Spectrophotometric determination of metformin hydrochloride via oxidative coupling reaction with 1-naphthol in pharmaceutical and environmental water sample

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Abstract

A simple, rapid, accurate and sensitive spectrophotometric method for determination of metformin hydrochloride has been developed. The proposed method is based on the oxidation of 1-naphthol by sodium hypochlorite and coupling with metformin hydrochloride in the presence of sodium hydroxide to form an intense blue soluble product with maximum absorption at 580 nm. Beer's law is obeyed over the concentration range of 2-20µg/ml, with molar absorptivity of $3.66x10^4$ l/mol.cm.The present method is considered to be simple because it does not need either heating or hydrolysis or solvent extraction steps. The ingredients often formulated with metformin hydrochloride have been shown not to interfere, and the proposed method is suitable for the routine determination of metformin hydrochloride. The method has been successfully applied for the determination of metformin hydrochloride in pure form, pharmaceutical preparations(Tablets) and environmental water sample .

Keywords: Metformin hydrochlorid, spectrophotometry, pharmaceutical preparations

الخلاصة

تم وصف طريقة طيفية سهلة وسريعة وحساسة لتقدير المتفورمين هيدروكلورايد . تعتمد الطريقة على اكسدة 1 – نافثول بواسطة هايبوكلورات الصوديوم واقترانة مع المتفورمين هيدروكلورايد بوجود هيدروكسيد الصوديوم لتكوين ناتج ازرق اللون ذائب في الماء وله اقصى امتصاص عند 580 نانو ميتر.لقد وجد بان قانون بير يسري على الكميات التي تتراوح بين2–20 مايكروغرام\ مل بامتصاصية مولارية مقدارها 3.66×10^4 لترامول.سم . تعد الطريقة الحالية بسيطة كونها لاتحتاج الى تسخين او تحلل مائي او استخلاص مذيبي حيث ان المواد الداخلة في تحضير المستحضرات المحللة لاتتداخل مع المتفورمين هيدروكلورايد مما جعلها طريقة ناجحة للتحليل الروتيني للمتفورمين هيدروكلورايد بشكله النقي وفي مستحضراته الصيدلانية (الحبوب)وفي نموذج بيئي من المياه

Introduction

Metformin hydrochloride (glucophage) [1] , chemically is 1,1-Dimethyl biguanide hydrochloride with a molecular formula of $C_4H_{12}Cl N_5$ (Fig 1). It is an oral antidiabetic drug that has been used in the treatment of non- insulin dependent diabetes which improves control of glycemia primarily by inhibiting hepatic gluconeogenesis and glucogenolysis[2] and seems to ameliorate hyperglycemia by improving preipheral sensitivity to insulin , reducing gastrointestinal glucose absorption and hepatic glucose production . Recently, metformin has also become available for the treatment of polycystic ovary syndrom and has been found to improve vascular function, prevent pancreatic cancer and revers fatty liver diseases[3]

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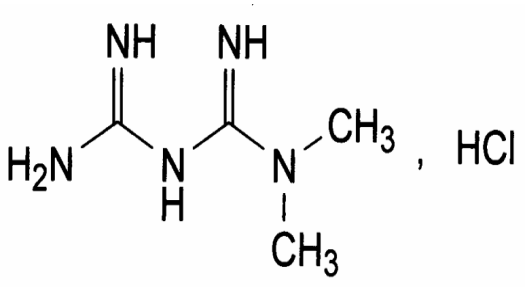


M. Wt = 165.6

Fig (1) : Chemical structure of metformin- HCl

Literature survey reveals that many HPLC methods for the detremination of metformin are reported . But most of the methods use either ion-pair reagent or cation exchange column[4-15]. Another different methods for the determination of metformin have been described , such as conductometric titration[16], flow-injection chemiluminescence[17-19], capillary electrophoresis[20], ion-selective electrode[21] and adsorptive catalytic squar-wave voltammetry[22].Very few spectrophotometric methods for the determination of metformin hydrochloride, in pharmaceutical formulation are described. The official method includes uv spectrophotometric method for estimation of the drug in the tablets[23]. The colorimetric methods include charge transfer complex with iodine in acetonitrile medium[24], reaction of metformin with Cu^{+2} in basic cyclohexylamine medium[25] and the reaction with ninhydrin to form a violet colored complex[26]. And spectrophotometric method using multi variate However , all of these technique[27]. methods suffered from several disadvantages including use of complex extraction procedures which were tedious and time consuming , ultra filtration and column-switching technique, have been suggested to improve specificity and selectivity . The proposed method can be applicable to routine analysis and content uniformity test of metformin hydrochloride in tablets and complies well with the validation requirments in the pharmaceutical industry[28].

Material and Methods

Apparatus

A Spectro scan 50 Uv visible spectrophotometer with 1.0 cm quartz cells was used .

Reagents

All chemicals used were of analytical grade and the metformin hydrochloride standard material was provided from state company of drug industries and medical appliance (NDI) Ninavah – Iraq. .

Metformin hydrochloride stock solution (1000 ppm) was prepared by dissolving 0.1g of metformin hydrochloride in 100ml distilled water in a volumetric flask .

Metformin hydrochloride standard solution (100 ppm) was prepared by diluting 10 ml of stock solution to 100 ml by distilled water in a volumetric flask.

Sodium hypochlorite solution (0.1%) was prepared by dilution 1.25 ml of 8% sodium hypochlorite to 100 ml by distilled water, this solution was standardized every 4-5 days and stored in a dark bottle[29].

Sodium hydroxide solution (0.1N)

1-Naphthol solution(0.1%) was prepared by dissolving 0.1g of 1-naphthol in 100ml ethanol 95% (in a (volumetric flask .

Recommended procedure

Aliquots of standard solution of metformin hydrochloride (50-500µg) were transferred into a series of 25 ml calibrated flasks, added

1 ml of 0.1% sodium hypochlorite solution ,3 ml of 0.1% 1-naphthol,and 1 ml of 0.1 N sodium hydroxide , dilute the solution to the mark with distilled water . The absorbance of the blue-colored product was measured at 580 nm against a reagent blank.

Procedure for pharmaceutical preparations (tablets)

Weigh and powder 10 tablets . Dissolve a quantity of the powdered tablets equivalent to 100mg of pure metformin hydrochloride in about 100 ml distilled water and mix for 20 min and then filtered. The filtrate was made up to 1L with distilled water . Treat 3 ml of this solution as mentioned under recommended procedure.

Procedure for tap water

To demonstrate the practical applicability of the proposed method, tap water sample from, Mosul-Iraq, were filtered and analyzed for metformin hydrochloride . Which shows negative results, the samples were spiked with the concentrations ranging from 5-15 $\mu g.ml^{-1}$ of metformin hydrochloride .The spiked water samples were analyzed as described above for recommended procedure and the concentration was calculated by using the calibration curve of the method.

Results and Discussion

The absorption spectra of the reagent and its metformin complex are shown in Fig.2. The maximum absorbance of the blue product is at 580nm which used in all subsequent experiments.

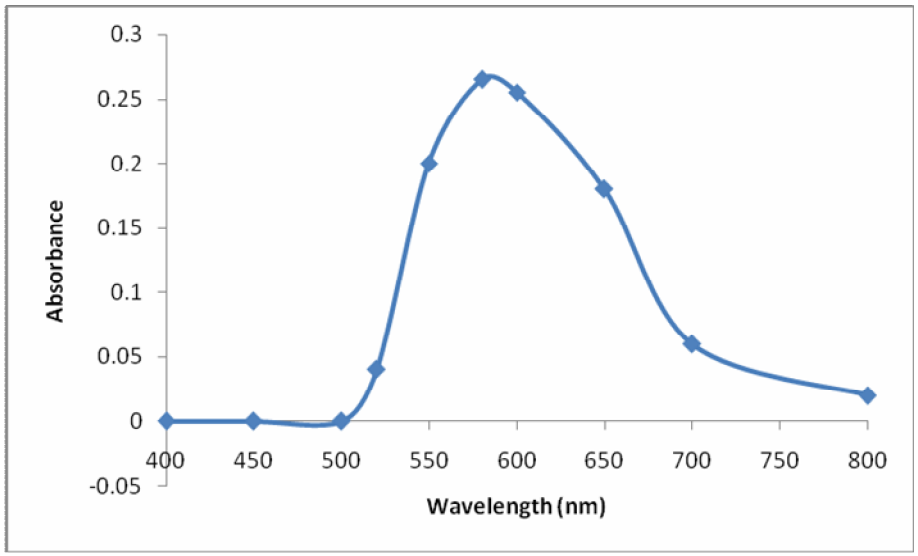


Fig 2 :Absorption spectra of 12µg/ml of metformin .HCL with 1-naphthol against reagent blank.

Effect of base

It was found that the presence of a base led to increase the intensity and cloudless of the the produced product, therefore some bases such as NaOH,Na_2CO_3 and NH_4OH are examined and was found that all these bases gave almost equal intensity, NaOH was selected and it was found that 1ml of this base gave ahigh sensitivity and was selected in all subsequent experiments.

Effect of reagent concentration

The amount of 1- naphthol solution for maximum color intensity was examined. 1 ml was found enough to develop the color to its full intensity and was selected in all subsequent experiments.

Effect of oxidant concentration

The amount of sodium hypochlorite solution for maximal color intensity was examined . 1 ml was found enough to develop the color to its full intensity which was selected in all subsequent experiments.

Effect of reaction time

Immediate color was developed at room temperature and the absorbance was stable for at least 6 hours.

Effect of order of addition

To obtain optimum results the order of addition of reagent should be followed as given under the recommended procedure , otherwise a loss in color intensity was observed.

Beer's law

Employing the condition described under recommended procedure , alinear calibration curve for metformin hydrochloride with the concentration range of 2-20µg/ml Fig 3 . Linear regression equation : y= 0.022x - 0.002 (r=0.999, n=6) where y is the absorbance and x is the concentration in µg/ml. The conditional molar absorptivity was found to be $3.66x10^4$ l/mol.cm ,with accuracy (average recovery%) was99.8 and the relative standard deviation of better than 1%was obtained.

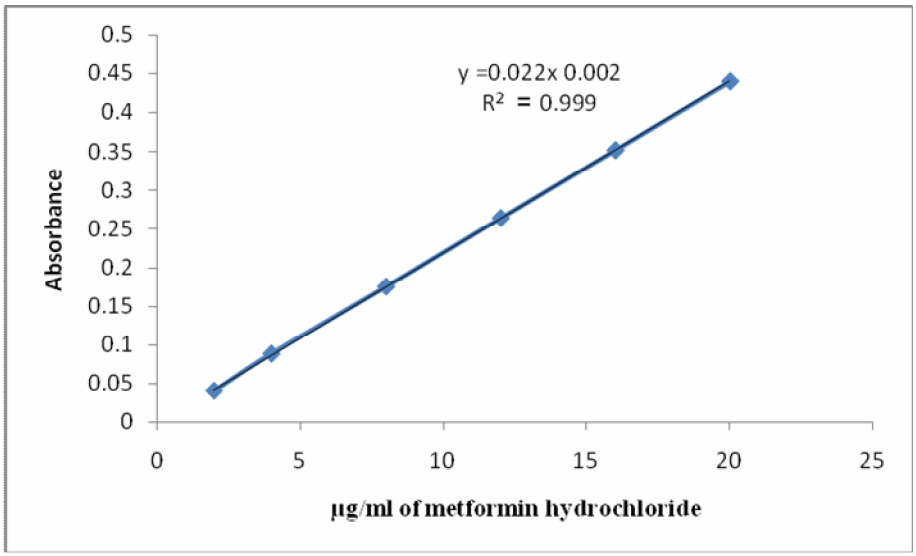


Fig 3 : Calibration graph of metformin hydrochloride.

Interference studies

In order to assess the possible applications of the proposed method, the effect of substance that often present with metformin hydrochloride in (Tablets) were studied by adding various amounts of excipients to 10 µg of metformin hydrochloride . An attractive feature of the method is its relative freedom from interference by the usual diluents and excipients in amounts for in excess of their normal occurence in pharmaceutical preparations. The results are given in Table (1).

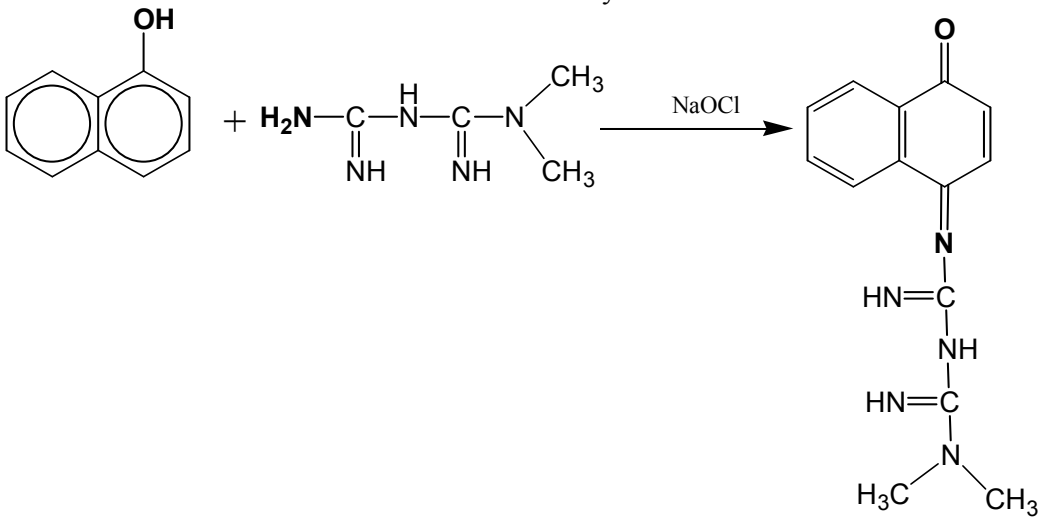
Table1: Determination of 10 µg / ml of metformin hydrochloride in the presence of excipients and other substances.

Interfering substances	Amount added/mg of interfering	Amount of drug found*,µg	Recovery, %
Lactose	40	10.06	100.6
Microcrystalline cellulose	20	9.96	99.6
Corn starch	30	9.97	99.7
Povidone	30	10.05	100.5
Magnesium stearate	40	10.07	100.7
Hydroxyl propyl methyl cellulose	40	10.07	100.7
Polyethylene glycol	20	10.04	100.4
Titanium dioxide	10	10.02	100.2

*Average of six determinations.

Composition of the colored product

The stiochiometry of reactants was investigated by the mole- ratio method [30]. The results obtained indicated that the existence of 1:1 metformin hydrochloride- 1-naphthol at 580 nm. Thus the suggested reaction may be written as



Blue color

Apparent stability constant of the product

The conditional stability constant of the product was estimated by using the following equation[31]:

$$K=a-(\Delta A/\varepsilon)\ /n^{n}(\Delta A/\varepsilon)$$

where :
a = metformin total concentration.
ΔA= sample absorbance in reagent excess minus the sample absorbance in stiochiometric reagent amount.
ε = molar absorptivity at the measured wavelength.
and n = number of ligand.

The stability constant (mean of five values) is found to be 1.31×10^{6} $l.mol^{-1}$, indicating that the product is stable.

Analytical application:

The proposed method was satisfactorily applied to the determination of metformin hydrochloride in its pharmaceutical preparations(tablets) and tap water samples ,the results of the assay of the pharmaceutical preparations revels that there is close agreement between the results obtained by the proposed method and the lable claim (Table 2), and the results of water samples (Table3) show that the recovery values obtained were closed to 100%.

Table 2 : Determination ofmetformin hydrochloride in pharmaceutical formulations

Pharmaceutical formulations	Lable amount(mg)	Found by proposed method(mg)*	Recovery%
Glucosam tablets(NDI-Iraq)	500mg/tab	498.75	99.75
	850mg/tab	852.125	100.25
METFORMAL (SPA-Italy)	500 mg/tab	499	99.8

*mean value of ten determinations

Table 3 : Determination of metformin hydrochloride in tap water samples

Wastewater samples	**Tadalafil added µg/ml**	**Found* µg/ml**	**Recovery %(n=10)**
Tap water	4	4.04	101
	12	11.94	99.5
	20	20.26	101.3

* mean value of ten determinations

Conclusions

The developed method is found to be sensitive ,accurate ,simple ,precise and economical ,and can be used for routine quality control analysis of metformin hydrochloride in pure form ,bulk, pharmaceutical formulations and environmental water samples.

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