

Development and Validation of a Stability-Indicating RP-UPLC Method for the Simultaneous Determination of Glimepiride and Lobeglitazone Sulfate in Pharmaceutical Dosage Forms

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Abstract

A simple and rapid RP-UPLC method was developed and validated for the simultaneous analysis of Glimepiride (GLM) and Lobeglitazone sulfate (LBZ) in pharmaceutical formulations. Chromatographic separation was performed on a Phenyl column using a mobile phase of 0.1% formic acid and acetonitrile (70:30, v/v) at 0.5 mL min⁻¹, with detection at 266 nm. Retention times were 1.267 min for GLM and 1.876 min for LBZ. Linearity was achieved over 25–150 µg/mL for GLM and 12.5–75 µg/mL for LBZ ($R^2 = 0.99956$ and 0.99980 , respectively). Recoveries ranged from 99.8–100.3% (GLM) and 99.3–100.1% (LBZ), with method precision (%RSD) below 0.64% and LOD/LOQ values of 0.6/2.0 µg/mL (GLM) and 0.3/1.0 µg/mL (LBZ). Stress degradation studies established its stability-indicating capability, with peroxide treatment producing the highest degradation (15.9% for GLM and 16.5% for LBZ). Peak purity analysis demonstrated that analyte peaks remained free from interference by degradation products, indicating the validated method is reliable for routine quality assessment and stability evaluation of Glimepiride and Lobeglitazone sulfate in formulations.

Keywords: Forced degradation, Glimepiride, Lobeglitazone sulfate, Method validation, RP-UPLC, Stability-indicating method.

Introduction

Type 2 diabetes mellitus (T2DM), the most common form of diabetes, comprises 90–95% of global cases and is driven by defects in insulin secretion, insulin action, or both (1,2). The worldwide diabetic population is projected to increase from 463 million adults in 2019 to nearly 700 million by 2045, reinforcing the importance of effective glycaemic control and pharmaceutical quality assurance (3-7).

Glimepiride (GLM) is a third-generation sulfonylurea that promotes insulin release from pancreatic β -cells, while Lobeglitazone (LBZ) is a PPAR- γ agonist that improves insulin sensitivity and peripheral glucose uptake (8–11). Owing to their complementary pharmacological actions, their fixed-dose combination provides effective glycaemic control in patients with T2DM (Figure 1).

Several analytical methods have been reported for the estimation of glimepiride, including RP-HPLC, HPLC, RP-UFLC, and UV spectrophotometric methods for bulk drugs,

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pharmaceutical formulations, degradation studies, and biological samples (12–17,20). In contrast, analytical methods for lobeglitazone are limited, with most studies focusing on LC-MS/MS-based bioanalytical quantification in rat and human plasma for pharmacokinetic applications (18,19). Although these methods offer high sensitivity, they require sophisticated instrumentation and are unsuitable for routine quality control. Simultaneous chromatographic methods involving glimepiride with other antidiabetic agents, such as pioglitazone, rosiglitazone, and metformin, have also been reported (21–24). However, to the best of our knowledge, no rapid, stability-indicating RP-UPLC method has been reported for the simultaneous estimation of glimepiride and lobeglitazone in pharmaceutical dosage forms.

Despite these reports, the published literature demonstrates that available chromatographic methods primarily focus on the individual estimation of Glimepiride, bioanalysis of Lobeglitazone, or simultaneous estimation of Glimepiride with other antidiabetic agents. Conventional RP-HPLC methods are time-consuming and solvent-intensive, whereas LC-MS/MS methods are costly for routine quality control. To our knowledge, no validated stability-indicating RP-UPLC method has been developed for the simultaneous estimation of Glimepiride and Lobeglitazone in combined dosage forms, representing a notable analytical gap.

RP-UPLC was selected in the present study because it provides higher chromatographic efficiency, improved resolution, shorter run time, and lower solvent consumption than conventional HPLC. A phenyl stationary phase was selected because both analytes possess multiple aromatic rings capable of π – π interactions with the phenyl ligand, resulting in improved selectivity and peak symmetry. Furthermore, 0.1% formic acid was employed as the aqueous mobile-phase modifier to suppress analyte ionisation, reduce peak tailing, improve reproducibility, and ensure compatibility with future LC-MS applications. This study aimed to

develop and validate a rapid, stability-indicating RP-UPLC method for the simultaneous estimation of glimepiride and lobeglitazone in pharmaceutical dosage forms.

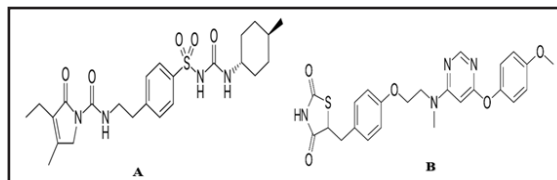


Figure .1 Chemical Structure of A) Glimepiride and B) Lobeglitazone

Material and Methods

Instrumentation

Chromatographic analysis was performed on an Agilent 1290 Infinity II UPLC system equipped with a quaternary pump, autosampler, vacuum degasser, and variable wavelength detector using Empower® 2 chromatography software. A Shimadzu UV–Visible spectrophotometer was used for wavelength selection.

Reagents and chemicals

Glimepiride and lobeglitazone sulfate reference standards were procured from Supriya Life Science Ltd., Mumbai, India. HPLC-grade solvents and reagents were purchased from Merck Life Science Pvt. Ltd., Mumbai, India, and Milli-Q water was used in all analytical preparations. Only AR- and HPLC-grade chemicals were employed.

Standard solution preparation

A stock solution containing 50 mg of lobeglitazone and 100 mg of glimepiride was prepared in a 100 mL volumetric flask using the diluent and sonication for 15 min. The working standard was obtained by diluting 5 mL of the stock solution to 50 mL, resulting in concentrations of 50 μ g/mL lobeglitazone and 100 μ g/mL glimepiride.

Determination of wavelength

A PDA detector was used to record the absorption spectrum of the drug solution between 200 and 400 nm, using a 70:30 (v/v) mix-

ture of 0.1% formic acid and acetonitrile as the blank. At 266 nm, the absorption curve displays the isosbestic point. Thus, using the UPLC chromatographic process, a detector wavelength of 266 nm was used (Figure 2).

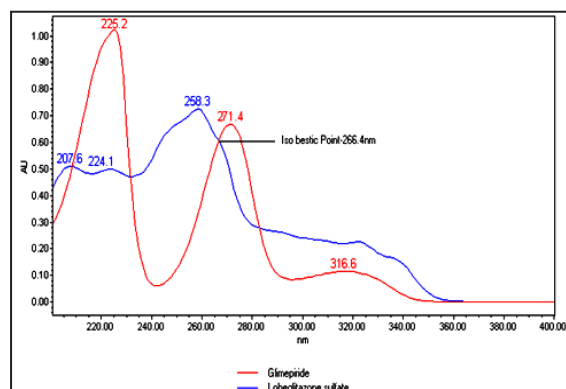


Figure. 2 UV spectrum of GLM and LBZ. The UV absorption spectra of Glimepiride and Lobeglitazone were recorded using a photodiode array (PDA) detector.

Method optimisation strategy

Preliminary chromatographic experiments were carried out to identify conditions capable of providing complete separation of Glimepiride and Lobeglitazone with acceptable peak symmetry, theoretical plates, and analy-

sis time. Different stationary phases, including BEH C18 and Phenyl columns, were evaluated together with orthophosphoric acid and formic acid buffers at different organic solvent compositions.

During Trial 1, an Acquity BEH C18 column with 0.1% orthophosphoric acid–acetonitrile (70:30) produced poor baseline stability. In Trial 2, changing the mobile phase composition improved retention but resulted in interference from an unknown peak. Therefore, a phenyl stationary phase was investigated because both analytes contain multiple aromatic ring systems capable of π – π interactions, which can improve chromatographic selectivity.

Initial trials using 0.1% orthophosphoric acid on the phenyl column produced inadequate peak responses. Subsequently, orthophosphoric acid was replaced with 0.1% formic acid to improve ionisation control and peak symmetry. Progressive optimization of the aqueous-to-organic ratio from 50:50 to 70:30 resulted in improved peak shape, increased theoretical plates, resolution greater than 3.0, and tailing factors below 1.2. Therefore, 0.1% formic acid: acetonitrile (70:30 v/v) on a phenyl column was selected as the optimized chromatographic condition (Table 1).

Table 1: Trials for optimization of chromatographic conditions

Trials	Mobile Phase	Stationary Phase	Reason for Rejection
1	Acetonitrile: 0.1% OPA (70:30)	Acquity UPLC BEH C ₁₈ Column with 130 Å°, 1.7 µm, 210 cm X 100 mm	Baseline is not good
2	Acetonitrile: 0.1% OPA (60:40)	Acquity UPLC BEH C ₁₈ Column with 130 Å°, 1.7 µm, 210 cm X 100 mm	Unknown Peak interference was found
3	Acetonitrile: 0.1% OPA (50:50)	Phenyl 210 cm X 100mm, 1.7 µ	Peak responses are low.
4	0.1% Formic acid: Acetonitrile (50:50)	Phenyl 210 cm X 100mm, 1.7 µ	Second peak tailing is not within the limit.
5	0.1% Formic acid: Acetonitrile (60:40)	Phenyl 210 cm X 100mm, 1.7 µ	resolution between the peaks can be improved
6	0.1% Formic acid: Acetonitrile (70:30)	Phenyl 210 cm X 100mm, 1.7 µ	optimized

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Method validation

Validation of an analytical technique is the process of establishing the validity of an analytical approach. To be more explicit, validating an analytical technique is all about gathering and submitting paperwork that guarantees a factory or business will reliably turn out goods that fulfil strict standards (25,26).

System suitability

System suitability the primary goal of a system suitability analysis is to guarantee that all components of the system (instrument, analyst, chemicals, and electronics) are appropriate for the planned use. After establishing the chromatographic conditions and allowing the mobile phase to equilibrate with the stationary phase, the suggested technique was implemented. Drug chromatograms were obtained after six duplicate injections of system suitability solutions.

Specificity

An experiment was done to determine the level of blank/placebo interference. The chromatograph was then used to record blank and placebo chromatograms after injecting diluent and placebo under the chromatographic conditions.

Linearity

Linearity is indicated by how close the plot of response x concentration becomes a linear graph in shape and form. Solitary observations at different concentrations may be utilized in order to evaluate linearity. Correlation coefficient can be seen in the ensuing plot by looking at the slope and intercept.

Accuracy

A measurement's accuracy can be defined as closely matching the actual value. It is possible to accurately measure a sample's "true value" using a procedure with great precision. Analyte recovery from a blank matrix or insertion of the analyte as per standard practice.

Precision

It is possible to define precision as "level of consistency or disagreement within independent test findings when conducting the technique numerous times on a homogeneous sample." According to ICH, it can be broken down into following distinct sorts: Repeatability, Reproducibility, and Intermediate precision. A method's repeatability is its capacity to perform consistently over a short period. The agreement of entire observations (along with references) after same procedure is repeated multiple times in same lab is referred to as Intermediate Precision. In joint investigations or method transfer trials, reproducibility examines the Precision in different laboratories.

LOD & LOQ

The smallest analyte concentration that can be sensed, but not always measured, is known as the limit of detection (LOD). Under the provided experimental circumstances, the smallest analyte concentration that can be reliably quantified is the limit of quantitation (LOQ).

Robustness

The ICH defines robustness as "Measure of a technique skill to persist unchanged even after tiny but intended modifications in technique parameters." To get the best results, it is best to experiment with the method's most significant parameters and see how they affect separation.

Assay solution preparation

Two tablets were weighed and placed in a mortar. Each tablet (avg. tablet weight 74mg) contained 0.5 mg of Lobeglitazone and 1.0 mg of Glimepiride. The tablets were then crushed into a fine powder and thoroughly mixed. Precisely measured and transferred a sample of 74mg of Lobeglitazone and Glimepiride (as mentioned in the above sample powder) into a 10mL clean volumetric flask, then added the diluent and sonicated it for a duration of 30 minutes to ensure dissolution. Finally, adjust the volume to the desired amount using the same solvent. The sample is subsequently subjected

to filtration using a 0.22-micron injection filter, containing 100ppm of Glimepiride and 50ppm of Lobeglitazone.

Forced degradation studies

An accurately weighed 74 mg sample was subjected to acid (1.0 N HCl, 60°C, 1 h), alkali (1.0 N NaOH, 30 min), oxidation (10% H₂O₂, 30 min), reduction (10% sodium bisulfite, 30 min), hydrolysis (water, 30 min), thermal (60°C, 48 h), and photolytic (1.2 million lux hours and 200 Wh/m² UV, 7 days) stress conditions. Acidic and alkaline samples were neutralised before dilution. Following filtration through a 0.22 µm membrane filter, the samples were analysed, and chromatograms were assessed for degradation behaviour, peak purity, and specificity (27–29).

PDA peak purity analysis demonstrated that the purity angle remained lower than the purity threshold for Glimepiride and Lobeglitazone under all stress conditions, confirming that the principal peaks were spectrally homogeneous and free from co-eluting degradation products. Therefore, the developed RP-UPLC method can be considered stability-indicating. Glimepiride exhibited maximum degradation under oxidative and acidic conditions, indicating the susceptibility of its sulfonylurea linkage toward oxidation and hydrolysis. Lobeglitazone showed similar degradation behaviour owing to oxidative degradation of the thiazolidinedione moiety. Hydrolytic degradation was minimal for both drugs, demonstrating acceptable stability under neutral aqueous conditions.

The acceptance criteria for all analytical method validation parameters were established according to the recommendations of the ICH Q2(R2) guideline and summarised in Table 2.

Table 2. Acceptance criteria for validation of the proposed method as per ICH Q2(R2).

Parameter	Acceptance Criteria
System suitability	%RSD ≤ 2.0%; Tailing factor ≤ 2.0; Resolution ≥ 2.0; Theoretical plates ≥ 2000

Specificity	No interfering peaks; Peak purity passed
Linearity	R ² ≥ 0.999
Accuracy	Recovery 98.0–102.0%
Precision	%RSD ≤ 2.0%
Robustness	%RSD ≤ 2.0% after deliberate changes
Assay	98.0–102.0% of label claim
Forced degradation	5–20% degradation with peak purity passed

Results and Discussion

Method development

Several chromatographic conditions were investigated to optimize the simultaneous determination of Glimepiride and Lobeglitazone. The selected conditions consisted of a phenyl column (2.1 × 100 mm, 1.7 µm) and a mobile phase of 0.1% formic acid (70:30, v/v) delivered at 0.5 mL/min, with UV detection at 266 nm. The optimized method produced sharp, well-resolved peaks within 5 min, with retention times of 1.267 min for Glimepiride and 1.876 min for Lobeglitazone (Table 1; Figure 3).

The phenyl stationary phase was selected because both Glimepiride and Lobeglitazone contain multiple aromatic rings capable of π–π interactions with phenyl ligands. These interactions provided enhanced chromatographic selectivity compared with conventional C18 columns, resulting in improved peak symmetry, higher theoretical plates, reduced peak tailing, and better resolution between the analytes.

Compared with reported RP-HPLC methods, the developed RP-UPLC method offers a shorter run time (5 min), lower solvent consumption, and improved chromatographic efficiency. Most published methods were developed using conventional C18 columns and focused on the quantitation of Glimepiride alone or combined with other antidiabetic drugs, whereas Lobeglitazone has mainly been analyzed using LC-MS/MS for bioanalytical use. In contrast, the present method employs a phenyl

stationary phase and a 0.1% formic acid :acetonitrile as mobile-phase, achieving rapid baseline separation of Glimepiride and Lobeglitazone with retention times of 1.267 and 1.876 min, re-

spectively, while demonstrating excellent sensitivity and stability-indicating capability, making it suitable for routine quality control (Table 3).

Table 3. Comparison of the developed RP-UPLC method with reported chromatographic methods.

Ref.	Drug(s)	Technique	Run Time (min)	Retention Time (min)
Mohd et al., 2014 (12)	Glimepiride	RP-HPLC	10	6.8
Maslarska, 2014 (13)	Glimepiride	HPLC	8	5.6
Kumar et al., 2020 (14)	Glimepiride	RP-HPLC	8	4.5
Kovaříková et al., 2004 (15)	Glimepiride	HPLC	15	9.3
Maruthi & Chandan, 2019 (17)	Glimepiride	RP-UFLC	6	2.9
Lee et al., 2009 (18)	Lobeglitazone	LC-MS/MS	4	2.1
Piyusha & Shailesh, 2023 (19)	Lobeglitazone	LC-MS/MS	3	1.8
Sakuntala et al., 2012 (21)	Glimepiride + Pioglitazone	RP-HPLC	10	3.4, 5.8
El-Enany et al., 2012 (22)	Rosiglitazone + Glimepiride	RP-HPLC	12	4.2, 6.3
Lakshmi et al., 2009 (23)	Pioglitazone + Glimepiride	RP-LC	10	3.6, 6.2
Jain et al., 2008 (24)	Metformin + Pioglitazone + Glimepiride	RP-HPLC	12	2.7, 4.8, 7.6
Present Study	Glimepiride + Lobeglitazone	RP-UPLC	5.0	1.267 (GLM), 1.876 (LBZ)

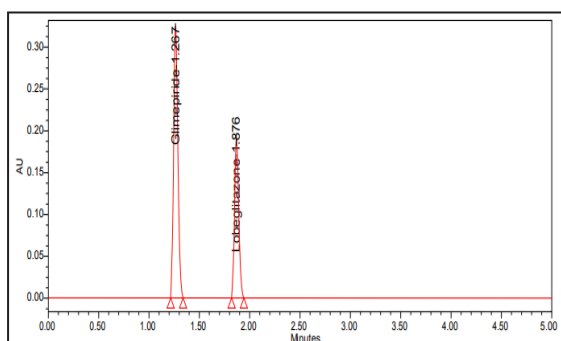


Figure. 3: Optimised chromatogram for Glimepiride and Lobeglitazone with retention times for of 1.267 minutes and 1.876 minutes, respectively

System suitability

The findings of six identical samples that were analyzed are presented in Table 4. Since the percentage RSD values for retention times, plate count, symmetry factor, and resolution were all below 2, the results of the system suitability test were considered acceptable. The low %RSD values (<2%) demonstrate excellent repeatability of the chromatographic system and indicate minimal variability between replicate injections, confirming that the developed RP-UPLC method provides highly reproducible quantitative analysis suitable for routine quality control.

Table 4: System suitability parameters for Glimepiride & Lobeglitazone Sulfate

Parameter	Glimepiride	Lobeglitazone sulfate
Retention time	1.267	1.876
Plate count	7119	5842
Tailing factor	1.05	1.17
Resolution	----	3.34
%RSD	0.19	0.22

Specificity

The chromatogram of the blank solution (Figure.4) has no peak at the RT of the GLM and LBZ sulfate peak. Similarly, the chromatogram of the placebo solution (Figure.5) did not show any peaks during the same retention period as the peak of Glimepiride & Lobeglitazone sulfate.

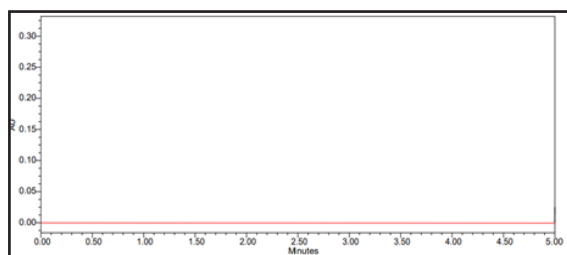


Figure.4: Blank chromatogram shows no significant peaks.

Table 5: Results of linearity for GLM and LBZ

S.no	GLM Standard Level(µg/ml)	Detector Response	LBZ Standard Level (µg/mL)	Detector Response
1	25	587392	12	368402
2	50	1073025	25	679372
3	75	1521134	37.5	1039463
4	100	2084502	50	1381917
5	125	2560044	62.5	1710682
6	150	3043429	75	2029838
Linear Regression	Y=20124.38x+43460.71		Y=27075.93x+14610.93	
Slope	20124.38		27075.93	
Intercept	43460.71		14610.93	
R2	0.99956		0.9998	

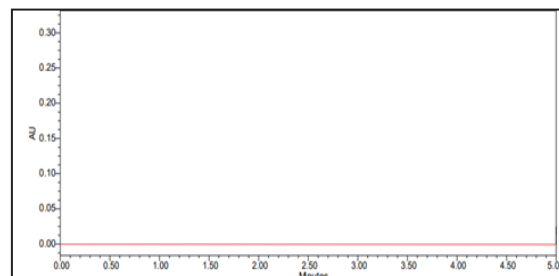


Figure.5: Placebo chromatogram was obtained to confirm the absence of Glimepiride and Lobeglitazone in the excipient matrix

Linearity

Excellent linearity was obtained over the ranges of 25.00–150.00 µg/mL for Glimepiride and 12.50–75.00 µg/mL for Lobeglitazone, yielding R² values of 0.99956 and 0.99980, respectively (Table 5; Figures 6 and 7).

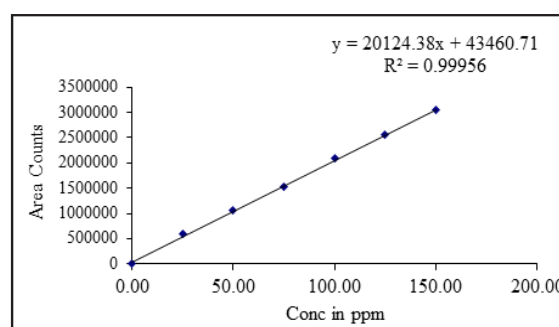


Figure.6: Calibration curve of GLM

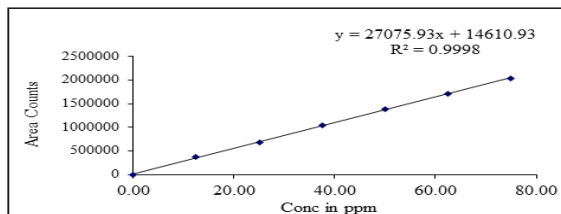


Figure.7: Calibration curve of LBZ

Accuracy/Recovery

The mean recoveries ranged from 99.8–100.3% for Glimepiride and 99.3–100.1% for Lobeglitazone (Table 6), indicating excellent agreement between the measured and added concentrations.

Table 6: Recovery study results GLM & LBZ

%Recovery Level	Glimepiride			Lobeglitazone sulfate		
	Area	% Recovery	Mean Recovery	Area	% Recovery	Mean Recovery
50%	1028425	98.6	99.8	685157	99.0	99.3
	1059366	101.6		688223	99.4	
	1035120	99.3		689428	99.6	
100%	2099981	100.7	100.3	1377281	99.5	100.1
	2084468	100.0		1393552	100.7	
	2092165	100.3		1386210	100.1	
150%	3130258	100.1	100.2	2077149	100.0	99.3
	3153150	100.8		2049485	98.7	
	3115381	99.6		2061122	99.3	

Method precision

The obtained assay values showed mean recoveries of 99.5% for Glimepiride and 99.7% for Lobeglitazone with %RSD values of 0.63% and 0.64%, respectively (Table 7). The low %RSD values indicate excellent repeatability of the analytical procedure and minimal variability between independently prepared samples.

Table 7: Method Precision for GLM & LBZ

S. No.	%Assay for GLM	%Assay for LBZ
1	99.4	100.7
2	98.6	99.8
3	100.0	98.8

4	99.6	100
5	100.3	99.4
6	99.0	99.6
Average	99.5	99.7
Standard Deviation	0.627	0.634
%RSD (n=6)	0.63	0.64

System precision

The %RSD values for peak area responses were 0.295% for Glimepiride and 0.378% for Lobeglitazone (Table 8), which are well below the acceptance limit of 2.0%. These results indicate excellent performance of the chromatographic system with minimal injection-to-injection variability. The low variability confirms the suitability of the UPLC system for consistent and reliable quantitative analysis.

Table 8. System precision results for GLM and LBZ

S. No.	Area for GLM	Area for LBZ
1	2080365	1383425
2	2083230	1377348
3	2088958	1389235
4	2094962	1390541
5	2078124	1385958
6	2086888	1379654
Average	2085421	1384360
Standard Deviation	6153.1	5229.01
%RSD	0.295	0.378

LOD & LOQ

The LOD values of 0.6 µg/mL for Glimepiride and 0.3 µg/mL for Lobeglitazone, with LOQ values of 2.0 µg/mL and 1.0 µg/mL, respectively.

Robustness

Deliberate modifications in flow rate (± 0.05 mL/min) and mobile phase composition ($\pm 2\%$ organic phase) were employed to evaluate method robustness. The %RSD values for both analytes were consistently below 2.0% (Table 9), indicating that these variations did not significantly influence assay performance.

Table 9: Robustness Results of GLM and LBZ

Inj.	Glimepiride				Lobeglitazone			
	Flow Rate (-)	Flow Rate (+)	MP organic (-)	MP organic (+)	Flow Rate (-)	Flow Rate (+)	MP organic (-)	MP organic (+)
1	99.1	99.1	98.4	100.3	99.7	100.6	100.1	99.1
2	100.1	100.4	100.3	100.0	101.1	99.4	99.3	98.7
3	101.5	99.8	101.1	101.5	100.1	101.3	100.7	101.2
Mean	100.2	99.8	99.9	100.6	100.3	100.4	100	99.7
SD	1.206	0.651	1.387	0.794	0.721	0.961	0.702	1.343
%RSD	1.2	0.65	1.39	0.79	0.72	0.96	0.70	1.35

Assay

The assay values obtained were 99.8% for Glimepiride and 100.0% for Lobeglitazone (Table 10), which are within the acceptable

range of 98–102%. These results indicate excellent agreement between the measured drug content and the labelled claim.

Table 10: Assay of GLM and LBZ

Drug	Avg. sample area (n=2)	Std. Conc. (µg/ml)	Sample Conc. (µg/ml)	Label amount (mg)	Std. purity	% assay
Glimepiride	2081796	10	10	1	99.9	99.8
Lobeglitazone sulfate	1385407	5	5	0.5	99.8	100.0

Forced Degradation

Oxidative stress produced the highest degradation for Glimepiride (15.9%) and Lobeglitazone (16.5%), indicating that the sul-

fonylurea functionality of Glimepiride and the thiazolidinedione moiety of Lobeglitazone are particularly susceptible to oxidative degradation. Acidic (14.3% and 15.4%) and alkaline

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(13.1% and 13.5%) stress conditions produced moderate degradation, suggesting partial hydrolysis of chemically labile functional groups. Thermal degradation was comparatively lower (9.0% and 7.1%), indicating acceptable thermal stability during short-term exposure, while photolytic (4.1% and 3.5%) and hydrolytic (2.8% and 1.8%) conditions resulted in minimal degradation, demonstrating good stability under light exposure and neutral aqueous conditions.

Oxidative stress produced the highest degradation for both analytes, indicating that the sulfonylurea functionality of Glimepiride and the thiazolidinedione moiety of Lomeglitazone are particularly susceptible to oxidation. Acid-

ic and alkaline degradation produced moderate degradation, suggesting partial hydrolysis of chemically labile functional groups. Thermal degradation remained comparatively lower, demonstrating acceptable thermal stability during short-term exposure. Minimal degradation under hydrolytic and photolytic conditions indicates that both drugs are relatively stable in aqueous media and under light exposure (see Figures 8 to 14). Under all stress conditions, the purity angle obtained from PDA peak purity analysis was lower than the corresponding purity threshold, demonstrating peak homogeneity, the absence of co-eluting degradants, and the stability-indicating performance of the developed RP-UPLC method.

Table 11: Stress studies of GLM.

Stress studies.	Mean Area Counts	% Degradation	Purity Angle	Purity Threshold
Control	2082968	0.1	0.198	1.717
Acid	1785241	14.3	0.114	1.796
Alkali	1810235	13.1	0.118	1.78
Peroxide	1752031	15.9	0.126	1.732
Reduction	1854276	11.0	0.145	1.711
Thermal	1895645	9.0	0.164	1.712
Photo	1998426	4.1	0.139	1.736
hydrolysis	2025147	2.8	0.136	1.746

Table 12: studies of LBZ.

Stress studies	Mean Area Counts	% Degradation	Purity Angle	Purity Threshold
Control	1384217	0.1	0.952	3.331
Acid	1171117	15.4	0.963	3.338
Alkali	1196598	13.5	0.957	3.335
Peroxide	1155478	16.5	0.955	3.321
Reduction	1246052	10	0.937	3.318
Thermal	1286052	7.1	0.921	3.366
Photo	1336052	3.5	0.949	3.347
hydrolysis	1358695	1.8	0.954	3.321

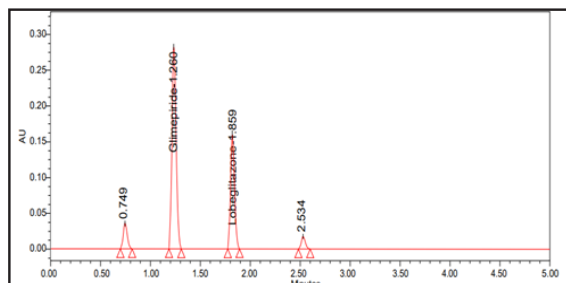


Figure 8: Acidic Degradation chromatogram with about 15.4% of the analytes degraded.

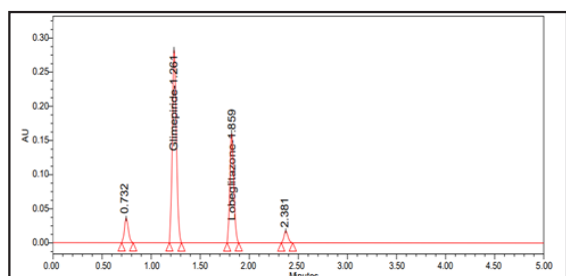


Figure 9: Alkali Degradation chromatogram with about 13.5% degradation, indicating susceptibility to basic conditions.

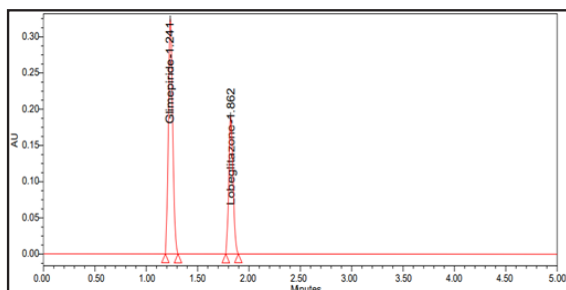


Figure 10: Hydrolysis Degradation chromatogram with about 1.8% degradation

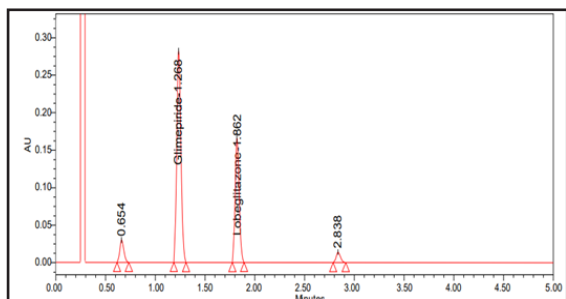


Figure 11: Peroxide Degradation chromatogram, the highest degradation was observed under peroxide stress, with 16.5% degradation.

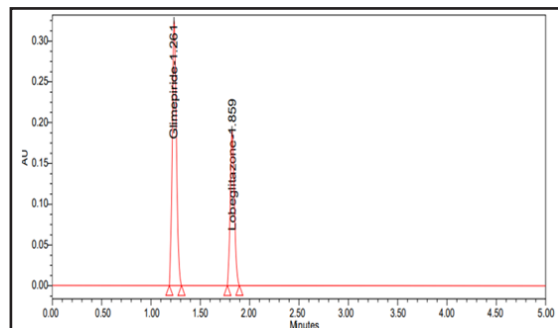


Figure 12: Photo Degradation chromatogram, Photolytic stress resulted in 3.5% degradation, highlighting a moderate impact from light exposure.

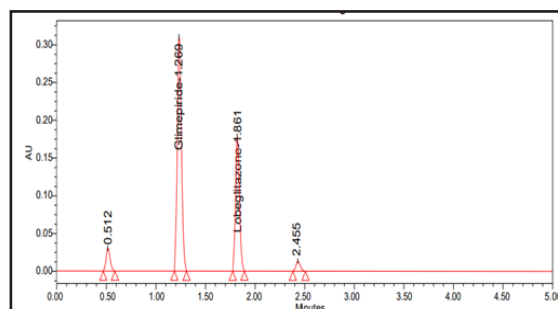


Figure 13: Reduction Degradation chromatogram, with about 10% degradation, indicating vulnerability to reducing agents.

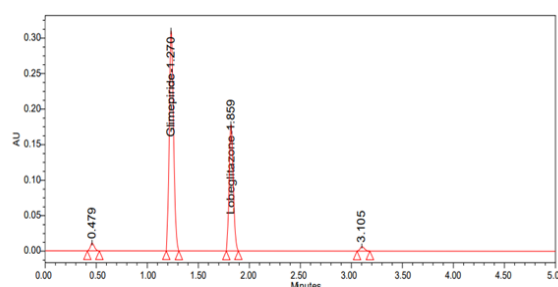


Figure 14: Thermal Degradation chromatogram, with about 7.1% degradation, demonstrating the impact of elevated temperatures.

Conclusion

A simple and stability-indicating RP-UP-LC method was successfully developed for the simultaneous determination of Glimepiride and Lobeglitazone sulfate. Chromatographic separation was achieved within 5 min, providing excellent peak symmetry, satisfactory resolu-

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tion, and high chromatographic efficiency. The method exhibited excellent chromatographic performance and fulfilled all ICH Q2(R2) validation criteria, demonstrating specificity, linearity, accuracy, precision, robustness, and sensitivity. Its ability to separate degradation products confirms its applicability for routine quality control and stability studies.

Compared with previously reported methods, the proposed RP-UPLC method provides shorter run time, lower solvent consumption, and improved efficiency for the simultaneous estimation of Glimepiride and Lomeglitazone sulfate. It is suitable for routine quality control, stability studies, and batch-release testing, with potential application in impurity profiling and LC-MS-based analysis of pharmaceutical formulations in industrial quality control and regulatory laboratories. Future studies may further extend the applicability of the method to impurity profiling and transfer to LC-MS-compatible analytical platforms for comprehensive pharmaceutical analysis.

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Author contributions

Anoop Bodapati: Conceptualization, Investigation, Methodology, Validation, Writing-original draft. **Pagala Bangaraiah:** Conceptualization, Supervision, Resources, Writing-review & editing.

Conflict of interest

The authors declare no conflict of interest.

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