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Original Research Article

Development and Validation of an LC-ESI-MS/MS Method for Metformin Hydrochloride and Its Application in the Pharmacokinetic Evaluation of Gastroretentive Floating Granules in Wistar Rats

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Abstract: ***Background:*** Metformin has low absorption in the gastrointestinal tract, which means it needs to be taken numerous times a day and has led to the creation of gastroretentive delivery methods. The goal of this study was to create and test a sensitive LC-ESI-MS/MS method for measuring metformin in rat plasma and then use that technology to look at how a new floating granule formulation works in the body. ***Methods:*** A robust LC-ESI-MS/MS method was optimized using a Phenomenex Kinetex C18 column with a mobile phase of 0.1% formic acid and ammonium acetate buffer. The method exhibited exceptional specificity, linearity (31.25–4000 ng/mL), precision, accuracy, recovery, and stability. In vivo assessment was conducted on Wistar rats (n = 12) receiving metformin floating granules (MET-GR) or an oral metformin solution (MET-SOL) at a dosage of 500 mg/kg. Plasma concentrations were measured using an approved method, and pharmacokinetic characteristics were evaluated by non-compartmental analysis. In vivo edema and stomach retention were subsequently assessed in rats utilizing X-ray imaging. ***Results:*** The validated technique demonstrated significant sensitivity (LOD 0.01 ng/mL; LLOQ 31.25 ng/mL) and displayed no interference during the analyte retention durations. Pharmacokinetic studies demonstrated that MET-GR exhibited significantly elevated AUC_{0-t} (91716.72 ± 8828.20 ng·h/mL vs. 65201.98 ± 2339.82 ng·h/mL) and $AUC_{0-\infty}$ values compared to MET-SOL ($p < 0.0001$), in addition to a prolonged half-life (12.79 ± 2.10 h vs. 8.67 ± 0.55 h). The Cmax was reduced, while the Tmax was notably postponed for MET-GR, signifying prolonged absorption. X-ray radiography verified stomach retention and the gradual enlargement of the granules for a minimum of 60 minutes. ***Conclusion:*** The established LC-ESI-MS/MS technique is extremely dependable for quantifying metformin levels in rat plasma. The gastroretentive floating granules exhibited persistent absorption, enhanced systemic exposure, and an extended half-life relative to an oral solution, indicating their potential as a superior controlled-release formulation. Additional pharmacodynamic and clinical investigations are necessary.

Keywords: Metformin; Gastroretentive drug delivery; Floating granules; LC-ESI-MS/MS; Bioanalytical method validation; Pharmacokinetics; In vivo swelling; Wistar rats.

INTRODUCTION

Metformin is the first-line drug treatment for type 2 diabetes mellitus because it is safe, effective, and cheap. Metformin is widely used, however it has low and variable oral bioavailability (40–60%). This is mainly because it is hydrophilic, doesn't pass through the intestines easily, and only absorbs in certain areas, mostly the upper gastrointestinal system [1, 2]. Additionally, the quick movement of typical immediate-release formulations through the digestive system sometimes means that the drug doesn't stay in its best absorption window for long enough, which means

that the therapeutic levels are not as high as they should be and the patient needs to take the drug numerous times a day. These restrictions have led to a lot of research into gastroretentive and controlled-release delivery methods that are meant to improve metformin absorption by keeping it in the stomach longer and allowing the medicine to be released over time [3, 4]. Floating drug delivery systems (FDDS) have garnered considerable interest as an efficient gastroretentive approach, owing to their capacity to remain suspended in the stomach for prolonged durations, hence enhancing the bioavailability and therapeutic effectiveness of medicines characterized by limited absorption windows. Floating granules, in example, provide benefits including being easy to give, allowing for more flexible dosing, and making patients more likely to follow directions. To evaluate the in vivo efficacy of these systems, real-time imaging modalities, such as X-ray radiography, are commonly utilized to observe stomach retention and monitor swelling or breakdown patterns [5, 6].
For pharmacokinetic testing of new formulations, it is important to be able to accurately measure metformin in biological samples. Liquid chromatography–electrospray ionization tandem mass spectrometry (LC-ESI-MS/MS) is the best way to analyze biological samples since it is more sensitive, specific, reproducible, and works well for high-throughput analysis. It is important to have a validated analytical approach in order to make sure that plasma metformin levels are measured accurately during pharmacokinetic studies. This study developed and validated a sensitive, specific, and reproducible LC-ESI-MS/MS method for quantifying metformin in rat plasma, utilizing Clopidogrel Carboxylic Acid as an internal standard to ensure consistent and accurate detection across a broad concentration range [7, 8].
The validated approach was then used to study the pharmacokinetic profile of a new gastroretentive floating granule formulation of metformin in Wistar rats, comparing it to a regular oral metformin solution. Moreover, the in vivo swelling and gastric retention characteristics of the floating granules were recorded by X-ray imaging to ascertain a relationship between formulation efficacy and pharmacokinetic results. This combined analytical and in vivo assessment gives us a full picture of how well the gastroretentive floating system works and how it might improve the effects of metformin.

2. Method Development

2.1 Chromatographic Conditions

The chromatographic method was fine-tuned in a methodical fashion to achieve optimal sensitivity for metformin detection in rat plasma, with sharp peak morphologies, sufficient retention, low matrix interference, and so on. A Phenomenex Kinetex C18 column, which is ideal for fast LC-MS/MS analysis of very polar analytes like metformin, was chosen for the separation process because of its efficiency, stability, and dimensions (50 × 3 mm, 5 µm). Two solvents, A and B, made up the mobile phase. Solvent A was a mixture of Milli-Q water with 10 mM ammonium acetate and 0.1% formic acid, and Solvent B was acetonitrile with 0.1% formic acid. In positive ESI mode, this combination guaranteed a predictable analyte response and produced good ionization efficiency [9, 10].

The chromatographic resolution and MS compatibility were balanced by maintaining a flow rate of 0.5 mL/min, and the injection volume was tuned to 10 µL to avoid column overloading and keep sensitivity. Complete elution of metformin and the internal standard without carryover was achieved in the allotted
7 minutes of run time, allowing for quick sample throughput. A positive electrospray ionization mode LC-MS/MS system was used for detection. This mode produced stable ion currents that were strong enough to reliably quantify the target analyte across the calibration range. For optimal retention, sensitivity, and repeatability, these chromatographic parameters were fine-tuned by testing various column chemistries, mobile phase mixes, and organic phase gradients [10, 11].

2.2 Mass Spectrometric Conditions

We used a tandem mass spectrometer that was set up with a Turbo Spray ionization source and ran in the positive electrospray ionization (ESI+) mode to detect and quantify metformin. The reason this approach was chosen is because metformin, being both hydrophilic and extremely basic, easily produces stable protonated ions when exposed to positive electrostatic interactions (ESI). To maximize the analyte response, improve the signal-to-noise ratio, and minimize matrix suppression, the instrument parameters were fine-tuned. For detection that was both sensitive and selective, multiple reaction monitoring (MRM) was used. The transition from precursor to product ions, m/z 130.0 → 71.0, was tracked for metformin with optimum voltages such as 40 V for declustering, 33 V for collision energy, and 15 V for collision cell exit potential [12, 13]. Under these conditions, the product ion signal was robust and steady, and the fragmentation was consistent. The internal standard (IS) employed to account for variations in extraction efficiency, ionization, and instrument performance was clopidogrel carboxylic acid. The parameters were tuned to ensure stable ionization and reproducible peak response, with the MRM transition set at m/z 308.1 → 198.0, DP at 20 V, CE at 26 V, and CXP at 15 V. The approach is well- suited for pharmacokinetic applications due to the designed mass spectrometric settings, which provide high selectivity, minimal background noise, and robust measurement of metformin in rat plasma [13, 14].

3. Sample Preparation

The isolation of metformin from rat plasma was accomplished using a liquid-liquid extraction (LLE)

technique because it is simple, reproducible, and well-suited for high-throughput bioanalytical procedures. Maximizing analyte recovery while minimizing matrix interference and ensuring acceptable compatibility with LC-ESI-MS/MS analysis were the goals of the optimization process. At a concentration of 100 ng/mL, the internal standard (IS), Clopidogrel Carboxylic Acid, was added to 100 µL of rat plasma that had been placed into a clean polypropylene tube for each sample. To make sure the IS was distributed evenly throughout the plasma matrix, the mixture was vortex-mixed for 1 minute. Proteins were precipitated and the analyte was made easier to partition into the organic phase by adding 1 mL of acetonitrile as the extraction solvent. Centrifugation at 12,000 rpm for 10 minutes was used to accomplish full phase separation after the samples were vortexed for an extra 5 minutes to improve extraction efficiency [14, 15].
To remove the organic solvent without degrading the analyte by heat, the transparent supernatant was delicately transferred and evaporated to dryness under a mild nitrogen stream at 40-45°C. After drying, the residue was mixed with 500 µL of a diluent that was chosen for its best solubility and chromatographic performance. The diluent was a 50:50 (v/v) ratio of acetonitrile and water. The last step was to inject the reconstituted samples into autosampler vials using low-volume inserts for LC-MS/MS. Reliable quantification of metformin across the calibration range was supported by clean samples yielded by this extraction protocol, which had high recovery and minimal matrix effects [16].

4. Calibration and QC Samples

Calibration standards and quality control (QC) samples were created to make sure that the amount of metformin in rat plasma could be measured accurately, precisely, and consistently across the range of concentrations that are usually found in pharmacokinetic investigations. We chose the range for the calibration curve (31.25–4000 ng/mL) based on early pharmacokinetic data and the method's sensitivity needs. There were eight non-zero calibration points at concentrations of 31.25, 62.5, 125, 250, 500, 1000, 2000, and 4000 ng/mL. This gave us enough data density for strong linear regression analysis. To make each calibration standard, blank rat plasma was spiked with the right amounts of metformin working solutions. Then, the samples were processed in the same way as the study samples. We used a weighted ($1/x^2$) linear regression model to plot the peak area ratio of metformin to the internal standard against the nominal concentration to see how linear the calibration curve was [17, 18].
Four different concentrations of quality control (QC) samples were made to test the method's precision, accuracy, recovery, and stability. The lower limit of quantification (LLOQ) was 31.25 ng/mL, the low QC (LQC) was 93.75 ng/mL, the medium QC (MQC) was 1500 ng/mL, and the high QC (HQC) was 3000 ng/mL. The QC levels were carefully chosen to represent the lowest, middle, and upper parts of the calibration range. This made sure that the analytical method was fully tested. All calibration and QC samples were divided into smaller parts and kept in controlled conditions until they could be analyzed. This calibration system gave accurate measurements of metformin over a wide range of values, making it useful for pharmacokinetic testing [18].

5. Method Validation

The developed LC-ESI-MS/MS technique was verified in accordance with current regulatory criteria to verify reliability, repeatability, and appropriateness for the quantification of metformin in rat plasma. The validation metrics assessed comprised specificity, sensitivity, linearity, precision, accuracy, recovery, stability, and matrix effects [19].

5.1 Specificity and Selectivity

Specificity was evaluated by examining blank rat plasma samples from various sources to examine potential endogenous interference at the retention durations of metformin and the internal standard (Clopidogrel Carboxylic Acid). There were no significant interference peaks in any of the plasma lots that were tested. This shows that the approach is very selective and may be used to accurately measure the analyte in biological matrices (figure 1) [19, 20].

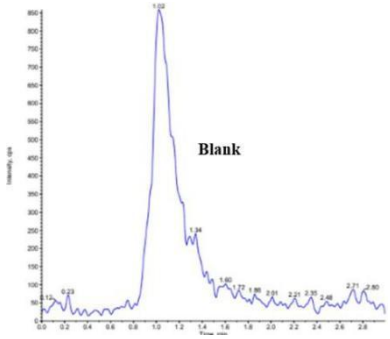

Figure 1A. Blank chromatogram

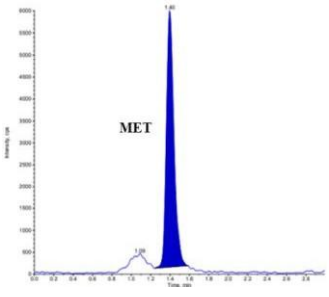

Figure 1B. Metformin chromatogram

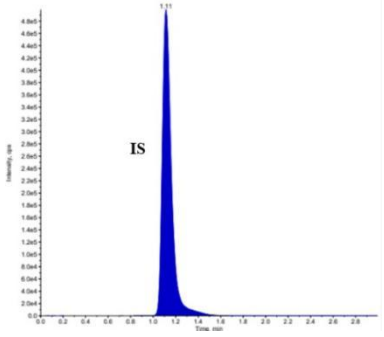

Figure 1C. Internal standard chromatogram

Figure 1: Combined LC-ESI-MS/MS chromatogram panel including (1A) blank plasma showing no interference, (1B) metformin peak at ~1.40 min, and (1C) internal standard peak at ~1.11 min.

5.2 Sensitivity

The approach showed a lot of analytical sensitivity. The limit of detection (LOD) was 0.01 ng/mL, which means that the instrument was very responsive. The lower limit of quantification (LLOQ) was set at 31.25 ng/mL. This is the lowest concentration that can be measured with acceptable precision (%CV ≤ 20%) and accuracy (within ±20%). The chromatographic results at the LLOQ exhibited a distinct and consistent signal-to-noise ratio, validating the method's efficacy for low-level detection of metformin in pharmacokinetic studies [19, 20].

5.3 Linearity

Eight non-zero calibration points were used to check the linearity over the calibration range of 31.25 to 4000 ng/mL. The approach showed a very strong linear relationship, with regression coefficients (r^2) always being higher than 0.99. All of the back-calculated concentrations for the calibration levels met the acceptance standards. The %CV values were below 10.6%, and the accuracy ranged from 92.05% to 106.27%. These results show that the approach is strong and works well across the whole analytical range (Table 1).

Table 1: Linearity data for metformin across the calibration range

Nominal (ng/mL)	Mean Back-Calculated (ng/mL)	SD	%CV	% Accuracy
31.25	30.12	1.02	3.39	96.43
62.5	63.85	2.75	4.31	102.16
125	120.44	8.10	6.72	96.35
250	265.67	15.22	5.73	106.27
500	510.22	30.12	5.91	102.04
1000	920.50	50.30	5.47	92.05
2000	1985.70	210.55	10.60	99.29
4000	3702.85	100.25	2.71	92.57

5.4 Precision and Accuracy

Intra-day precision and accuracy were assessed utilizing quality control samples at four concentration tiers: LLOQ, LQC, MQC, and HQC. All values fell within acceptable parameters, exhibiting a %CV below 10% and an accuracy of ±15%. The results are presented below Table 2. These results illustrate the method's precision and consistency for bioanalytical quantification.

Table 2: Intra-day precision and accuracy of metformin at QC levels

Sr. no.	QC Level	Nominal (ng/mL)	Mean (ng/mL)	SD	%CV	% Accuracy
1	LLOQ	31.25	32.45	2.35	7.24	103.84
2	LQC	93.75	95.88	5.28	5.51	102.27
3	MQC	1500	1465.52	135.20	9.22	97.70
4	HQC	3000	2855.47	175.42	6.14	95.18

5.5 Recovery

Absolute recovery was evaluated at low quality control (LQC), medium quality control (MQC), and high quality control (HQC) levels by comparing extracted samples to post-extraction spiked standards. The recovery values were consistent and reproducible across various concentrations. The elevated and consistent recovery validates the efficacy of the liquid–liquid extraction method (Table 3).

Table 3: Recovery of metformin at different QC levels.

Sr. no.	QC Level	% Recovery
1	LQC	93.75
2	MQC	100.92
3	HQC	98.10

5.6 Stability

The stability of metformin was assessed under diverse settings to replicate sample handling and storage situations. The analyte exhibited stability with negligible degradation, as seen by the subsequent data (Table 4). All data fell within ±15% of nominal concentrations, so affirming the stability of metformin under all evaluated settings.

Sr. no.	Stability Study	% Stability
1	Freeze-Thaw (3 cycles)	98.24
2	Short-Term (24 h)	96.42
3	Long-Term (30 days, −80°C)	97.50
4	Autosampler (24 h)	99.30

5.7 Matrix Effect

Matrix effects were assessed to determine ion suppression or amplification caused by plasma components. The results indicated negligible matrix interference, with values presented in Table 5. The minimal matrix effect values signify exceptional technique resilience and uniform ionization efficiency.

Table 5: Matrix effect evaluation at QC levels.

Sr. No.	QC Level	% Matrix Effect
1	LQC	12.45
2	MQC	9.78
3	HQC	5.32

The approach exhibited outstanding specificity, linearity, accuracy, precision, and stability for quantifying Metformin in rat plasma. The recovery was uniform, and the matrix effects remained within permissible ranges. This established approach is appropriate for pharmacokinetic investigations of Metformin.

3.0 Pharmacokinetic Study:

A pharmacokinetic study was conducted to assess the efficacy of a novel gastroretentive floating granule formulation of metformin in comparison to an oral metformin solution, acknowledging that metformin demonstrates unique pharmacokinetic properties, including flip-flop absorption and substantial gastrointestinal interactions that affect its therapeutic response. Because the time spent in the gastrointestinal tract and the rate of absorption are so important for metformin's pharmacodynamic effects, this study looked at the plasma pharmacokinetics of both formulations after a single oral dose of 500 mg/kg in male Wistar rats. The goals were to compare how well they were absorbed, looking at things like the maximum plasma concentration (Cmax), the time it took to reach peak concentration (Tmax), the overall systemic exposure (AUC), and how well the floating granules kept plasma levels up compared to the regular solution [20-22].

3.1 Study Design

A single-dose, parallel-group, comparative pharmacokinetic study was performed to assess and compare the systemic exposure profiles of a novel gastroretentive floating granule formulation of metformin with a standard oral metformin solution. The study involved 12 healthy male Wistar rats, each weighing between 180 and 220 grams. The animals were randomly divided into two groups, each with six rats (n = 6), to make sure the comparison was fair and the statistical power was strong enough. Both groups got the same amount of metformin, 500 mg/kg, by mouth. Group I received metformin floating granules that were made to stay in the stomach longer and release the drug slowly. Group II received the regular oral metformin solution, which is the immediate-release reference formulation. All doses were given through oral gavage to make sure they were given correctly and could be repeated. Table 6 below gives a summary of the whole study design:

Table 6: Treatment groups and dosing details for the pharmacokinetic study

Group	Treatment	Dose (mg/kg)	Route
I	Metformin Floating Granules	500	Oral
II	Oral Metformin Solution	500	Oral

3.2 Dose Preparation

The test formulation, which was made with the right polymers to lower density and keep the metformin in the stomach for a longer time, was made using gastroretentive floating granules. Before giving the granules, they were evenly suspended in a 0.5% w/v carboxymethylcellulose (CMC) solution to make dosing and delivery easier. To make the reference formulation, an oral metformin solution, metformin hydrochloride was dissolved in purified water while being stirred gently until a clear, homogeneous solution was formed. To keep the drugs stable and make sure the right amount of medicine was in each dose, both formulations were made fresh on the day of dosing [22, 23].

3.3 Administration

To make sure that the dose was accurate and could be repeated, the animals got the allotted formulation by mouth through a calibrated stainless-steel feeding needle. The dose volume given to each rat was based on its weight, making sure that each rat got the same amount of metformin, which was 500 mg/kg. All animals were deprived of food overnight

while having unrestricted access to water to reduce variability in gastrointestinal transit and absorption.

3.4 Blood Collection

Blood samples were taken at set times to get a full picture of how metformin works in the body. Samples were taken at 0 (before the dose), 0.25, 0.5, 1, 2, 4, 6, 8, 12, 24, 48, and 72 hours after the dose. Blood was taken from the retro -orbital plexus while the patient was lightly sedated with isoflurane (3% induction, 1.5–2% maintenance) to reduce stress and speed up the process. The blood that was collected was put into tubes with EDTA on them and spun at 4000 rpm for 10 minutes at 4°C to separate the plasma. To keep metformin from breaking down, the plasma samples were refrigerated at −80°C right away until they could be tested.

3.5 Bioanalytical Method

We used the validated LC-ESI-MS/MS technology we talked about earlier to find out how much metformin was in the plasma. We used the standard liquid–liquid extraction method to process all samples, calibration standards, and quality control samples. We also analyzed them under the best chromatographic and mass spectrometric conditions to make sure we got the right amounts over the pharmacokinetic range [23, 24].

3.6 *In-Vivo* Drug Release Study

The in vivo plasma concentration–time profiles of metformin were assessed for both the metformin solution (MET-SOL) and the gastroretentive floating granules (MET-GR) after oral delivery. Blood samples obtained within 72 hours post-administration were analyzed utilizing the validated LC-ESI-MS/MS technique. Table X displays the mean plasma concentrations and standard deviations at each time point. MET-SOL demonstrated a rapid absorption phase, with a pronounced increase in plasma concentration, attaining peak values within 1–2 hours. Conversely, MET-GR exhibited a slower, more gradual absorption characterized by lower starting concentrations yet maintained plasma levels over an extended duration. The MET-GR formulation sustained elevated concentrations beyond 6 hours and demonstrated extended drug exposure for up to 72 hours, signifying excellent controlled release and improved stomach retention. The mean plasma concentration–time curve (Figure 2) further exemplifies these tendencies, revealing a quick peak followed by a sharp decrease for MET-SOL, whereas MET-GR exhibits a flattened, prolonged profile indicative of enhanced in vivo performance [25, 26].

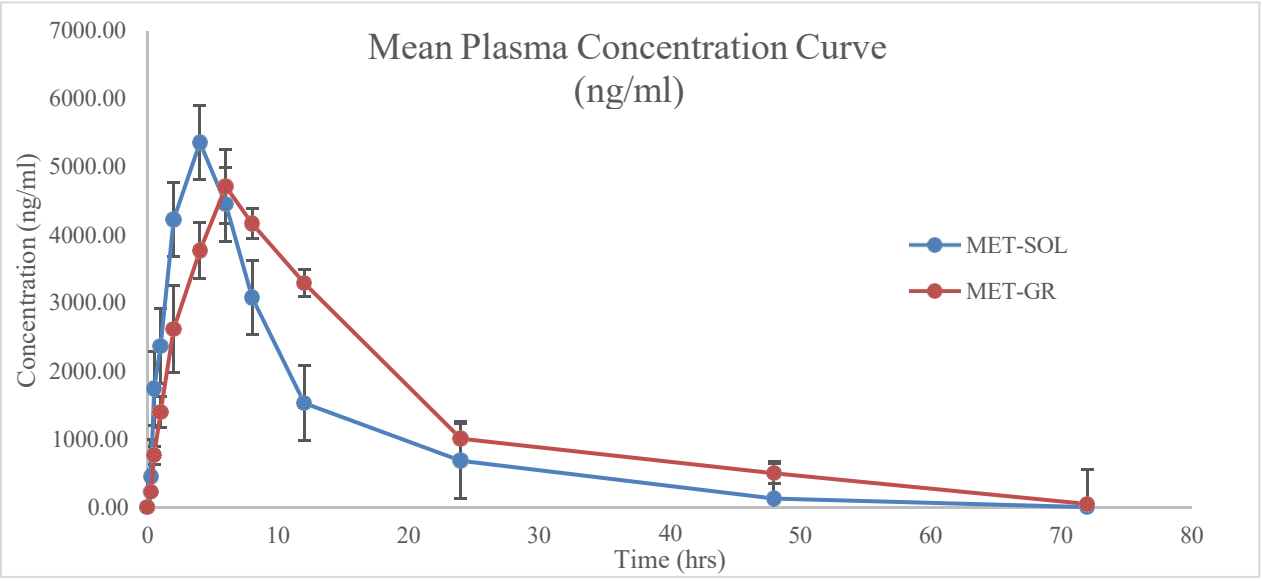

Figure 2: Mean plasma concentration–time profiles of metformin following oral administration of metformin solution (MET-SOL) and gastroretentive floating granules (MET-GR) in Wistar rats (n = 6 per group). The graph shows rapid absorption and early peak concentration for MET-SOL, whereas MET-GR exhibits slower absorption with sustained plasma levels and prolonged drug exposure up to 72 hours.

4. Pharmacokinetic Analysis

Pharmacokinetic assessment revealed significant disparities in the absorption and distribution of metformin between the two formulations. The MET-SOL group had a faster Tmax, which meant that it was absorbed faster. The MET-GR formulation, on the other hand, had a much slower Tmax, which demonstrated that it was designed to be released slowly and stay in the stomach longer. The Cmax of MET-GR was lower than that of MET-SOL, but the floating granules' ability to release the medication over time led to much greater $AUC_{0–t}$ and $AUC_{0–\infty}$ values. This means that the drug was more available and had a longer effect on the body. The elimination rate constant (Kel) was much lower, and the

half-life (t½) was significantly longer for MET-GR. This further supports the idea that metformin takes longer to leave the body and stays in the bloodstream longer. These findings indicate that the gastroretentive floating granule formulation significantly extends absorption, improves bioavailability, and sustains plasma concentrations for an extended period relative to the standard oral solution [27, 28].

Table 7: Comparison of pharmacokinetic parameters between MET-SOL and MET-GR.

PK-Parameters	MET-SOL		MET-GR		p-value
	Mean	SD	Mean	SD	
Cmax	5582.215	627.3503	4828.395	314.4304	0.012553*
T max	4.333333	1.505545	6.333333	0.816497	0.008473*
Auc 0-t	65201.98	2339.816	91716.72	8828.197	0.000016*
Auc 0-a	65696.37	2730.062	95722.47	6979.074	< .00001**
Kel	0.080215	0.005177	0.055313	0.00843	0.000053*
T 1/2	8.668937	0.552377	12.79392	2.099929	0.000452*

*Significant at $p < 0.05$; **Highly significant.*

RESULT AND DISCUSSION

The comparative pharmacokinetic evaluation of metformin gastroretentive granules (MET-GR) and the oral metformin solution (MET-SOL) in Wistar rats demonstrated considerable formulation-dependent disparities in absorption, systemic exposure, and elimination. The Cmax of MET-SOL (5582.22 ± 627.35 ng/mL) was much greater than that of MET-GR (4828.39 ± 314.43 ng/mL; p = 0.0126). This means that the solution formulation gave the drug quick and strong systemic exposure right after it was given. This result is anticipated, as pre-dissolved medication in solution form circumvents the dissolving phase, facilitating expedited absorption via the gastrointestinal mucosa [28, 29].

The Tmax, on the other hand, was much longer for MET-GR (6.33 ± 0.82 h) than for MET-SOL (4.33 ± 1.51 h; p = 0.0085). This shows that the granule formulation absorbed more slowly. This delayed Tmax is in line with the controlled-release properties of floating granules, which slowly absorb water, expand, and break down in the stomach. This lengthens the time it takes for metformin to be released and absorbed in its main absorption window [30].

Even though MET-GR took longer to be absorbed, it had a far higher overall systemic exposure. The AUC_0–t (91716.72 ± 8828.20 ng·h/mL) and AUC_0–∞ (95722.47 ± 6979.07 ng·h/mL) values for MET-GR were both much greater than those for MET-SOL (65201.98 ± 2339.82 ng·h/mL and 65696.37 ± 2730.06 ng·h/mL; $p < 0.0001$ for both). These results indicate increased bioavailability from the granule formulation, presumably attributable to higher stomach retention, extended residence time in the upper gastrointestinal tract, and more thorough absorption over a protracted period [31].

Elimination measurements also corroborated the sustained-release properties of MET-GR. The elimination rate constant (Kel) for MET-GR (0.0553 ± 0.00843 h^{-1}) was much lower than for MET-SOL (0.0802 ± 0.00518 h^{-1}; p = 0.000053). This meant that MET-GR had a much longer half-life (t½) of 12.79 ± 2.10 h compared to MET-SOL's 8.67 ± 0.55 h (p = 0.000452). This prolonged half-life implies extended systemic persistence of metformin when taken in the granule form, validating the controlled-release profile of the formulation [32].

These pharmacokinetic results show that the metformin solution reaches high plasma concentrations quickly, but the gastroretentive granule formulation gives the body more exposure to the medication, keeps it in the body longer, and absorbs it more slowly. These pharmacokinetic benefits may help with glycemic control by keeping therapeutic plasma levels higher for longer periods of time and lowering the number of doses needed, which may help patients stick to their diabetes treatment plan over the long term. The observed enhancements in pharmacokinetic behavior highlight the possibility of the floating granule formulation as an efficacious controlled-release oral dose form of metformin [32, 33].

CONCLUSION

The LC-ESI-MS/MS method we developed was very good at measuring metformin in rat plasma because it was very specific, linear, accurate, precise, and stable. The method's robustness and dependability were further proven by consistent recovery values and low matrix effects, which showed that it was suitable for pharmacokinetic applications. Using this proven method, the pharmacokinetic study showed that the gastroretentive floating granule formulation of metformin was much better than the regular oral

solution. The floating granules showed delayed absorption, a much higher systemic exposure, and a longer elimination half-life. This means that they worked well for controlled-release and stayed in the stomach longer. These characteristics indicate that the floating granule formulation may enhance treatment efficacy, mitigate peak-associated side effects, and decrease dose frequency, thereby improving patient adherence. Additional pharmacodynamic research and clinical trials are advised to validate these results and confirm the clinical advantages of metformin gastroretentive floating granules.

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Conflict of Interest:
None

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