International Journal of Scientific Research and Engineering Development-– Volume 7 Issue 4, July-Aug 2024
Available at www.ijsred.com

RESEARCH ARTICLE OPEN ACCESS

A REVIEW ON VARIOUS ANALYTICAL METHODS FOR ANALYSIS OF METFORMIN HYDROCHLORIDE

J. Lavanya[1*] B. Uma Reddy[2], T. Ramarao[3]
[1]Associate Professor, Department of Pharmaceutical Analysis, CMR college of pharmacy, Medchal, Hyderabad, India.
[2]Scholar, Department of Pharmaceutical Analysis, CMR college of Pharmacy, Medchal, Hyderabad, India.
[3]principal of CMR college of Pharmacy, Medchal, Hyderabad, India.
*Author for Correspondence:J.Lavanya, M.Pharm, Ph.D
Email: umareddybalemla@gmail.com

ABSTRACT:
Metformin is an oral Anti-Diabetic medicine in helping complications of type 2 diabetes and it is a good first- line remedy for an over-obese with type 2 diabetes, it's presently available in further than 60 countries worldwide. As a result of the significance of this oral hypoglycemic agent in the treatment of non-insulin-dependent diabetes mellitus, which leads to end- stage renal complaint, this work aims to collect the published logical styles reported so far in the literature for the determination in natural samples and pharmaceutical phrasings. This composition narrates different ways like high performance liquid chromatography. It can be seen that high- performance liquid chromatography styles have been used considerably. therefore, this paper will help in the selection and development of proper logical methodologies estimation of Metformin to achieve satisfactory results.

KEY WORDS: Metformin, HPLC, UV spectroscopy, Anti-Diabetic

INTRODUCTION:

Metformin (dimethylbiguanide) has become the preferred first-line oral blood glucose-lowering agent to manage type 2 diabetes. Its history is linked to Galega officinalis (also known as goat's rue), a traditional herbal medicine in Europe, found to be rich in guanidine, which, in 1918, was shown to lower blood glucose. Guanidine derivatives, including metformin, were synthesised and some (not metformin) were used to treat diabetes in the 1920s and 1930s but were discontinued due to toxicity and the increased availability of insulin. Metformin was rediscovered in the search for antimalarial agents in the 1940s and, during clinical tests, proved useful to treat influenza when it sometimes lowered blood glucose. This property was pursued by the French physician Jean Sterne, who first reported the use of metformin to treat diabetes in 1957. However, metformin received limited attention as it was less potent than other glucose-lowering biguanides (phenformin and buformin), which were generally discontinued in the late 1970s due to high risk of lactic acidosis. Metformin's future was precarious, its reputation tarnished by association with other biguanides despite evident differences. The ability of metformin to counter insulin resistance and address adult-onset hyperglycaemia without weight gain or increased risk of hypoglycaemia gradually gathered credence in Europe, and after intensive scrutiny metformin was introduced into the USA in 1995. Long-term cardiovascular benefits of metformin were identified by the UK Prospective Diabetes Study (UKPDS) in 1998, providing a new rationale to adopt metformin as initial therapy to manage hyperglycaemia in type 2 diabetes. Sixty years after its introduction in diabetes treatment, metformin has become the most prescribed glucose-lowering medicine worldwide with the potential for further therapeutic applications. [1]. Metformin is the first-line anti diabetic agent, which is used for (A

Review on Various Analytical Methods for Analysis of Metformin Hydrochloride)type 2 diabetes treatment, particularly in overweight patients where diabetes not controlled by adequate diet.[2]

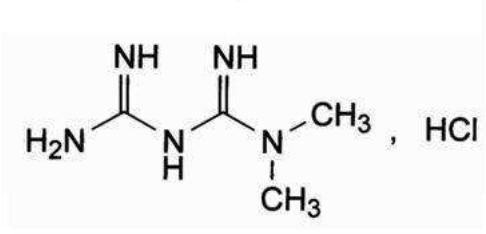

Fig. 1. Structure of Metformin Hydrochloride

Physical And Chemical Properties

- Metformin is white and almost white crystalline powder
- IUPAC name 3-(diamino methylidene)-1,1- dimethylgunidine,
- Molecular formula $C_4H_{11}N_5$
- Molecular weight 129.197g/mol,
- Melting point of 223-226 ºC,
- It has water solubility freely soluble as HCl salt, stable under recommended storage conditions,hazardous decomposition products formed under fire conditions.[3]

Pharmacokinetic of Metformin:

The optimal oral metformin dose for many diabetic patients is 2g/day. After a single oral dose, metformin is rapidly distributed to many tissues following partial absorption by the small intestine, but the luminal concentration in the gastrointestinal tract remains high. The peak plasma concentration occurs in 3 hr (increasing from 1.0 to 1.6 mg/ml [about 6 to 10 mM] after a 0.5 g dose and to 3 mg/ml [about 18 mM] after a 1.5 g dose) with a mean plasma half-life of about 20 hr. When the human metformin dose of 20 mg/kg/day orally is translated to the mouse equivalent dose of 250 mg/kg/day, according to the normalization to body surface area, murine plasma levels of metformin of up to 1.7 mg/ml (about 10 mM) are achieved. This is in the range achieved when conventional antidiabetic doses are used in humans. Biodistribution studies in mice using 14C-labeled metformin showed accumulation mainly in the gastrointestinal tract, kidney, and liver. It is important to note that being supplied directly by blood coming from the portal vein, the liver may contain a concentration of orally administered metformin substantially higher than in the general circulation and other organs. Metformin liver concentrations of greater than 180 mmol/kg wet weight and 250 mmol/kg wet weight in normal and diabetic rodents, respectively, can be achieved after a single dose of 50 mg/kg.[4]

UV- SPECTROPHOTOMETRIC METHOD:

A Sharma et al, 2023: was developed and validated for the simultaneous estimation of Metformin Hydrochloride (MH) and Pravastatin Sodium (PS) in pure form. Simultaneous estimation of Metformin Hydrochloride (MH) and Pravastatin Sodium (PS) was estimated by ultraviolet (UV) spectrophotometry using the absorbance subtraction method. The method was based on the measurement of absorbance at two wavelengths 232 nm. and 238 nm, of Metformin and Pravastatin Sodium respectively. These studies were performed at three different levels (75%, 100%, and 125%) and the % recovery of MH and PS was calculated. The LOD and LOQ were found to be 0.481 µg/ml and 0.670µg/ml for MH and 1.15µg/ml and 1.68µg/ml for PS respectively. All the statically analyses were within the standard limits. It proves that the method was repeatable and selective for the simultaneous. Therefore, the present study concludes that it can be successfully used for simultaneous estimation of MH and

PS in pure and pharmaceutical dosage forms. The developed method was found to be simple, precise, and accurate.[5]

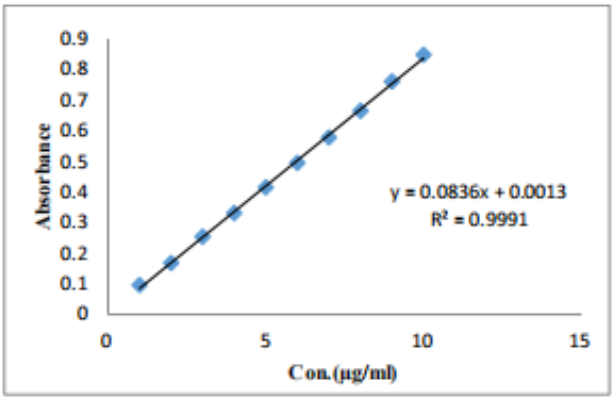

Fig. 2.Standard calibration curve

Kakade et al, 2019: estimated Metformin Hydrochloride in Bulk and Formulation by UV Spectroscopic Area Under Curve Method. The current work is carried out for estimation of Metformin Hydrochloride by using area under curve (AUC) method using UV visible spectrophotometer. For this purpose the wavelength range 221-241 nm was selected. Distilled water was used as a solvent throughout the work. Linearity was observed in concentration range 5-25 µg/ml (R2= 0.994) for the method. The present method was found to be simple and linear which can be used for routine quality control analysis for spectrophotometric estimation of Metformin hydrochloride in bulk.[6]

K Mishra et al,2011: developed and Validation of Metformin Hydrochloride in Tablet Dosage Form: A simple, reproducible and efficient method for the determination of Metformin hydrochloride (MET) was developed and validated. The analysis complied with Beer's law in the concentration range of 8-13 g/mL at 233 nm for MET. In our study the validation of analytical method for determination of MET by UV in tablets formulation was performed in accordance the parameters including-system suitability, specificity, limit of quantification, limit of detection, linearity of response, accuracy, precision (reproducibility & repeatability), robustness (change of wave length ±2 nm). [7]

Validation Parameter	MET
Recovery, %	99.91
Intraday precision, %RSD	1.312
Interday precision, %RSD	0.093
Linearity (r^2)	0.99996
Robustness, %RSD*	1.55
LOD, µg/mL*	1.0µg
LOQ, µg/mL*	3.0 µg
Specificity	Specific

Fig. 3. Analytical method validation

Ambadas R. Rote etal,2014: Estimated Metformin Hydrochloride by UV Spectrophotometric Method in Pharmaceutical Formulation. A simple and sensitive UV spectrophotometric method has been developed and validated for the estimation of metformin hydrochloride in tablet formulation. Metformin hydrochloride is determined spectrophotometrically at 232 nm using distilled water as solvent. It obeyed Beer's law in the range of 2-10 µg/ml. Percentage recovery of the drug for the proposed method ranged from 102-105% indicating no interference

of the tablet excipients. The proposed method was found to be accurate and precise for routine estimation of metformin hydrochloride in bulk and pharmaceutical formulation.[8]

Level of addition	Drug added µg/ml	Tablet solution	Drug found µg/ml	%recovery	SD	%RSD
80%	6	4.8	6.16	102.6		
100%	6	6	6.25	104.1	0.006164	1.09
120%	6	7.2	6.3	105		

SD-standard deviation, RSD-relative standard deviation

Fig. 4. Precision of Metformin hydrochloride

D Yuvraj Dilip et al, 2017: developed and Validation of UV-Spectrophotometric Method for Estimation of Metformin in Bulk and Tablet Dosage Form. Diabetes mellitus, a metabolic disorder characterized by increased blood sugar level. Metformin hydrochloride is used to treat type I Diabetes mellitus. Metformin hydrochloride chemically 1, 1-dimethylbiguanide hydrochloride, is white crystalline powder, hygroscopic and freely soluble in water, Officially UV spectrophotometric method used for estimation of Metformin Hydrochloride from the bulk and tablets formulations. Develop and validate a simple, rapid, accurate, economic and precise UV/VIS method for Metformin Hydrochloride in bulk and tablets formulation. of a common solvent were essential so various solvent ranges including methanol, ethanol, acetonitrile and phosphate buffer and various concentrations ranges of various buffers were analyzed. Among different solvents water has showed better results, hence water was selected as a solvent for the proposed method. Metformin Hydrochloride showed maximum absorbance at 234 nm. The percentage recoveries for Metformin Hydrochloride were found in the range of 99- 101 %. Method was quantitatively evaluated in terms of linearity, accuracy, precision, ruggedness, robustness and recovery. The method was simple, convenient and suitable for the determination of Metformin Hydrochloride from bulk and tablet dosage forms.[9]

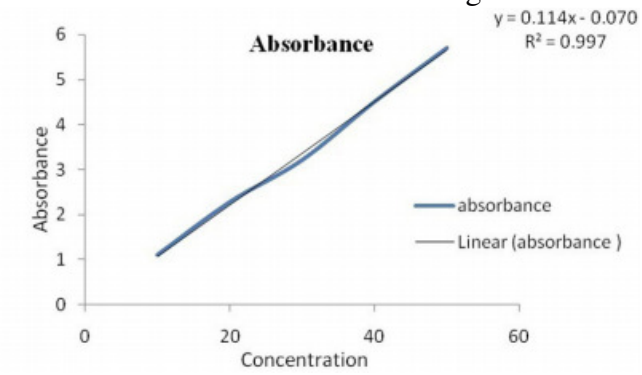

Fig. 5. Calibration curve of Metformin at 234 nm

HPLC METHOD:

N Nilesh et al, 2019: Was Developed and Validation of Metformin Hydrochloride by using RP-HPLC with ICH Guidelines. A simple and reproducible method was developed for Metformin (MET) by Reverse Phase High Performance Liquid Chromatography (RP-HPLC). Metformin was separated on C18 column [4.6x250mm, particle size 5µm], using combination of phosphate buffer with pH of 3.0 and Methanol at the UV detection of 238nm. Isocratic elution of phosphate buffer with pH of 3.0 and Methanol was used as a mobile phase with various ratios and flow rates, eventually 30:70 v/v phosphate buffer with pH of 3.0 and Methanol was being set with the flow rate of 1mL/min. The statistical validation parameters such as linearity, accuracy, precision, inter-day and intra-day variation were checked, assay studies of Metformin were within 98% to 102% indicating that the proposed method can be adoptable for quality control analysis of Metformin.[10]

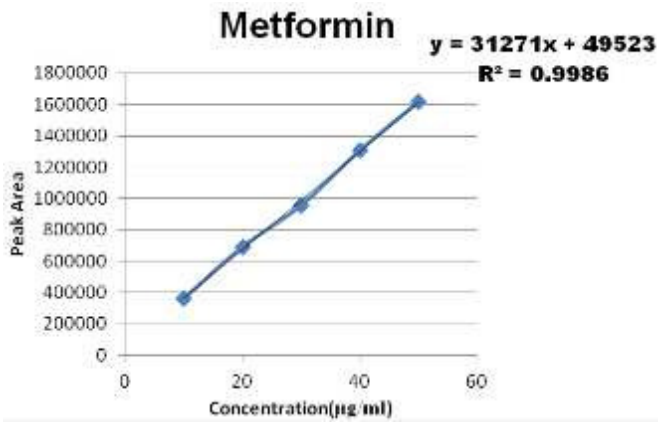

Fig. 6. Linearity

Saeed Arayne et al, 2006: Was developed and validation of RP-HPLC method for the analysis of metformin. The reversed-phase high-performance liquid chromatographic (RP-HPLC) method has been developed to quantify metformin hydrochloride (MfCl) in raw material and pharmaceutical formulations using C18 analytical reverse-phase column. Diazepam was used as an internal standard. Mobile phase consisted of methanol-water (30:70 v/v), pumped at a flow rate of 0.5 ml/min at ambient temperature and the retention time was about 4.4 min with symmetrical peaks. (MfCl) was detected by ultraviolet absorbance at 233 nm with no interference of commonly used excipients. The method was linear over the concentration range 0.312–5 µg/mL (R2 = 0.9995). The limit of detection of metformin was 0.1 µg/mL and the limit of quantitation was 0.3 µg/mL. The results obtained showed a good agreement with the declared contents in case of pharmaceutical formulations. The proposed method is rapid, accurate, economical and selective and it may be used for the quantitative analysis of metformin in Neodipar tablets because of its sensitivity and reproducibility.[11]

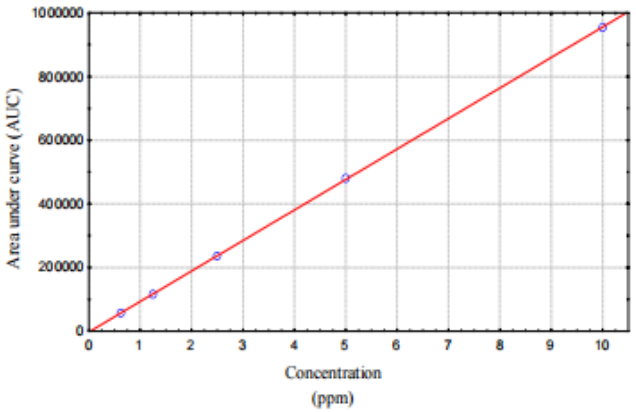

Fig. 7. Linearity of metformin in methanol: water (30: 70 v/v).

T Anil Kumar et al, 2023: developed analytical techniques for determination of metformin: present and perspectives. Metformin is an oral anti-diabetic drug in preventing complications of type 2 diabetes and it is a good first-line therapy for an over-obese with type 2 diabetes, it is currently available in more than 60 countries worldwide. As a result of the importance of this oral hypoglycaemic agent in the treatment of non-insulin-dependent diabetes mellitus, which leads to end-stage renal disease, this work aims to compile the published analytical methods reported so far in the literature for the determination in biological samples and pharmaceutical formulations. This article narrates different techniques like high performance liquid chromatography. It can be seen that high-performance liquid chromatography methods have been used extensively. Thus, this paper will help in the selection and development of proper analytical methodologies estimation of Metformin to achieve satisfactory results.[12]

Madiha Fatima, 2018: was review on Metformin and its gastrointestinal problems. Metformin is a biguanide class of drugs and has been recommended as first-line therapy for type 2 diabetes. It has a good safety profile, efficacy, comparatively reduced cost, and potential cardiovascular benefits. Metformin is an insulin-sensitizing agent, its bio availability is 50%-60%. Generally, A1C levels are lowered by 1.5% points by metformin mono therapy. Treatment with metformin decreases fasting plasma glucose concentrations by 25% to 30% and decreases the production of glucose. Metformin reduces hepatic glucose production and absorption of glucose in the intestine. In addition to it, decreases fatty acids oxidation. In liver and skeletal muscles the mitochondrial function and AMP Activated Protein Kinase (AMPK) activity are considered as potential mechanisms and has gained much attention by which metformin exerts its advantageous effects. In the gut enteroendocrine cells secret glucagon-like peptide-1 and glucose-dependent insulinotropic peptide, which are considered as important determinants for the disposal of glucose following a meal. Glucose production is reduced either by decreasing gluconeogenesis or by glycogenolysis. Treatment with metformin is, nevertheless, very often associated with gastrointestinal side effects and quality of life and treatment adherence is negatively affected in patients of type 2 diabetes. The most common gastrointestinal symptoms are diarrhea, heartburn, and nausea, followed by abdominal pain, bloating, and retching. The mechanism lying under gastrointestinal intolerance caused by metformin is unclear. However, there are different hypothesis proposed, including stimulation of intestinal secretion of serotonin, alteration in in-cretin and metabolism of glucose, and malabsorption of bile salts. Metformin is used clinically in diabetes, poly cystic ovary syndrome, and in obese for weight reduction. It has cardio protective effect and its use is recently being studied in cancer and HIV associated metabolic abnormalities.[13]

S K. Godasu et al, 2017: proposed Analytical Method Development and Validation of Metformin Hydrochloride by using RP-HPLC with ICH Guidelines. A simple and reproducible method was developed for Metformin (MET) by Reverse Phase High Performance Liquid Chromatography (RP-HPLC). Metformin was separated on C18 column [4.6x250mm, particle size 5µm], using combination of phosphate buffer with pH of 3.0 and Methanol at the UV detection of 238nm. Isocratic elution of phosphate buffer with pH of 3.0 and Methanol was used as a mobile phase with various ratios and flow rates, eventually 30:70 v/v phosphate buffer with pH of 3.0 and Methanol was being set with the flow rate of 1mL/min. The statistical validation parameters such as linearity, accuracy, precision, inter-day and intra-day variation were checked, assay studies of Metformin were within 98% to 102% indicating that the proposed method can be adoptable for quality control analysis of Metformin.[14]

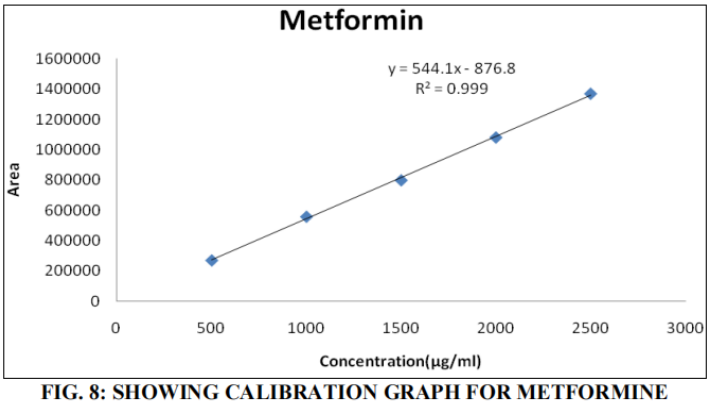

FIG. 8: SHOWING CALIBRATION GRAPH FOR METFORMINE

A Gedawy et al, 2019: developed and validation of a new analytical HPLC method for simultaneous determination of the anti diabetic drugs, metformin and gliclazide. An efficient and simple HPLC method has been developed and validated for the simultaneous determination of gliclazide and metformin hydrochloride in bulk and was applied on marketed metformin and

gliclazide products. The mobile phase used for the chromatographic runs consisted of 20 mm ammonium formate buffer (pH 3.5) and acetonitrile (45:55, v/v) The separation was achieved on an Alltima CN (250 mm 4.6 mm x5m) column using Isocratic mode. Drug peaks were well separated and were detected by a UV detector at 227 nm. The method was linear at the concentration range 1.25e150 mg/ml for gliclazide and 2.5e150 mg/ml for metformin respectively. The method has been validated according to ICH guidelines with respect to system suitability, specificity, precision, accuracy and robustness. Metformin limit of detection (LOD) and limit of quantification (LOQ) were 0.8 mg/ml and 2.45 mg/ml respectively while LOD and LOQ for gliclazide were 0.97 mg/ml and 2.95 mg/ml respectively.[15]

CONCLUSION:

Presented systematic review covers the current analytical methods for the determination of Metformin Hydrochloride. And UV, HPLC methods were found to be most widely used for Metformin. The presented information is useful for the future study for researcher involved in formulation development and quality control of Metformin Hydrochloride.

REFERENCE:

[1] Clifford J.B , Metformin: historical overview. *Diabetologia*, 2017; 60,1566–1576
[2] Madiha Fatima1, Sadeeqa1 S, Rashid Nazir S.U, Metformin and its gastrointestinal problems: A review. *Biomedical Research,* 2018; 29 (11): 2285-2289.
[3] Teja Kumar Reddy K, Vijaya Babu P, Rajinikanth S, & Sudhakar P. A review of artificial intelligence in treatment of covid-19. *Journal of Pharmaceutical Negative Results*,2022; 254–264.
[4] Foretz M, Metformin: From Mechanisms of Action to Therapies. *Cell Metabolism*, December 2, 2014;20.
[5] Sharma A, Kapil Kumar V , Inder Kumar b, Bala A , Bhumika T and Vandana T, Development and Validation of UV-Spectroscopic Method for Simultaneous Estimation of Metformin Hydrochloride and Pravastatin Sodium. *journal of Pharmaceutical Research International,* 2023; 35(6), 1-13.
[6] Kakade Vrushali B, Estimation of Metformin Hydrochloride in Bulk and Formulation by UV -Spectroscopic Area Under Curve Method .*Journal of Drug Delivery & Therapeutics.* 2019; 9(3):163-167
[7] Mishra K, Soni H, Nayak G, Sharan patel S and Singhai A.K, Method Of Development and Validation of Metformin Hydrochloride in Tablet Dosage Form. *Journal of Chemistry*, ISSN: 0973-4945, 2011, 8(3), 1309-1313
[8] Ambadas R. Rote1, Ravindranath B. Saudagar, Estimation of Metformin Hydrochloride by UV Spectrophotometric Method in Pharmaceutical Formulation. *World Journal of Pharmaceutical Sciences*, 2014;ISSN: 2321-3310.
[9] Dange *et al,* UV-Spectrometric validation of Metformin HCl. *Indian Journal of Pharmaceutical Education and Research,* Oct-Dec 2017; 51(4).
[10] N Nilesh, Avish M, Anil J, Prashant M , Analytical Method Development and Validation of Metformin Hydrochloride by using RP-HPLC with ICH Guidelines. *International Journal of Trend in Scientific Research and Development,* Mar-Apr 2019;ISSN: 2456 - 6470, 3(3).
[11] M.Saeed Arayne,Sultana N,Hashim zuberi M, Development and validation of -RP-HPLC method for the analysis of Metformin, *Pakistan Journal of Pharmaceutical Sciences,* 2006;19(3), 231-235.
[12] Anil Kumar T, A review of analytical techniques for determination of metformin: present and perspectives . *International Journal of Health care and Biological Sciences, 2023; 4(1), 18-24.*

[13] Madiha Fatima1 ,Sadeeqa S, Rashid Nazir S.U, Metformin and its Gastrointestinal problems: A review *Biomedical Research 2018; 29 (11): 2285-2289.*
[14] Godasu and Sreenivas, *A* new validated RP-HPLC method for the determination of Metformin HCl and Empagliflozin in its bulk and pharmaceutical dosage forms. *International journal of pharmaceutical science and research , 2017; 8(5): 2223-2232.*
[15] Ahmed Gedawy et al Development and validation of a new analytical HPLC method for simultaneous determination of the Anti Diabetic drugs, Metformin and Gliclazide *journal of food and drug analysis,* 2019;27, 315 - 322.

