*, 3(4), 464-470.
11. Majidano, A.A.; Sodho, N.A.; Rajper, G.N.; Alamgir, M.; and Khuhawar, M.Y. (2014). Liquid chromatographic determination of dopamine, methyldopa, L-dopa and tyrosine in pharmaceutical preparations using 4-dimethylamino-benzalaldehyde as a derivatizing reagent. *Sindh University Research Journal* 46(1), 77-82.
12. Shpigun, L.K.; and Ryabenko, V.S. (2012). Flowinjection method of spectrophotometric determination of catecholamines in pharmaceutical formulations. *Theoretical Foundations of Chemical Engineering*, 46(4), 419-423.
13. Beitollahi, H.; Tajik, S.; Asadi, M.H.; and Biparva, P. (2014). Application of a modified graphene nanosheet paste electrode for voltammetric determination of methyldopa in urine and pharmaceutical formulation. *Journal of Analytical Science and Technology*, 5(29), 2-9.

14. Ribeiro, P.S.; Pezza, L.; and Pezza, H.R. (2006). Determination of methyldopa in pharmaceutical formulations by combined spot test-diffuse reflectance spectroscopy. *Journal of the Brazilian Chemical Society*, 17(4), 674-679.
15. Ferreira Garcia, L.; Peixoto, C. E.; Kussmaul, E.; Vieira, D.; Sanz, G.; Luque, R.; Somerset, V.; and Souza, E. D. (2018). Nanostructured tio_2 carbon paste based sensor for determination of methyldopa. *Pharmaceuticals*, 11(4), 99-109.
16. Zubaidi, S.; Al-Bugharbee, H.; Ortega, M.S.; Gharghan, S.; Olier, I.; Hashim, K.; Al-Bdairi, N.; and Kot, P. (2020). A novel methodology for prediction urban water demand by wavelet denoising and adaptive neuro-fuzzy inference system approach. *Water*, 12(6), 1-17.
17. Al Samarrai, S.Y.; Abdoon, F.M.; and Hashim, K.K. (2019). A simple method to determine tramadol using a coated-wire electrode as a detector in the flow injection analysis. *Microchemical Journal*, 146, 588-591.
18. Zubaidi, S.L.; Ortega-Martorell, S.; Al-Bugharbee, H.; Olier, I.; Hashim, K.S.; Gharghan, S.K.; Kot, P.; and Al-Khaddar, R. (2020). Urban water demand prediction for a city that suffers from climate change and population growth: gauteng province case study. *Water*, 12(7), 1-18.
19. Al Sultani, K.H.; Al-Rashidy, A.M.; and Al-Samrrai, S.Y. (2019). Determination of tartrazine and sodium benzoate as food additives in some local juices using continuous flow injection analysis. *Engineering in Agriculture, Environment and Food*, 12(2), 217-221.
20. Hashim, K.K.; Yahyaa, S.Y.; and Al-Rashidy, A.M. (2018). Fluorescence quantum yield determination of propylparaben using flow injection spectroscopy. *Journal of Fluorescence*, 28(6), 1275-1280.
21. Zubaidi, S.L.; Ortega-Martorell, S.; Kot, P.; Alkhaddar, R.M.; Abdellatif, M.; Gharghan, S.K.; Ahmed, M.S.; and Hashim, K. (2020). A method for predicting long-term municipal water demands under climate change. *Water Resources Management*, 34(3), 1265-1279.
22. Hashim, K.; Kot, P.; Zubaid, S.; Alwash, R.; Al Khaddar, R.; Shaw, A.; Al-Jumeily, D.; and Aljefery, M. (2020). Energy efficient electrocoagulation using baffle-plates electrodes for efficient Escherichia Coli removal from Wastewater. *Journal of Water Process Engineering*, 33(20), 1-7.
23. Hashim, K.S.; Ali, S.M.; Alrifaie, J.K.; Kot, P.; Shaw, A.; Al Khaddar, R.; Idowu, I.; and Gkantou, M. (2020). Escherichia coli inactivation using a hybrid ultrasonic–electrocoagulation reactor. *Chemosphere*, 247, 1-7.
24. Hashim, K.S.; Alkhaddar, R.; Shaw, A.; Kot, P.; Al-Jumeily, D.; Alwash, R.; and Aljefery, M. H. (2020). *Electrocoagulation as an eco-friendly River water treatment method, in advances in water resources engineering and management* (1st ed.). Berlin: Springer.
25. Hashim, K.S.; Idowu, I.A.; Jasim, N.; Al Khaddar, R.; Shaw, A.; Phipps, D.; Kot, P.; Pedrola, M.O.; Alattabi, A.W.; and Abdulredha, M. (2018). Removal of phosphate from River water using a new baffle plates electrochemical reactor. *Methodsx*, 5, 1413-1418.
26. Hashim, K.S.; Al-Saati, N.H.; Hussein, A.H.; and Al-Saati, Z.N. (2018). An investigation into the level of heavy metals leaching from canal-dreged sediment: a case study metals leaching from dreged sediment. *Proceedings of*

the First International Conference on Materials Engineering & Science. Istanbul Aydın University , Turkey, 12-22.

27. Al-Saati, N.H.; Hussein, T.K.; Abbas, M.H.; Hashim, K.; Al-Saati, Z.N.; Kot, P.; Sadique, M.; Aljefery, M.H.; and Carnacina, I. (2019). Statistical modelling of turbidity removal applied to non-toxic natural coagulants in water treatment: a case study. *Desalination and Water Treatment*, 150, 406-412.
28. Gotardo, M.; Lima, L.; Sequinel, R.; Rufino, J.; Pezza, L.; and Pezza, H.R. (2008). A simple spectrophotometric method for the determination of methyldopa using p-chloranil in the presence of hydrogen peroxide. *Eclética Química*, 33(3), 7-12.
29. Abdulhadi, B.A.; Kot, P.; Hashim, K.S.; Shaw, A.; and Khaddar, R.A. 2019). Influence of current density and electrodes spacing on reactive red 120 dye removal from dyed water using electrocoagulation/electroflotation (EC/EF) process. *Proceedings of the First International Conference on Civil and Environmental Engineering Technologies*. University of Kufa, Iraq, 12-22.
30. Zubaidi, S.L.; Al-Bugharbee, H.; Muhsen, Y.R.; Hashim, K.; Alkhaddar, R. M.; Al-Jumeily, D.; and Aljaaf, A.J. (2019). The prediction of municipal water demand in Iraq: a case study of Baghdad governorate. *Proceedings of the 12th International Conference on Developments in esystems Engineering*. Kazan, Russia, 274-277.
31. Zubaidi, S.L.; Kot, P.; Hashim, K.; Alkhaddar, R.; Abdellatif, M.; and Muhsin, Y.R. (2019). Using LARS –WG model for prediction of temperature in Columbia City, USA. *Proceedings of the First International Conference on Civil and Environmental Engineering Technologies*. University of Kufa, Iraq, 31-38.
32. Hashim, K.S.; Hussein, A.H.; Zubaidi, S.L.; Kot, P.; Kraidi, L.; Alkhaddar, R.; Shaw, A.; and Alwash, R. (2019). Effect of initial ph value on the removal of reactive black dye from water by electrocoagulation (EC) method. *Proceedings of the 2nd International Scientific Conference*. Al-Qadisiyah University, Iraq 12-22.
33. Hashim, K.S.; Al-Saati, N.H.; Alquzweeni, S.S.; Zubaidi, S.L.; Kot, P.; Kraidi, L.; Hussein, A.H.; Alkhaddar, R.; Shaw, A.; and Alwash, R. (2019). Decolourization of dye solutions by electrocoagulation: an investigation of the effect of operational parameters. *Proceedings of the First International Conference on Civil and Environmental Engineering Technologies*. University of Kufa, Iraq, 25-32.
34. Omran, I.I.; Al-Saati, N.H.; Hashim, K.S.; Al-Saati, Z.N.; P., K.; Khaddar, R. A.; Al-Jumeily, D.; Shaw, A.; Ruddock, F.; and Aljefery, M. (2019). Assessment of heavy metal pollution in the Great Al-Mussaib irrigation channel. *Desalination and Water Treatment*, 168, 165-174.
35. Emamjomeh, M.M.; Mousazadeh, M.; Mokhtari, N.; Jamali, H.A.; Makkiabadi, M.; Naghdali, Z.; Hashim, K.S.; and Ghanbari, R. (2019). Simultaneous removal of phenol and linear alkylbenzene sulfonate from automotive service station wastewater: Optimization of coupled electrochemical and physical processes. *Separation Science and Technology*, 1, 1-11.
36. Kaushik, R.D.; Yadav, R.; Sushma; Manilz; and Singh, J. (2014). Development and validation of kinetic-spectrophotometric method for

determination of methyl dopa in aqueous formulations and tablets *Der Pharma Chemica*, 6(4), 102-108.

37. Al-Abachi, M.Q.; Mohmmed, S.S.; and Hadi, A.J. (2015). The spectrophotometric determination of Famotidine drug via coupling with diazotized metochlopramide hydrochloride. *Baghdad Science Journal*, 12(4), 730-739.
38. Mabrouk, M.; Hammad, S.F.; Abdelaziz, M.A.; and Mansour, F.R. (2018). Ligand exchange method for determination of mole ratios of relatively weak metal complexes: a comparative study. *Chemistry Central Journal*, 12(1), 143.
39. Madrakian, T.; Afkhami, A.; Khalafi, L.; and Mohammadnejad, M. (2006). Spectrophotometric determination of catecholamines based on their oxidation reaction followed by coupling with 4-aminobenzoic acid. *Journal of the Brazilian Chemical Society*, 17(7), 1259-1265.
40. Mohammed, I.S.; Nasser, K.A.; and Mhemeed, A.H. (2017). Spectrophotometric determination of bisacodyl in pure and pharmaceutical preparation via oxidative coupling organic reaction. *Baghdad Science Journal*, 14(1), 181-188.
41. Fazakerley, G.V.; Linder, P.W.; Torrington, R.G.; and Wright, M.W. (1979). Potentiometric and spectrophotometric studies of the copper (II) complexes of methyldopa, methyltyrosine, and catechol in aqueous solution. *Journal of the Chemical Society, Dalton Transactions*, 12, 1872-1880.
42. Shahabadi, N.; and Maghsudi, M. (2014). Multi-spectroscopic and molecular modeling studies on the interaction of antihypertensive drug; methyldopa with calf thymus DNA. *Molecular biosystems*, 10(2), 338-347.
43. Hlongwane, G.N.; Sekoai, P.T.; Meyyappan, M.; and Moothi, K. (2018). Simultaneous removal of pollutants from water using nanoparticles: A shift from single pollutant control to multiple pollutant control. *Science of The Total Environment*, 656, 808-833.
44. Alattabi, A.W.; Harris, C.; Alkhaddar, R.; Alzeyadi, A.; and Hashim, K. (2017). Treatment of residential complexes' wastewater using environmentally friendly technology. *Procedia Engineering*, 196, 792-799.
45. Alattabi, A.W.; Harris, C.B.; Alkhaddar, R.M.; Hashim, K.S.; Ortoneda-Pedrola, M.; and Phipps, D. (2017). Improving sludge settleability by introducing an innovative, two-stage settling sequencing batch reactor. *Journal of Water Process Engineering*, 20, 207-216.
46. Hashim, K.S. (2017). *The innovative use of electrocoagulation-microwave techniques for the removal of pollutants from water* Ph.D. Thesis. Civil Engineering Liverpool John Moores University, Liverpool, United Kingdom.
47. Hashim, K.S.; Khaddar, R.A.; Jasim, N.; Shaw, A.; Phipps, D.; Kota, P.; Pedrola, M.O.; Alattabi, A.W.; Abdulredha, M.; and Alawsh, R. (2018). Electrocoagulation as a green technology for phosphate removal from River water. *Separation and Purification Technology*, 210, 135-144.
48. Gökkuş, Ö.; and Yıldız, Y.Ş. (2016). Application of electro-Fenton process for medical waste sterilization plant wastewater. *Desalination and Water Treatment*, 57(52), 24934-24945.
49. Safari, S.; Azadi Aghdam, M.; and Kariminia, H.R. (2015). Electrocoagulation for COD and diesel removal from oily wastewater. *International Journal of Environmental Science and Technology*, 13(1), 231-242.

50. Hashim, K.S.; Shaw, A.; Al Khaddar, R.; Ortoneda Pedrola, M.; and Phipps, D. (2017). Defluoridation of drinking water using a new flow column-electrocoagulation reactor (FCER) - Experimental, statistical, and economic approach. *Journal of Environmental Management*, 197, 80-88.
51. Hashim, K.S.; Shaw, A.; Al Khaddar, R.; Pedrola, M.O.; and Phipps, D. (2017). Iron removal, energy consumption and operating cost of electrocoagulation of drinking water using a new flow column reactor. *Journal of Environmental Management*, 189, 98-108.
52. Hashim, K.S.; Shaw, A.; Al Khaddar, R.; Pedrola, M.O.; and Phipps, D. (2017). Energy efficient electrocoagulation using a new flow column reactor to remove nitrate from drinking water - Experimental, statistical, and economic approach. *Journal of Environmental Management*, 196, 224-233.
53. Shubbar, A.A.; Jafer, H.; Dulaimi, A.; Hashim, K.; Atherton, W.; and Sadique, M. (2018). The development of a low carbon binder produced from the ternary blending of cement, ground granulated blast furnace slag and high calcium fly ash: An experimental and statistical approach. *Construction and Building Materials*, 187, 1051-1060.
54. Al-Jumeily, D.; Hashim, K.; Alkaddar, R.; Al-Tufaily, M.; and Lunn, J. (2019). Sustainable and environmental friendly ancient reed houses (inspired by the past to motivate the future). *Proceedings of the 11th International Conference on Developments in esystems Engineering*. Cambridge, UK, 214-219.
55. Shubbar, A.A.; Sadique, M.; Shanbara, H.K.; and Hashim, K. (2020). *The development of a new low carbon binder for construction as an alternative to cement, in advances in sustainable construction materials and geotechnical engineering* (1st ed.). Berlin: Springer.
56. Shubbar, A.A.; Al-Shaer, A.; Alkizwini, R.S.; Hashim, K.; Hawesah, H.A.; and Sadique, M. (2019). Investigating the influence of cement replacement by high volume of GGBS and PFA on the mechanical performance of cement mortar. *Proceedings of the First International Conference on Civil and Environmental Engineering Technologies*. University of Kufa, Iraq, 31-38.
57. Abdulredha, M.; Al Khaddar, R.; Jordan, D.; Kot, P.; Abdulridha, A.; and Hashim, K. (2018). Estimating solid waste generation by hospitality industry during major festivals: A quantification model based on multiple regression. *Waste Management*, 77, 388-400.
58. Abdulredha, M.; Rafid, A.; Jordan, D.; and Hashim, K. (2017). The development of a waste management system in Kerbala during major pilgrimage events: determination of solid waste composition. *Procedia Engineering*, 196, 779-784.
59. Idowu, I.A.; Atherton, W.; Hashim, K.; Kot, P.; Alkhaddar, R.; Alo, B.I.; and Shaw, A. (2019). An analyses of the status of landfill classification systems in developing countries: Sub Saharan Africa landfill experiences. *Waste Management*, 87, 761-771.
60. Gkantou, M.; Muradov, M.; Kamaris, G.S.; Hashim, K.; Atherton, W.; and Kot, P. (2019). Novel electromagnetic sensors embedded in reinforced concrete beams for crack detection. *Sensors*, 19(23), 5175-5189.
61. Ryecroft, S.; Shaw, A.; Fergus, P.; Kot, P.; Hashim, K.; Moody, A.; and Conway, L. (2019). A first implementation of underwater communications in

raw water using the 433 MHz frequency combined with a bowtie antenna. *Sensors*, 19(8), 1813.

62. Teng, K.H.; Kot, P.; Muradov, M.; Shaw, A.; Hashim, K.; Gkantou, M.; and Al-Shamma'a, A. (2019). embedded smart antenna for non-destructive testing and evaluation (ndt&e) of moisture content and deterioration in concrete. *Sensors*, 19(3), 547.
63. Ryecroft, S.P.; Shaw, A.; Fergus, P.; Kot, P.; Hashim, K.; and Conway, L. 2020). A novel gesomin detection method based on microwave spectroscopy. *Proceedings of the 12th International Conference on Developments in eSystems Engineering*. Kazan, Russia, 14-23.