

Simultaneous Estimation of Pioglitazone, Glimepiride & Metformin Hydrochloride in Bulk and Tablet Dosage Form by FTIR and LC-MS

Dr. Shoheb Shaikh

shoheb.shaikhnnsattha@gmail.com

RSM's NN Sattha college of Pharmacy Ahilynagar <https://orcid.org/0000-0001-5821-5109>

Research Article

Keywords: Pioglitazone, Glimepiride, Metformin HCl, FTIR spectroscopy, LC-MS, Method validation, pharmaceutical analysis

Posted Date: July 27th, 2026

DOI: <https://doi.org/10.21203/rs.3.rs-10469688/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.
[Read Full License](#)

Additional Declarations: The authors declare potential competing interests as follows: NA

Simultaneous Estimation of Pioglitazone, Glimepiride & Metformin Hydrochloride in Bulk and Tablet Dosage Form by FTIR and LC-MS

Abstract:

Background:

Combination therapy involving pioglitazone, glimepiride, and metformin hydrochloride is widely prescribed for the management of type 2 diabetes mellitus due to their complementary mechanisms of action. Accurate and simultaneous estimation of these drugs in bulk and combined dosage forms is essential to ensure quality, safety, and therapeutic efficacy. However, analytical challenges arise from differences in physicochemical properties and the concentration ranges of these drugs.

Objective:

The present study aims to develop and validate simple, rapid, and reliable Fourier Transform Infrared (FTIR) spectroscopic and Liquid Chromatography–Mass Spectrometry (LC-MS) methods for the simultaneous estimation of pioglitazone, glimepiride, and metformin hydrochloride in bulk and tablet dosage forms.

Methods:

FTIR analysis was performed using characteristic absorption peaks with suitable calibration models, while LC-MS analysis employed optimized chromatographic conditions with appropriate mobile phase composition and mass detection parameters. Method validation was carried out in accordance with ICH guidelines, evaluating parameters such as linearity, accuracy, precision, specificity, limit of detection (LOD), and limit of quantification (LOQ).

Results:

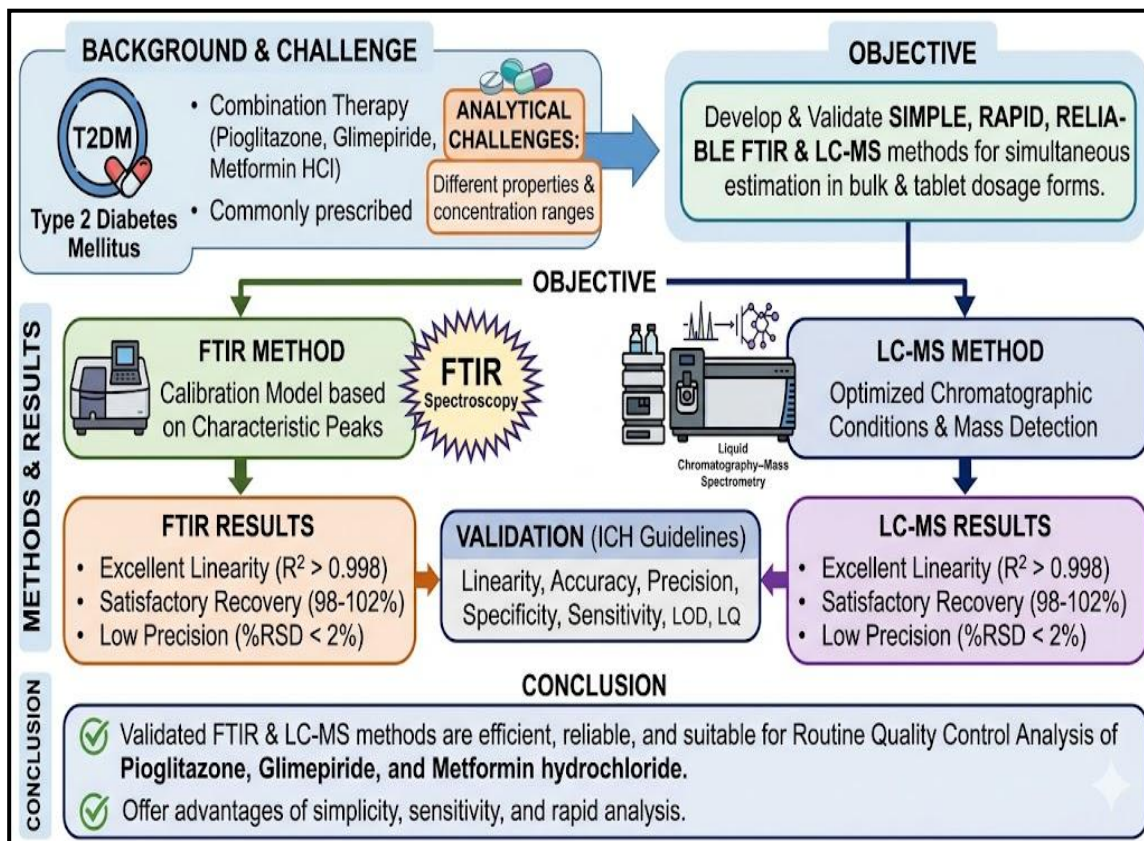
Both FTIR and LC-MS methods demonstrated excellent linearity within the selected concentration ranges, with correlation coefficients (R^2) exceeding 0.998. Accuracy studies showed satisfactory recovery (98–102%), and precision studies indicated low %RSD values (<2%). The developed methods were found to be specific, sensitive, and reproducible for all three drugs without interference from excipients.

Conclusion:

The validated FTIR and LC-MS methods are efficient, reliable, and suitable for routine quality control analysis of pioglitazone, glimepiride, and metformin hydrochloride in bulk and pharmaceutical formulations. These methods offer advantages of simplicity, sensitivity, and rapid analysis for multi-component drug estimation.

Keywords: Pioglitazone, Glimepiride, Metformin HCl, FTIR spectroscopy, LC-MS, Method validation, pharmaceutical analysis.

Graphical Abstract Image:



1. Introduction

1.1 Background

1.1.1 Diabetes Mellitus and Importance of Combination Therapy

Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The global burden of type 2 diabetes mellitus (T2DM) has increased significantly, necessitating effective therapeutic strategies. Combination therapy involving drugs with complementary mechanisms—such as insulin sensitizers, insulin secretagogues, and glucose production inhibitors—has emerged as a cornerstone in T2DM management. The combination of pioglitazone, glimepiride, and metformin hydrochloride offers synergistic glycemic control by targeting insulin resistance, pancreatic β -cell function, and hepatic glucose production simultaneously [1].

1.1.2 Analytical Challenges in Multi-Component Estimation

Simultaneous estimation of multi-drug formulations presents significant analytical challenges due to differences in polarity, solubility, concentration ratios, and spectral

overlap. These factors complicate accurate quantification, necessitating advanced analytical techniques capable of resolving such complexities with high precision and specificity [2].

1.2 Drug Profile

1.2.1 Pioglitazone (PPAR- γ Agonist):

Pioglitazone is a thiazolidinedione class drug that acts as a peroxisome proliferator-activated receptor gamma (PPAR- γ) agonist, enhancing insulin sensitivity in adipose tissue, liver, and skeletal muscle. It plays a crucial role in reducing insulin resistance in T2DM patients [3].

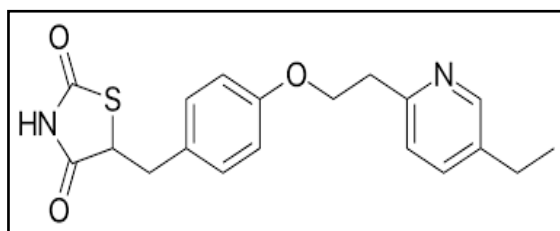


Fig. No.1 Pioglitazone³

1.2.2 Glimepiride (Sulfonylurea)

Glimepiride belongs to the sulfonylurea class and functions by stimulating insulin secretion from pancreatic β -cells. It improves glycemic control by increasing endogenous insulin availability [4].

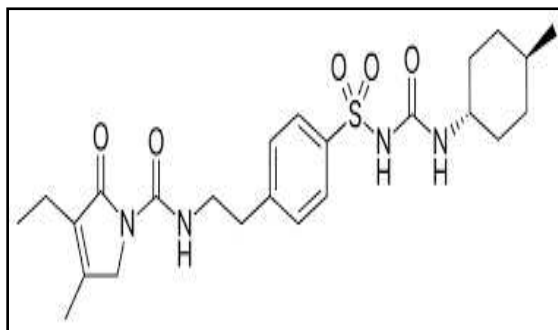


Fig. No.2 Glimepiride⁴

1.2.3 Metformin Hydrochloride (Biguanide)

Metformin hydrochloride, a biguanide, is the first-line therapy for T2DM. It primarily decreases hepatic glucose production and enhances peripheral glucose uptake without causing significant hypoglycemia [5].

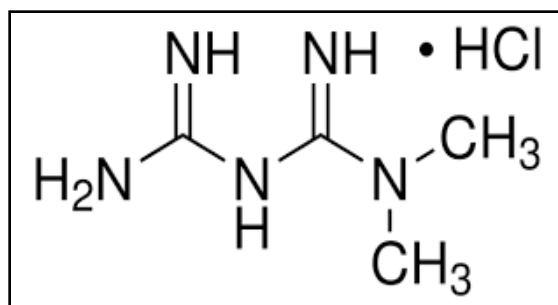


Fig. No.3 Metformin Hydrochloride⁵

1.3 Literature Review

1.3.1 Existing Analytical Methods (HPLC, UV, LC-MS)

Various analytical methods have been reported for the estimation of antidiabetic drugs, including high-performance liquid chromatography (HPLC), UV-visible spectrophotometry, and LC-MS. HPLC methods are widely used due to their robustness and reproducibility, while UV methods offer simplicity and cost-effectiveness. LC-MS techniques provide high sensitivity and selectivity, especially for trace-level analysis and complex matrices [6–8].

1.3.2 Limitations of Current Methods

Despite their advantages, existing methods have several limitations. HPLC methods often require extensive sample preparation, longer analysis time, and the use of large volumes of organic solvents. UV spectrophotometric methods suffer from poor specificity due to spectral overlap. Although LC-MS is highly sensitive, it involves high operational costs and requires skilled personnel. These limitations highlight the need for alternative analytical approaches that are rapid, cost-effective, and environmentally friendly [9].

1.4 Rationale of Study

1.4.1 Need for Rapid, Eco-Friendly FTIR Method

Fourier Transform Infrared (FTIR) spectroscopy has gained attention as a green analytical technique due to minimal solvent usage, rapid analysis, and non-destructive nature. It offers a promising alternative for simultaneous drug estimation using characteristic functional group vibrations [10].

1.4.2 Sensitivity and Specificity Advantages of LC-MS

LC-MS combines the separation efficiency of liquid chromatography with the detection power of mass spectrometry, enabling precise identification and quantification of analytes even at low concentrations. It is particularly useful for confirming results obtained from other analytical techniques [11].

1.5 Objective

1.5.1 To Develop and Validate FTIR and LC-MS Methods for Simultaneous Estimation

The primary objective of this study is to develop and validate simple, accurate, precise, and reliable FTIR and LC-MS methods for the simultaneous estimation of pioglitazone, glimepiride, and metformin hydrochloride in bulk and tablet dosage forms, in accordance with ICH guidelines.

1.6 Materials and Methods

1.6.1 Materials

1.6.1.1 Pure Drug Samples

Reference standards of pioglitazone, glimepiride, and metformin hydrochloride were obtained as gift samples from a certified pharmaceutical manufacturer. All standards were of analytical grade with purity $\geq 99\%$ and used without further purification [12].

1.6.1.2 Tablet Formulation Details

Commercially available tablet formulations containing a fixed-dose combination of pioglitazone, glimepiride, and metformin hydrochloride were procured from the local market. The label claim was recorded, and tablets were stored under appropriate conditions before analysis [13].

1.6.1.3 Chemicals and Reagents

All solvents and reagents, including methanol, acetonitrile, formic acid, and potassium bromide (KBr), were of analytical or HPLC grade. Ultrapure water was obtained using a Milli-Q purification system [14].

1.6.2 Instrumentation

FTIR Spectrophotometer

FTIR analysis was performed using a double-beam FTIR spectrophotometer equipped with ATR (Attenuated Total Reflectance) accessory and compatible spectral analysis software. The instrument was operated within a range of $4000\text{--}400\text{ cm}^{-1}$ [15].

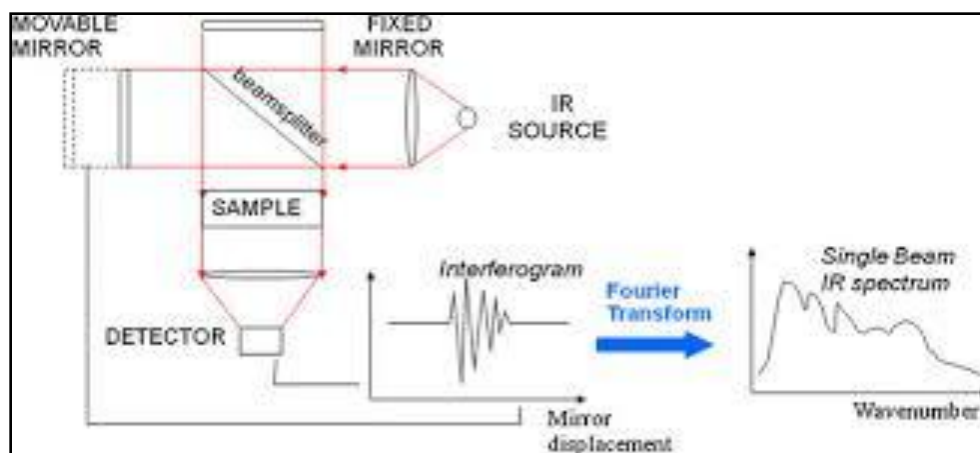


Fig. No.4 FTIR Spectrophotometer¹⁵

LC-MS System:

LC-MS analysis was carried out using a high-performance liquid chromatography system coupled with a mass spectrometer. Separation was achieved using a C18 reverse-phase column (e.g., 150 mm × 4.6 mm, 5 μm particle size), and detection was performed using an electrospray ionization (ESI) source with a triple quadrupole mass analyzer [16].

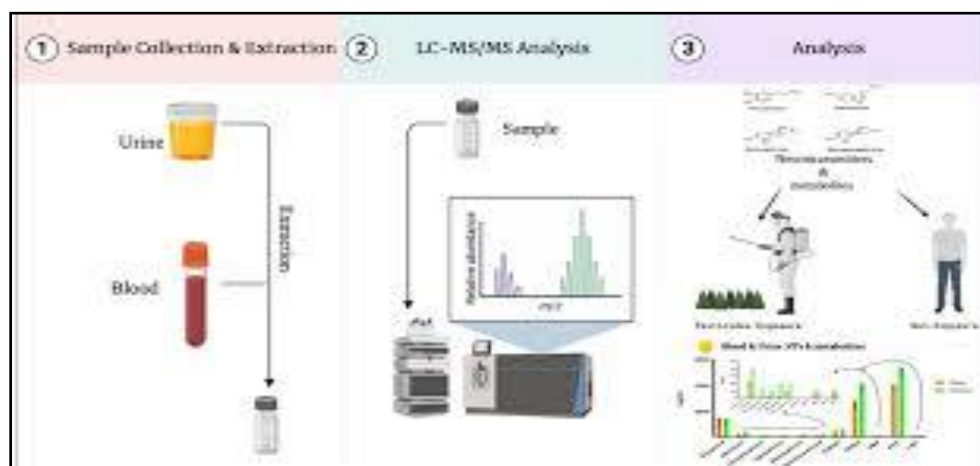


Fig. No.5 LC-MS System¹⁶

1.7 Method Development

1.7.1 FTIR Method

Sample Preparation: Samples were prepared using KBr pellet technique and ATR method for direct analysis. Powdered drug samples and tablet mixtures were finely triturated with KBr and compressed into pellets [17].

Selection of Characteristic Peaks: Distinct absorption peaks corresponding to functional groups of each drug were identified and selected to avoid spectral overlap.

Calibration Model Development: Calibration curves were constructed by plotting absorbance versus concentration for each drug using selected peaks [18].

1.7.2 LC-MS Method

Chromatographic Conditions:

Column Type: Reverse-phase C18 column

Mobile Phase Composition: Mixture of acetonitrile and water with 0.1% formic acid (gradient or isocratic mode)

Flow Rate: ~0.8–1.0 mL/min

Injection Volume: 10–20 μ L [19]

1.7.3 Mass Spectrometry Conditions:

Ionization Mode: Electrospray ionization (ESI) in positive and/or negative mode

m/z Transitions: Specific precursor-to-product ion transitions were selected for each drug for quantification [20]

1.7.4 Preparation of Standard Solutions

Stock solutions (1000 μ g/mL) of each drug were prepared in methanol. Working standard solutions were prepared by appropriate dilution with mobile phase or solvent system [21].

1.7.5 Calibration Curve

Concentration Range: Selected based on expected assay levels (e.g., 5–50 μ g/mL)

Regression Analysis: Calibration curves were plotted, and linear regression equations along with correlation coefficients (R^2) were determined [22]

1.8 Method Validation (as per ICH Q2(R1))

1.8.1 Linearity

Linearity was evaluated by analysing standard solutions at multiple concentrations and assessing the correlation coefficient [23].

1.8.2 Accuracy (Recovery Studies)

Accuracy was determined by the standard addition method at 80%, 100%, and 120% levels, and % recovery was calculated [24].

1.8.3 Precision

Repeatability: Intra-day precision assessed by analyzing multiple replicates

Intermediate Precision: Inter-day variation evaluated over different days [25]

1.8.4 Limit of Detection (LOD)

LOD was calculated based on signal-to-noise ratio or standard deviation of response and slope [26].

1.8.5 Limit of Quantification (LOQ)

LOQ was determined similarly, ensuring acceptable precision and accuracy at the lowest quantifiable level [27].

1.8.6 Specificity

Specificity was assessed by analyzing the ability of the method to measure analytes in the presence of excipients [28].

1.8.7 Robustness

Robustness was evaluated by making small deliberate changes in analytical conditions and observing their effects [29].

1.8.9 System Suitability (for LC-MS)

System suitability parameters such as retention time, peak area, resolution, and theoretical plates were evaluated prior to analysis [30].

1.9 Results

The spectra of all three drugs, pioglitazone, metformin and glimepiride, are shown below for Infrared spectroscopy and for mass spectroscopy.

FTIR Spectroscopy:

Spectra of Metformin:

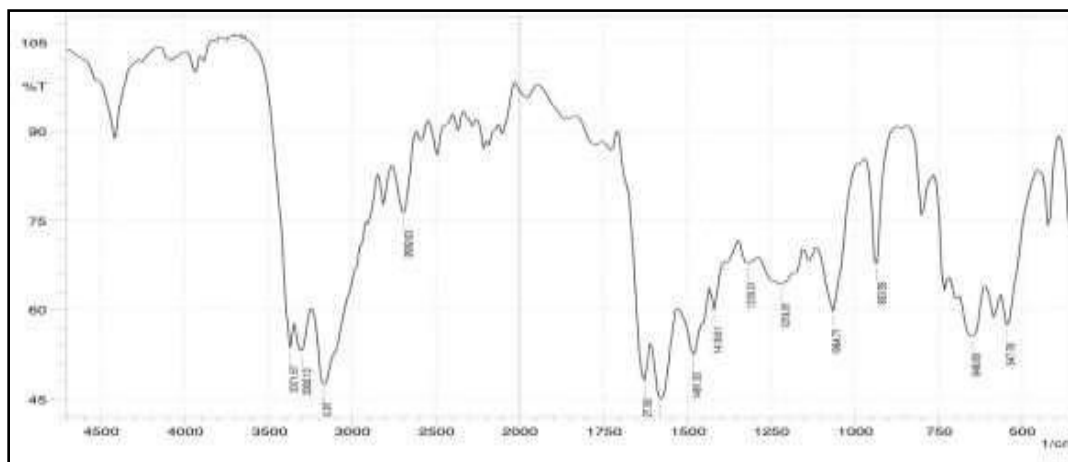


Fig. No.6 IR Spectrum of Metformin¹⁷

Sr. No.	Functional Group	Standard Range (cm ⁻¹)	Observed Range (cm ⁻¹)
1.	N-H Stretch	3500-3350 cm ⁻¹	3371.57 cm ⁻¹
2.	-C-H	2950-2840 cm ⁻¹	2862.36 cm ⁻¹

Interpretation of FTIR Spectrum of Metformin

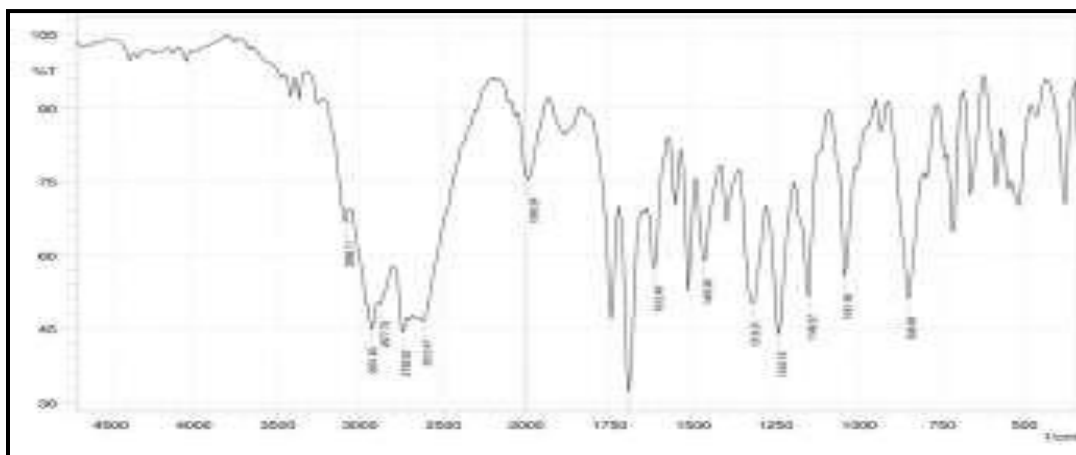


Fig. No.7 IR Spectrum of Pioglitazone

Sr. No.	Functional Group	Standard Range (cm ⁻¹)	Observed Range (cm ⁻¹)
1.	Symmetric CH Stretching	3000-2850cm ⁻¹	2931.8 cm ⁻¹
2.	=C-H stretch	3100-3000 cm ⁻¹	3086.11 cm ⁻¹
3.	-C-H	2950-2840 cm ⁻¹	2877.79 cm ⁻¹
4.	C=O amide	1700-1500 cm ⁻¹	1689.64 cm ⁻¹ 1612.49 cm ⁻¹
5.	CH ₂ bend	1480-1440 cm ⁻¹	1465.9 cm ⁻¹
6.	C-O-C stretch	1250-1050 cm ⁻¹	1242.16 cm ⁻¹ 1149.57 cm ⁻¹

			1041.56 cm ⁻¹
--	--	--	--------------------------

Interpretation of FTIR Spectrum of Pioglitazone

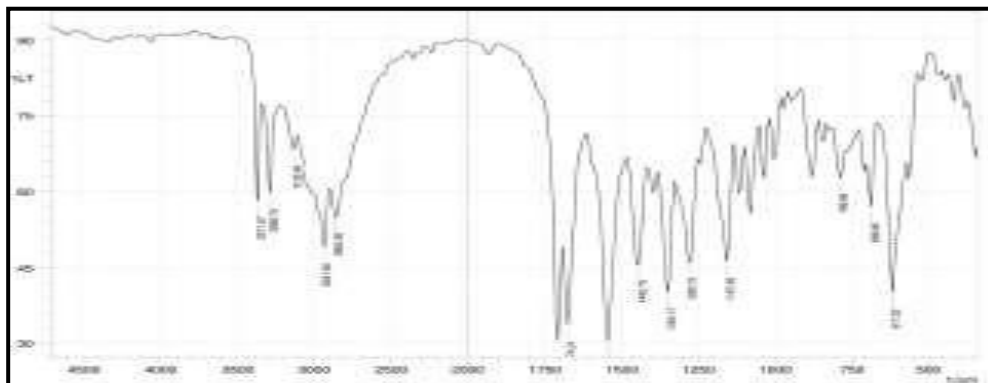


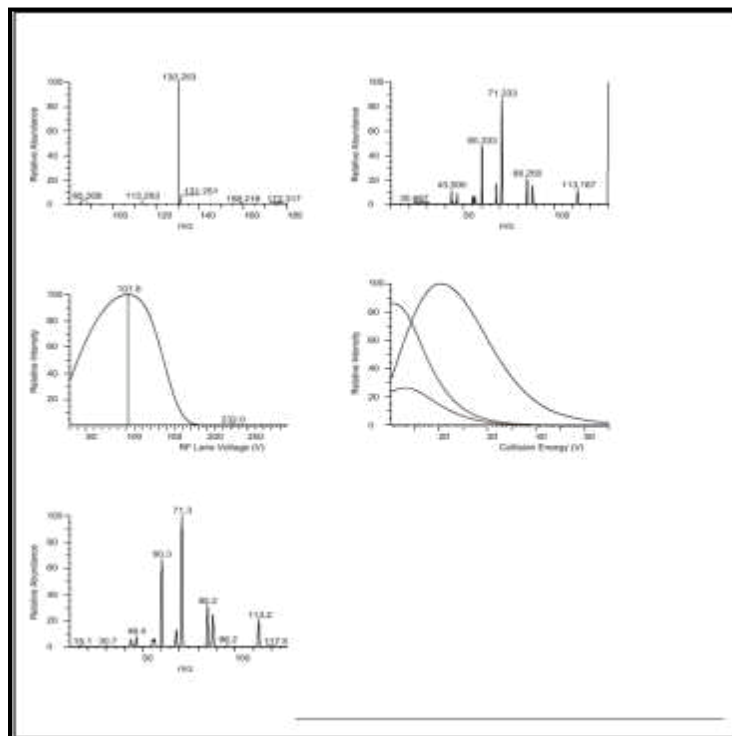
Fig. No.8 FTIR SpectraIR Spectrum of Glimepiride

Sr. No.	Functional Group	Standard Range (cm ⁻¹)	Observed Range (cm ⁻¹)
1.	Symmetric CH Stretching	3000-2850cm ⁻¹	2931.8 cm ⁻¹
2.	N-H Stretch	3500-3350 cm ⁻¹	3371.57 cm ⁻¹
3.	-C-H	2950-2840 cm ⁻¹	2862.36 cm ⁻¹
4.	C=O amide	1700-1500 cm ⁻¹	1543.05 cm ⁻¹ 1674.21 cm ⁻¹
5.	CH2 bend	1480-1440 cm ⁻¹	1442.75 cm ⁻¹
6.	C=C Aromatic	1600-1400 cm ⁻¹	1674.21 cm ⁻¹ 1543.05 cm ⁻¹ 1442.75 cm ⁻¹

Interpretation of FTIR Spectrum of Glimepiride

Liquid Chromatography Mass Spectrometry (LC-MS).

Fig. No.9 Mass Spectra of Metformin



Molecular weight = 129.16g/mol

- **Observed Molecular weight = 130.25 g/mol that's the molecular ion peak.**
- **131.25 ion peak could be the isotopic peak.**
- **113.253 for this expected removed fragment would be -NH₂.**
- **85- Changes of fragmentation of 2-NH₂ moieties.**
- **71 is the fragmentation of nascent 'N' as molecular weight is reduced by 14 only. (Base Peak).**
- **60 ion peak could be the fragmentation of C₃H₇N₂.**

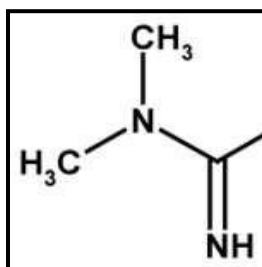


Fig. No.10 Fragment of metformin

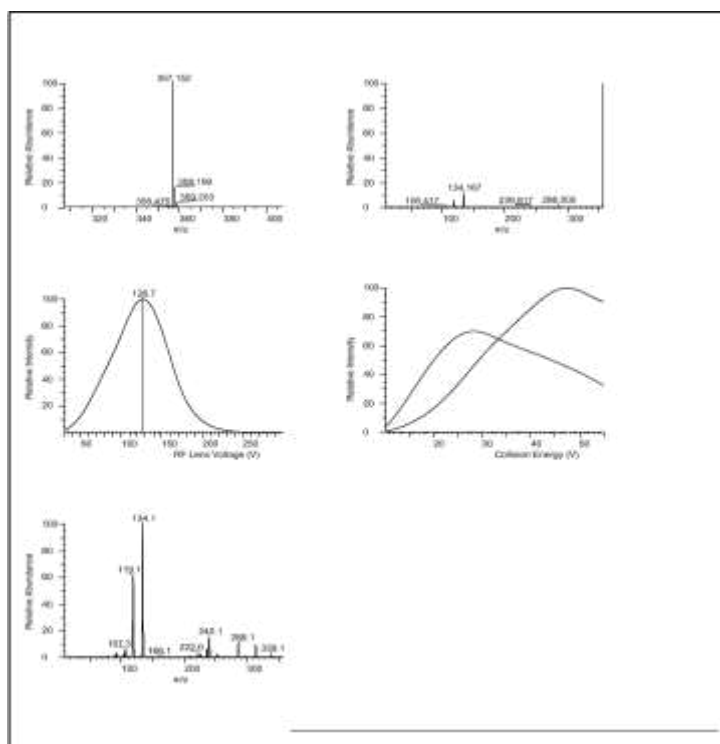


Fig. No.11 Mass Spectra of Pioglitazone

Molecular weight = 356.4g/mol.

- 357.1g/mol is the molecular ion peak.
- 358g/mol would be the isotopic peak
- 134g/mol is base peak:

- 119g/mol is due to fragmentation of -CH₃ after the base peak.

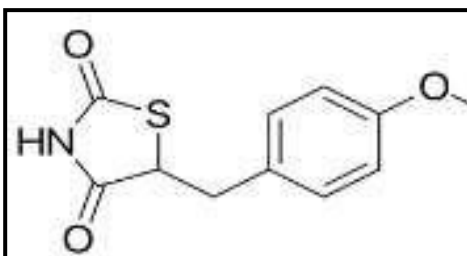


Fig No.12 Fragment of pioglitazone

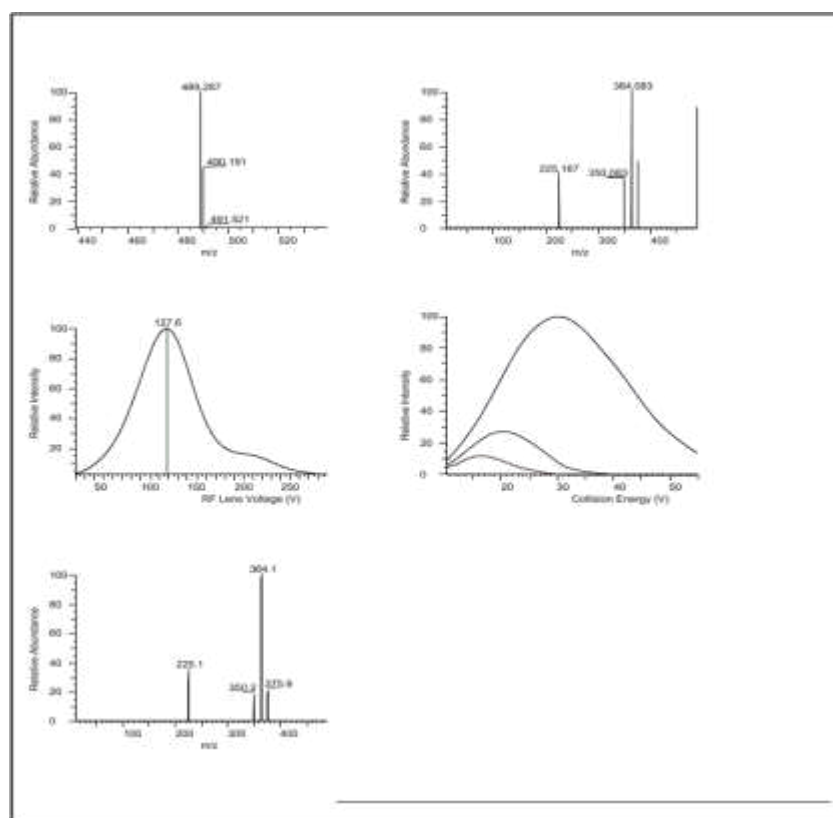
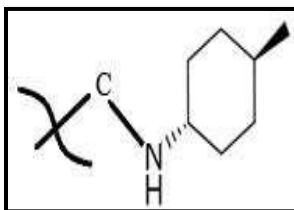


Fig No.13 Mass Spectra of Glimepiride

Molecular Weight = 490.6 g/mol.

- **Observed Molecular weight – 489.28g/mol.**
- **491 may be the isotopic peak.**
- **364 is base peak which is the fragmentation of below image as shown:**

Fig No.14 Fragment of glimepiride 01



- 350 is the further fragmentation of nascent 'N'.
- 225 is the further fragmentation of the 350 fragments, whose fragment is shown in the below image:

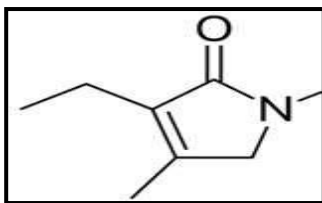


Fig No.15 Fragment of glimepiride 02

1.10 Discussion

For simultaneous estimation of metformin, pioglitazone, and Glimepiride, an analytical method was developed and scanned for infrared spectroscopy and liquid chromatography mass spectrometry. The scanned results were found to be accurate and precise. All three drugs which was studied at their respective concentration. More than 0.99 regression coefficient values were observed, which were within the limit for mass analysis. With the help of the FTIR, the functional group was analysed, and with the mass spectra, expected fragmentation was observed, which will help to elucidate the structure of all three drugs, that is of Pioglitazone, Glimepiride and Metformin. The method was found to be simple, rapid and robust for the analysis of metformin, Pioglitazone and Glimepiride. [31-33].

1.11 Conclusion

A novel LC-MS and FTIR method was analysed for the simultaneous estimation of Pioglitazone, Glimepiride and metformin in bulk and pharmaceutical dosage form. The obtained results were found to be satisfactory, indicating the analysis was sensitive, rapid, accurate and relevant for all three drugs that are carried out using guidelines of the Indian Council of Harmonisation. Hence, the proposed method was carried out for the routine analysis in the tablet dosage form, which in turn helps confirm the drugs by elucidating the respective structures of the three drugs.

Declarations:

Data Availability Statement: All data generated or analysed during this study are included in this published article.

Consent to Participate: Not applicable. This study did not involve human participants or animals.

Ethics Approval: Not applicable for this review article.

Consent for Publication: Not applicable.

Availability of Data: No new data were created or analysed in this study.

Funding: No funds received from anywhere while writing this manuscript

Conflict of Interest: The authors declare no conflict of interest.

Acknowledgement: We acknowledge the use of artificial intelligence tools, specifically ChatGPT (OpenAI, GPT-5.2), to assist in generating graphical abstract and Napkin AI tool for other diagrams and visualizations for more detailed elaboration of the contents which later enhanced by Adobe Illustrator. Prompts were designed to emulate BioRender-style illustrations. All outputs were reviewed, modified, and validated by the authors to ensure scientific accuracy and originality. All the references were made by using Elsevier's Mendeley and plagiarism and AI contents were checked using Turnitin and Quillbot premium.

1.12. References:

1. Jadon AS, Kaushik MP, Anitha K, Bhatt S, Bhadauriya P, Sharma M. 2024. Types of diabetes mellitus, mechanism of insulin resistance and associated complications. In: *Biochemical Immunology of Diabetes and Associated Complications*. Academic Press. p. 1–18.
2. Snyder LR, Kirkland JJ, Dolan JW. 2010. *Introduction to Modern Liquid Chromatography*. 3rd ed. New York: Wiley.
3. Yki-Järvinen H. 2004. Thiazolidinediones. *N Engl J Med*. 351(11):1106–1118.
4. Holstein A, Egberts EH. 2003. Risk of hypoglycemia with oral antidiabetic agents. *Exp Clin Endocrinol Diabetes*. 111(7):405–414.
5. Rena G, Hardie DG, Pearson ER. 2017. The mechanisms of action of metformin. *Diabetologia*. 60(9):1577–1585.
6. Beckett AH, Stenlake JB. 2002. *Practical Pharmaceutical Chemistry*. 4th ed. London: CBS Publishers.

7. Morkhade S, Vaishnav MS. 2025. Role of reverse phase high performance liquid chromatography (RP-HPLC) in simultaneous estimation of anti-diabetic drug. *Journal of Advance and Future Research*. 3(11):470–495.
8. Niessen WMA. 2006. *Liquid Chromatography–Mass Spectrometry*. 3rd ed. Boca Raton: CRC Press.
9. Deshpande MM, Bhalerao MH, Pabale PD. 2022. A review on impurity profiling, degradation studies, and bioanalytical methods of anti-diabetic drugs. *Journal of Pharmaceutical Research International*. 34(34B):43–71.
10. Ramakrishna B, Mondal SU. 2021. A review of analytical methods for determination of type-II antidiabetic drugs in pharmaceuticals and biological matrices. *Asian J Pharm Clin Res*. 14(1):69–76.
11. Gross JH. 2011. *Mass Spectrometry: A Textbook*. 2nd ed. Berlin: Springer.
12. Chaudhari P, Prajapati M, Patel N, Patel D, Prajapati A, Patel K. 2024. Newer approaches for analytical method validation of anti-diabetic agents: A review. *International Journal of Innovative Science and Research Technology*. 9(12):650–669.
13. Stuart B. 2004. *Infrared Spectroscopy: Fundamentals and Applications*. Chichester: Wiley.
14. Patidar A, Kamble P. 2025. A comprehensive review on liquid chromatography-mass spectrometry (LC-MS): A hyphenated technique. *Asian Journal of Pharmaceutical Research and Development*. 13(1):95–103.
15. Marie AA, Hammad SF, Salim MM, Elkhodary MM, Kamal AH. 2023. Deduction of the operable design space of the RP-HPLC technique for the simultaneous estimation of metformin, pioglitazone, and glimepiride. *Scientific Reports*. 13(1):4334.
16. Shende MA, AA. 2022. Spectrophotometric method for simultaneous estimation of metformin, voglibose, and pioglitazone in triple fixed combined pharmaceutical dosage form. *International Journal of Innovative Research in Technology*. 8:96–102.
17. Zhao D, Wimalasinghe RM, Wang L, Rustum AM. 2022. Development and validation of a reversed-phase high-performance liquid chromatography (RP-HPLC) method for identification, assay and estimation of related substances of ivermectin in bulk drug batches of ivermectin drug substance. *Journal of Chromatographic Science*. 60(7):620–632.
18. Mohandoss S, Velu KS, Wahab R, Ahmad N, Dhanasekaran K, Napoleon AA, Akhtar MS, Lee YR. 2024. Synergistic effects of β -cyclodextrin derivative inclusion complexes with favipiravir to enhance solubility and antiviral efficacy

- against Herpes Simplex virus type 1 (HSV-1). *Journal of Molecular Liquids*. 413:126064.
19. Rahnenführer J, De Bin R, Benner A, Ambrogi F, Lusa L, Boulesteix AL, Migliavacca E, Binder H, Michiels S, Sauerbrei W, McShane L. 2023. Statistical analysis of high-dimensional biomedical data: A gentle introduction to analytical goals, common approaches and challenges. *BMC Medicine*. 21(1):182.
 20. Zuvela P, Skoczylas M, Jay Liu J, Bączek T, Kaliszan R, Wong MW, Buszewski B. 2019. Column characterization and selection systems in reversed-phase high-performance liquid chromatography. *Chemical Reviews*. 119(6):3674–3729.
 21. Naidong W, Shou WZ, Addison T, Maleki S, Jiang X. 2002. Liquid chromatography/tandem mass spectrometric bioanalysis using normal-phase columns with aqueous/organic mobile phases—a novel approach of eliminating evaporation and reconstitution steps in 96-well SPE. *Rapid Communications in Mass Spectrometry*. 16(20):1965–1975.
 22. Chen HY, Chen C. 2022. Evaluation of calibration equations by using regression analysis: An example of chemical analysis. *Sensors*. 22(2):447.
 23. Musuku A, Tan A, Awaiye K, Trabelsi F. 2013. Comparison of two-concentration with multi-concentration linear regressions: Retrospective data analysis of multiple regulated LC–MS bioanalytical projects. *Journal of Chromatography B*. 934:117–123.
 24. Saxberg BE, Kowalski BR. 1979. Generalized standard addition method. *Analytical Chemistry*. 51(7):1031–1038.
 25. O'Connor E, McGovern FM, Byrne DT, Boland TM, Dunne E, McHugh N. 2021. Repeatability of gaseous measurements across consecutive days in sheep using portable accumulation chambers. *Journal of Animal Science*. 99(11):skab288.
 26. Desimoni E, Brunetti B. 2015. About estimating the limit of detection by the signal-to-noise approach. *Pharmaceutica Analytica Acta*. 6(3):1–4.
 27. Ng B, Quinete N, Gardinali PR. 2020. Assessing accuracy, precision and selectivity using quality controls for non-targeted analysis. *Science of the Total Environment*. 713:136568.
 28. Chadha R, Bhandari S. 2014. Drug–excipient compatibility screening—role of thermoanalytical and spectroscopic techniques. *Journal of Pharmaceutical and Biomedical Analysis*. 87:82–97.
 29. Ferreira SL, Caires AO, Borges TD, Lima AM, Silva LO, dos Santos WN. 2017. Robustness evaluation in analytical methods optimized using experimental designs. *Microchemical Journal*. 131:163–169.

30. Abbatiello SE, Mani DR, Schilling B, MacLean B, Zimmerman LJ, Feng X, Cusack MP, Sedransk N, Hall SC, Addona T, Allen S. 2013. Design, implementation and multisite evaluation of a system suitability protocol for the quantitative assessment of instrument performance in liquid chromatography-multiple reaction monitoring-MS (LC-MRM-MS). *Molecular & Cellular Proteomics*. 12(9):2623–2639.
31. Pandit V, Pai RS, Singh G, Devi K, Narayana S, Suresh S. 2012. Development and validation of the liquid chromatographic method for simultaneous estimation of metformin, pioglitazone, and glimepiride in pharmaceutical dosage forms. *Pharmaceutical Methods*. 3(1):9–13.
32. Nedelcu RG, Persson AS. 2014. Scanning accuracy and precision in 4 intraoral scanners: An in vitro comparison based on 3-dimensional analysis. *The Journal of Prosthetic Dentistry*. 112(6):1461–1471.
33. Shaikh S. 2021. Simultaneous estimation of pioglitazone, glimepiride, metformin hydrochloride in bulk tablet dosage form by UV, RP-HPLC method. *International Journal of Pharmaceutical Chemistry and Analysis*.