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Development and Validation of a Novel and Robust RP-HPLC Method for the Simultaneous Estimation of Metformin and Sesamol, and its Application in an In-house Developed Nanoliposomal Formulation

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ABSTRACT

This study introduces a unique High-Performance Liquid Chromatography (HPLC) method for quantifying Metformin and Sesamol simultaneously in pharmaceuticals. The technique uses a reverse-phase C18 column with acetonitrile and water in a gradient pattern with UV detection. The method demonstrated good linearity for Metformin and Sesamol at concentrations ranging from 20-1.25 µg/mL, with coefficients of correlation (r^2) of 0.9947 and 0.9908, respectively. The method demonstrated excellent repeatability, with precision measured as RSD below 2% for intra-day and inter-day measurements. The method's accuracy was proven through recovery tests. The Metformin's LOD and LOQ reported as 0.89 µg/mL & 2.71 µg/mL, and Sesamol's as 1.27 µg/mL & 3.86 µg/mL, respectively. The approach also demonstrated resilience to minor fluctuations in method parameters, making it suitable for regular analysis. This validated HPLC technique was successfully used for the simultaneous measurement of Metformin and Sesamol in a nanoliposomal formulation, providing a reliable tool for quality control.

Keywords: Metformin, Sesamol, Medicine, High-Performance Liquid Chromatography, Simultaneous Estimation, Liposomes.

INTRODUCTION

Since the beginning of time, the pharmaceutical industry has been looking for reliable and efficient analytical methods to guarantee the high quality and effectiveness of the pharmaceuticals that they manufacture. HPLC method stands out as especially significant among these techniques since it has exceptional reproducibility, sensitivity, and resolution[1]. Metformin is an antihyperglycemic medicine that belongs to the Biguanide family. Type 2 diabetes mellitus(T2DM) treatment is commonly used for this medication[2-3]. Its primary functions consist glucose amount reduction that is produced by the liver and increasing the insulin sensitivity of tissues in the periphery[4]. The findings of several studies, however, indicate that metformin may possess anticancer characteristics via influencing several different pathways. mTOR & AMPK (AMP-activated protein kinase) signalling pathways

reported to be mechanisms via which metformin has its impact on the process of protein synthesis. Metformin inhibits the mitochondrial respiratory chain complex on tumor cells, which in turn activates the AMPK pathway, which is involved in the process of protein synthesis and cell division[5-6]. The activation of the AMPK pathway results in the production of glycolysis, fatty acid oxidation, along with fatty acid inhibition & protein production[7]. One of the primary pathways that human breast tumors use to spread is via an increase in the synthesis of proteins that are reliant on mTOR[8–9]. Metformin may potentially have anticancer effects since it lowers glycemia and insulin resistance. This will result in insulin & IGF-1(insulin-like growth factor 1) levels declining, and it may also prevent the spread of cancer cells[10-12]. It has been shown that growth factors nd hormones, such as insulin, may trigger the phosphatidylinositol 3-kinase signaling pathway[8], which in turn promotes the development of cancer. Metformin inhibits cancer cell growth *in vivo* & *in vitro*. This is accomplished by influencing a wide variety of pathways. The stability and quantification of metformin in various formulations need precise analytical procedures to ensure consistent treatment efficacy and patient safety[13–15]. Metformin is extensively utilized, but its stability and quantification in diverse formulations must be determined. Sesamol, a naturally occurring phenolic antioxidant that can be found in sesame seeds and oil[16–17], has garnered attention due to several pharmacological properties that it has, such as anti-inflammatory, anti-cancer, and neuroprotective activity[18–19]. Recent studies have shown that it has the potential to enhance the therapeutic efficacy of a variety of drugs; hence, it is imperative that reliable analytical methods for its detection in combination formulations be established. A synergistic approach to managing diabetes and the oxidative damage that is associated with it may be achieved via the use of therapeutic regimens that include sesamol and metformin. As a result, developing a suitable HPLC technique for simultaneous measurement of these substances is of fairly critical importance[20–21]. Not only would this strategy make it possible to conduct the pharmacokinetic and pharmacodynamic research that is necessary for their clinical validation, but it would also make it possible to conduct quality control examinations on mixed formulations[22-23] Designing and verifying analytical techniques for simultaneous pharmaceutical component estimation is of crucial importance in process of ensuring safety, quality, & pharmacological formulations efficacy. Many pharmaceutical companies employ HPLC analysis[24] given it's accurate, precise, & has ability to separate compound combinations that are complex. A number of analytical challenges are brought about by the simultaneous estimation of metformin and sesamol due to the fact that these two substances have distinct chemical properties. In contrast to metformin, which is a polar and hydrophilic substance, sesamol is a phenolic compound that has a higher degree of hydrophobicity[25-26]. Due to this disparity, it is necessary to carefully optimize the parameters of the HPLC, which includes selecting an appropriate stationary phase, mobile phase composition, & detector wavelength. Previous research[27–28] has resulted in the development of methodologies that may be used to determine the individual dosage of metformin and sesamol. On the other hand, there are not many methods that allow for their simultaneous measurement, and most of them often lack stability. Consequently, the purpose of our study is to fill this void by using a proven HPLC method that is capable of accurately and fairly quantifying both compounds in a single round of analysis. Metformin and Sesamol, two medications with significant therapeutic value, are concurrently calculated by means of a reliable HPLC method that was developed and verified in the course of this study. ICH regulation used in method validation process[29]. We conducted an in-depth analysis of a number of parameters, including a linear pattern, accuracy, precision, LD, LOQ, & robustness[30–31]. In conclusion, the validated HPLC approach employed in this investigation will provide a reliable instrument for the simultaneous measurement of metformin and sesamol. This will make it possible to maintain quality control and ensure that combined formulations are successful in treating their intended purpose.

MATERIALS AND METHODS

Chemicals and Reagents

The standard Metformin and Sesamol procured through chemical store of Integral University, Lucknow, India. Acetonitrile (HPLC-grade), Solvent methanol (HPLC-grade), along with other chemicals acquired from Thermo Scientific & Merck India.

Compatibility Analysis

Using FTIR spectrum analysis, we were able to evaluate whether or not metformin and sesamol are compatible with one another. Scans were performed on samples in an atmosphere of dry nitrogen at temperatures ranging from 40 to 400°C. The heating rate was 20°C per minute, and the resulting curve was formed was thoroughly investigated to identify any possible interactions[32-33]. It was determined by the use of Fourier transform infrared spectra (FTIR) (4000-500cm Bruker Tensor 37, Japan) that a pharmaceutical combination was compatible[32,34].

Instrumentation and Chromatographic Conditions

An HPLC system equipped with a variable wavelength programmable UV/VIS detector (SPD-10AVP, Shimadzu, Tokyo, Japan), system controller (SCL 10AVP), Rheodyne injector with a 20-mL loop, quaternary LC-10A VP pumps, & Class-VP 5.032 software package helped conduct the investigation. Furthermore, C18 (Purospher® STAR RP-18 end-capped (5 µm) Hibar® RT 250-4.6) HPLC column applied. Whole system was kept at what were regarded as ambient temperatures.

Mobile Phase Preparation

Mobile phase was prepared by simply dispersing acetonitrile in water. After preparing acetonitrile and water combination at ratio of 30:70, mixture was degassed in an ultrasonicator for 15 minutes.

Stock and Standard Solutions Preparation

An exact weight of ten milligrams of the standard powder (Metformin and Sesamol) was taken, and then it was dissolved in ten milliliters of HPLC-grade water. The stock solutions were fully dissolved, subjected to sonication, and filtered using nylon membrane filters with pore size of 0.22 µm along with 25 mm diameter before the formation of the final working solution. By diluting the medications with water in an appropriate manner, a standard working solution of the medicines was produced. By establishing a stock calibration curve, a range of 20 to 1.25 µg/mL was created. To reach the required concentrations of working solutions, the stock solution was diluted by the use of the serial dilution process. When doing the study, the concentration range that was used was 20, 10, 5, 2.5, and 1.25 µg/mL.

Development and Optimization of HPLC Method
HPLC Analysis Common Wavelength Selection

Primary research objective is developing technique that would be capable of accurately determining the concentrations of metformin and sesamol in the nanoformulation, as well as during the process of separating them. Specifically, the absorbance maxima of Metformin and Sesamol were observed at λ = 266 nm and λ = 307 nm, respectively. A single wavelength i.e. 230 nm was determined by evaluating the absorbance of the solution of the Metformin Sesamol and a combination of 200 µg/mL solution of Metformin and Sesamol was selected to detect both medicines simultaneously. At a wavelength of 230 nm, the concurrent detection was carried out. This was done at the point when the compounds were considered to be isosbestic. The two drugs were assessed separately at their specific absorption maxima of 266 & 307 nm[35]. This was accomplished to allow for individual comparison of the analytes.

Mobile Phase Optimization

Assessing the ability to dissolve and systemic elution of the two drugs across different mobile phases, while keeping a steady flow rate, facilitated the identification of optimal conditions for enhanced analyte resolution. Prior to the use of mobile phases, they underwent filtration through a Whatman filter with a concentration of 0.45 µM and were subsequently degassed.

To achieve optimal chromatographic conditions, a number of different combinations of acetonitrile and water were investigated, including 30:70, 40:60, 50:50, 60:40, and 70:30 percentages. After that, the chromatographic conditions were validated in line with the recommendation Q2 (R1) of the International Council for Harmonization[36].

Methodology Validation

The methodology was verified in conformity with ICH recommendations. Specificity, linearity, sensitivity (LOQ and LOD), accuracy, range, precision, & robustness had been factors that were evaluated throughout the validation process[29,37].

Specificity

In the presence of chemicals that might interfere with the analysis of the analyte, specificity refers to capacity for evaluating analyte without any uncertainty. In general, they could consist of

matrices, degradants, contaminants, and other similar substances[29]. The method's specificity was assessed by independently injecting 20 µL solutions of the standard, sample, blank, and placebo into each other[38].

Accuracy

An analytical procedure's accuracy is determined by the degree to which the value observed corresponds with the value recognized as a conventional true value or an acceptable reference value[29]. Three samples from each concentration were injected, then recovery experiments were subsequently conducted at three distinct concentrations for evaluating the assay method's accuracy: 50%,100%,200%, which correspond to 85, 170, and 255 µg/mL. The percentage of recovered metformin and sesamol, in addition to the relative standard deviation, was estimated for each and every duplicate sample[39].

Precision

The consistency of measurements acquired from many samplings of an identical homogeneous sample under prescribed conditions is what is meant by the term "accuracy" when referring to an analytical procedure. Variance, standard deviation, or coefficient of variation are often used to quantify it[29]. It is possible to measure it using samples that are homogeneous or samples that have been purposefully produced. The proposed methodologies were evaluated by performing many measurements on a reference solution to determine system accuracy & method precision (repeatability) of recommended procedures. A total of six measurements of the standard solution of Metformin and Sesamol at a concentration of 100% were taken on same day in order to assess the accuracy of the system. These evaluations were used to verify the precision of the method. For the purpose of determining whether or not they are repeatable, relative standard deviation of collected results were estimated[40].

Linearity and Range

An analytical technique's linearity indicates its ability of yielding test results that correlate with analyte concentration in sample, while its range defines interval between upper & lower concentration limits. Several standard solutions had been prepared by diluting standard stock solution with acetonitrile to concentrations of 20, 10, 5, 2.5, & 1.25 µg/mL for evaluating linearity & range of method. Under uniform conditions, three injections from each concentration have been examined. Linear regression analysis assessed the calibration curve's linearity through the least square's linear regression method[41,42].

Robustness

The term "robustness" refers to the ability of an analytical process to withstand slight, purposeful changes in method parameters. This ability reflects the process's dependability while running under typical operating conditions[29]. In order to confirm the robustness of the process, a number of alterations, both slight and purposeful, were made to the experimental parameters which were as follows: Flow rate: ±0.2 milliliters per minute Wavelength: ±3 nanometre wavelength[43].

Sensitivity

LOD & LOQ determined using formulae LOD = 3.3 σ/s and LOQ = 10 σ/s on basis of standard deviation of y-intercepts of regression line & calibration curve slopes. At the same time as the slope of the curve is represented by the symbol s[44], the standard deviation is represented by the symbol.

Preparation of Liposomes

In order to manufacture combinatorial Metformin and Sesamol liposomes, the ethanol injection method was used. These liposomes had the required quantity of phospholipid, cholesterol, and Tween-20. Particle size, polydispersity index (PDI), & zeta potential among numerous properties that were assessed for this formulation using a Zetasizer Nano-ZS90 from Malvern Instruments.

Simultaneous Quantitative Assessment of Metformin and Sesamol in Formulated Liposomes

To determine Metformin and Sesamol liposomes, an equivalent amount of formulation containing metformin & sesamol (10 mg) accurately weighed then shifted in volumetric flask containing mobile phase (10 mL). Sonicating nanoformulation for 20 min dissolved it. Passing through a membrane filter (0.45µm), Metformin & Sesamol concentrations measured in triplicate applying newly designed and verified HPLC technique at 25±2°C.

RESULTS AND DISCUSSION

Compatibility Study

Drug formulation compatibility verified

through the acquisition of FTIR spectra. FTIR spectrum of a combination of medications demonstrated existence of the functional peaks that are typical of both metformin and sesamol. This confirmed that there was no chemical interaction between the two drugs (Figure 1).

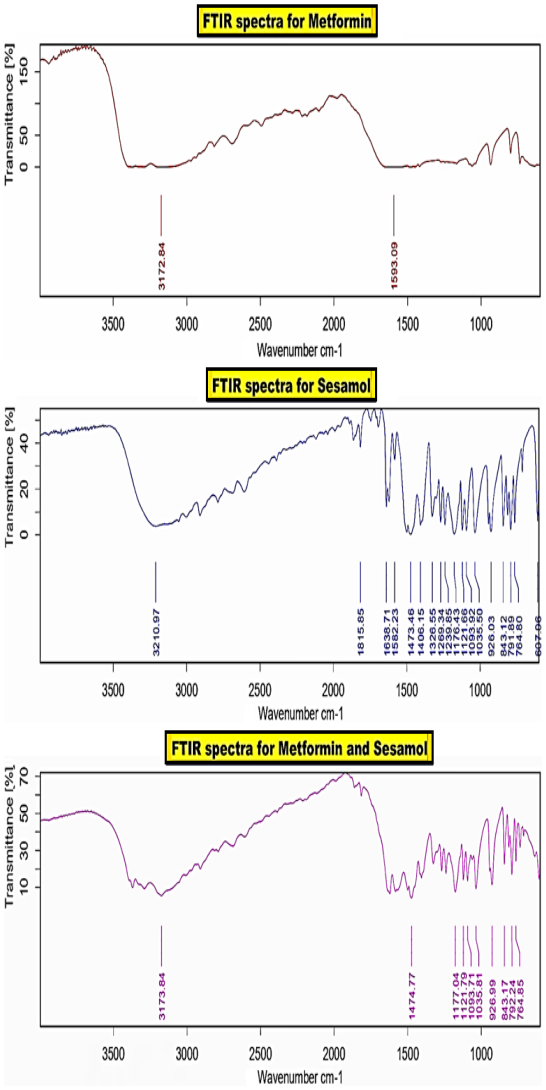

Fig. 1. FTIR spectra of metformin, sesamol and physical mixture of metformin and sesamol

Optimization of Mobile Phase and HPLC Conditions

In this study, a reversed-phase HPLC method that includes UV-Vis detection is presented to measure metformin and sesamol in liposomal formulation. As a result of its broad use in chromatographic techniques for the identification of metformin and sesamol, the C-18 column was selected because it produces better peak shape and resolution on the analytical instrument. The use of isocratic elution was chosen due to its ease of usage, which included the use of a single pump. This method significantly reduced baseline variance and ghost peaks, as seen in Fig. 2. In Tables 1 and 2, mobile phase optimization is shown for clarity. For analyzing metformin and sesamol, it was established that mobile phase consisting of water & acetonitrile at a ratio of 70:30 volume/volume was considered to be suitable. To obtain optimal conditions for the measurement of Metformin and Sesamol at a UV detection wavelength of 230 nm, the flow rate of 1.0 ml/min and the injection volume of 20 µL were used.

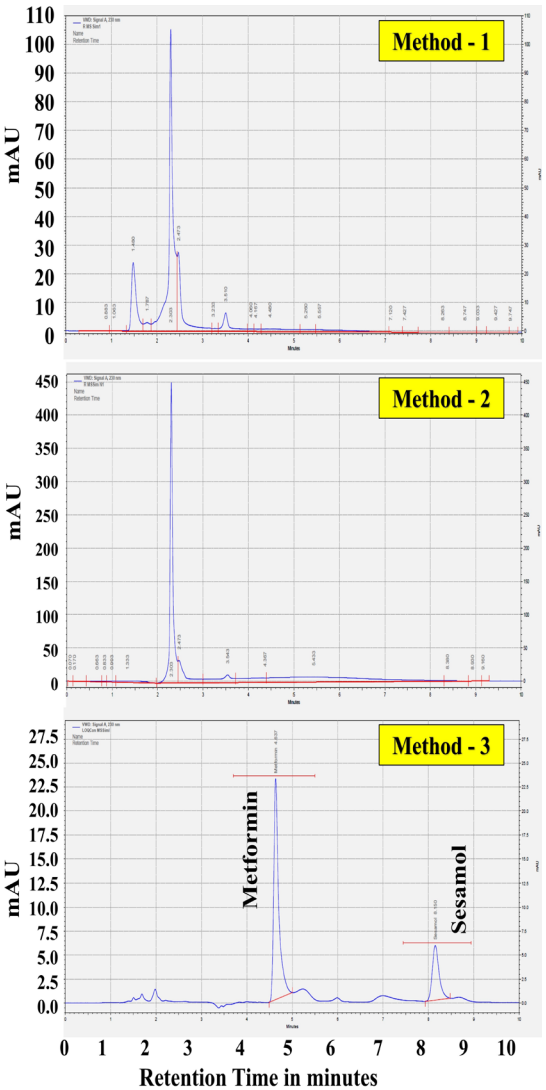

Fig. 2. Chromatograms obtained by Method-I, II, and III (Optimized) followed for simultaneous determination of Metformin and Sesamol

Table 1: Optimization of mobile phase

Method	Mobile phase composition	Wavelength	Flow rate	Resolution
I	Water:Acetonitrile (Isocratic)	230nm	1 mL/min	Average
II	Water:Acetonitrile (Gradient)	230nm	1 mL/min	Average
III	Water:Acetonitrile (Isocratic)	230nm	1 mL/min	Good (used for further study)

Table 2: Optimized chromatographic conditions

Parameter	Conditions
Stationary phase	AgilentTC-C18(2), 4.6x250 mm, 5 µm
Mobile phase	Acetonitrile: Water
Mobile phase ratio	30:70
Detection Wavelength	230 nm
Flow Rate	1.0 mL/min
Sample Volume	20 µL
Temperature	Ambient
LC System	Agilent test system and Open Lab CD S2

Validation Parameters

The analytical technique was validated in accordance with the criteria specified in the ICH guideline Q2(R1), emphasizing precision, specificity, accuracy, LOD, robustness, linearity, & LOQ.

Specificity

New method's specificity evaluated by separating two medicines, which showed no extra peak Note that a blank sample was conducted for comparison. Since (Fig. 3) does not show a solvent system chromatogram, this newly verified approach showed that both medicines were eluted without interference. The instrument's specificity was tested using blank, diluent, and standard runs.

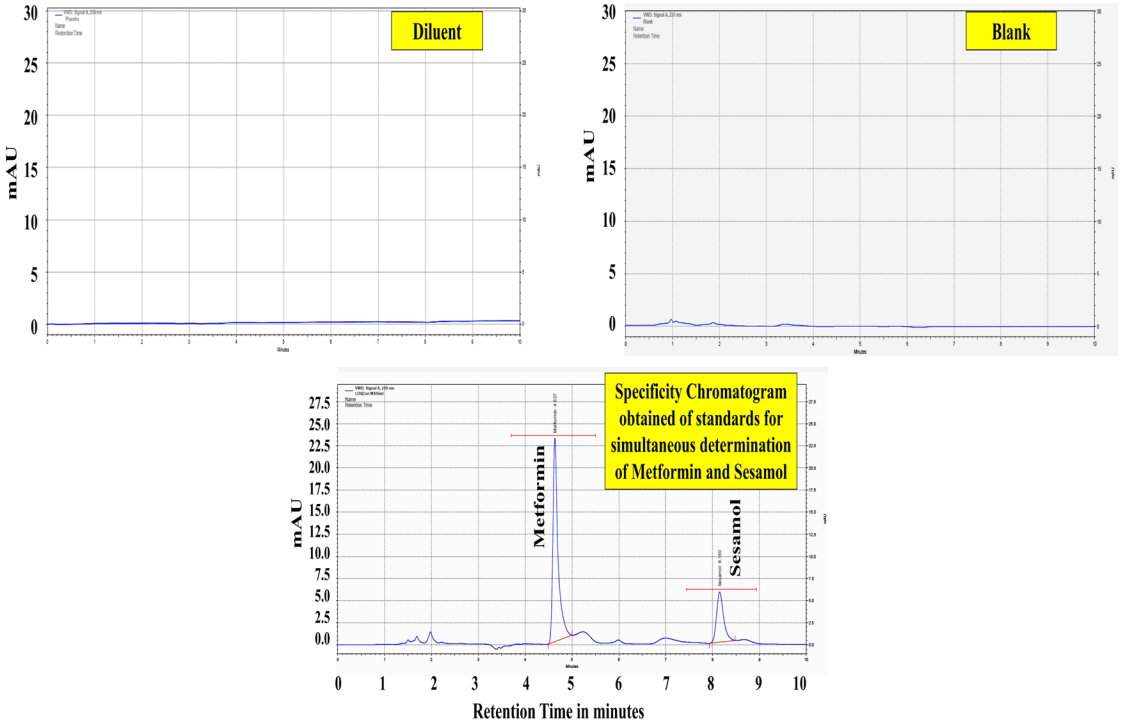

Fig. 3. Specificity Chromatogram obtained of standards for simultaneous determination of Metformin and Sesamol

Recovery and accuracy

When evaluating the accuracy of the developed analytical method, the drug was included into the pre-quantified solution at various concentration levels. These concentration levels included 1.4, 2.8, and 5.6 µg/mL of standard. Additionally, the spiking levels were set at 80%, 100%, and 180%. This was done while considering bulk drug samples percent purity that were added. According to Table 3, the percent recovery accuracy for Metformin varied from 98.432% to 98.790%, but for Sesamol, it ranged from 82.810% to 97.941%. On the other hand, the fact that the %RSD value for both drugs was lower than 2 demonstrated that the HPLC procedure that was developed was accurate (Figure 4).

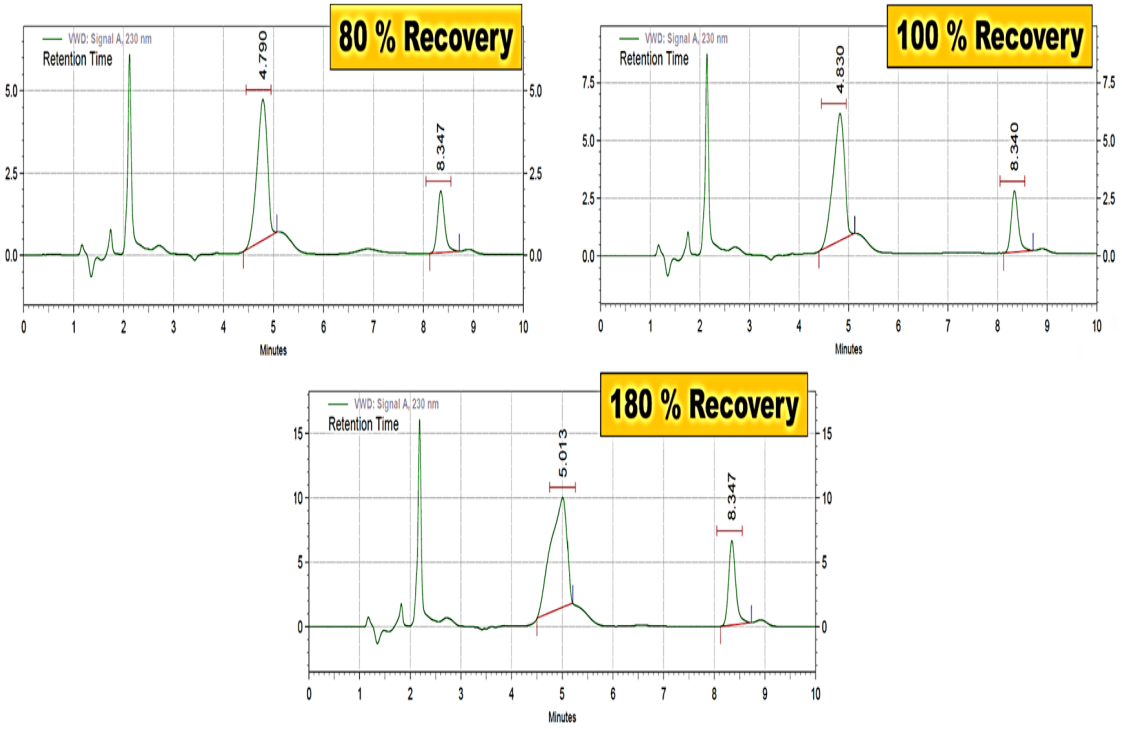

Fig. 4. Chromatogram obtained of Recovery with 80%, 100%, and 180% spiking level for simultaneous determination of Metformin and Sesamol

Table 3: Recovery of metformin and sesamol

Sr. No.	Conc.(µg/mL)	% Spiked level	Metformin		Sesamol	
			%Recovery	Mean %Recovery	%Recovery	Mean %Recovery
1		100% sample	98.432	98.893	96.779	92.510
2	1.4	80% 0.75 µg/mL std				
3		100% sample+80% 0.75 µg/mL std				
1		100% sample				
2	2.8	80% 0.75 µg/mL std	99.458		97.941	
3		100% sample+80% 0.75 µg/mL std				
1		100% sample	98.790		82.810	
2	5.6	80% 0.75 µg/mL std				
3		100% sample+80% 0.75 µg/mL std				

Precision studies

Consistent with ICH guideline Q2(R1), the sample that was created was used to evaluate the precision of both intra- and inter-day measurements, as well as the determination of precision & intermediate accuracy within analytical range of both medications. On separate days, triplicates of three different doses of metformin and sesamol were evaluated in order to see if the variability differed from one day to the next. Tables 4, 5, 6, 7, and 8 provide evidence that accuracy was achieved since all the data falls within the permitted range of less than 2% RSD. This substantiates HPLC technique's repeatability that has been developed (Fig. 5, 6, 7) each contains chromatograms that are relevant to research using precision techniques.

Table 4: Repeatability at LOQ of Metformin and Sesamol

	Metformin	Sesamol
Mean RT	4.6435	8.09383
SD	0.0306969	0.04613
%RSD	0.6610726	0.0057

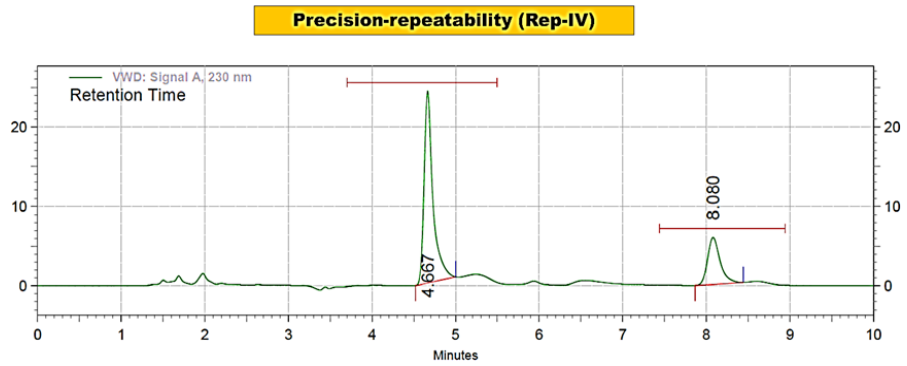

Fig. 5. Chromatogram obtained of Precision-repeatability (Rep-V) for simultaneous determination of Metformin and Sesamol

Table 5: Inter-day precision of Metformin

				Metformin				
Sr. No	Day	Concentration (µg/mL)	Replicate	RT	Mean	SD	%RSD	Mean% RSD
1			1	4.563				
2	1		2	4.597	3254588	37089.077	1.14	
3			3	4.59				
4			1	4.637				
5	2	2.8	2	4.683	2996411	30043.818	1.003	1.048
6			3	4.667				
7			1	4.637				
8	3		2	4.683	2996411	30043.818	1.003	
9			3	4.667				

Table 6: Inter-day precision of Sesamol

				Sesamol				
Sr. No	Day	Concentration(µg/ml)	Replicate	RT	Mean	SD	%RSD	Mean% RSD
1			1	7.993				
2	1		2	8.033	996271.7	13221.222	1.327	
3			3	8.023				
4		3.9	1	8.027				1.584
5	2		2	8.1	1058572	28547.074	2.697	
6			3	8.08				
7			1	8.027				
8	3		2	8.1	1084562	7903.656	0.729	
9			3	8.08				

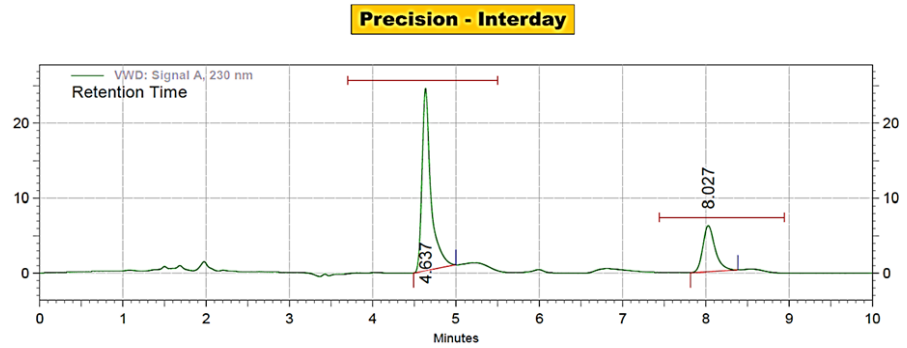

Fig. 6. Chromatogram obtained of Precision-Inter day for simultaneous determination of Metformin and Sesamol

Table 7: Intraday precision of Metformin

Sr. No	Time	Concentration (µg/mL)	Replicate	RT	Metformin Peak area	Mean	SD	% RSD	Mean %RSD
1	1st h		1	4.563	3293835				
2			2	4.597	2949810	3009044.333	260278.783	0.086	
3			3	4.683	2783488				
4	2nd h		1	4.677	3178049				
5		2.8	2	4.653	3030830	3051470	117625.092	0.039	0.048
6			3	4.637	2945531				
7	3rd h		1	4.667	2871345				
8			2	4.65	2784318	2843346	51142.192	0.018	
9			3	4.68	2874375				

Table 8: Intraday precision of Sesamol

Sr. No	Time	Concentration (µg/mL)	Replicate	RT	Sesamol Peak area	Mean	SD	% RSD	Mean %RSD
1	1st h		1	7.993	1111510	1094567.333	14777.127	1.35	
2			2	8.033	1087849				
3			3	8.1	1084343				
4	2nd h	3.9	1	8.107	1049361	1026836.333	29328.728	2.856	1.692
5			2	8.133	1037475				
6			3	8.15	993673				
7	3rd h		1	8.167	971345	981033.333	8534.043	0.87	
8			2	8.165	984318				
9			3	8.168	987437				

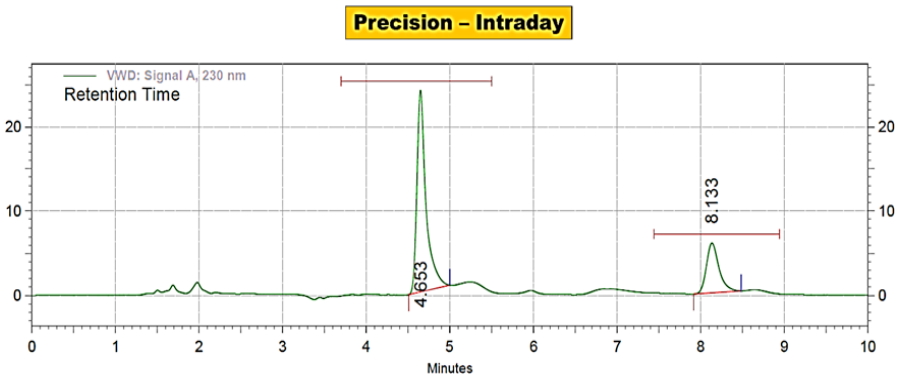

Fig. 7. Chromatogram obtained of Precision-Intraday for simultaneous determination of Metformin and Sesamol

Linearity and Range

Metformin and sesamol's linearity were evaluated via development of calibration curves for each of these medications. In (Fig. 8) it was observed that the peak areas of Metformin and Sesamol exhibited linearity within the concentration ranging 20-1.25 µg/mL for both the drugs. When solutions were injected in triplicate for the purpose of determining linearity, results demonstrates that RSD of peak area and RSD of retention time for Metformin and sesamol were 1.7114 and 1.9700, and 0.7934 and 0.1739, respectively, in combination with the calibration. When both drugs were administered at the concentrations that were indicated, the graphs displayed a linear pattern. The excellent linearity of the graphs is shown by the fact that correlation coefficients (R2) for calibration curves of metformin and sesamol were found to be 0.9947 and 0.9908, respectively. Chromatograms obtained for linearity experiments are shown in (Figure 9).

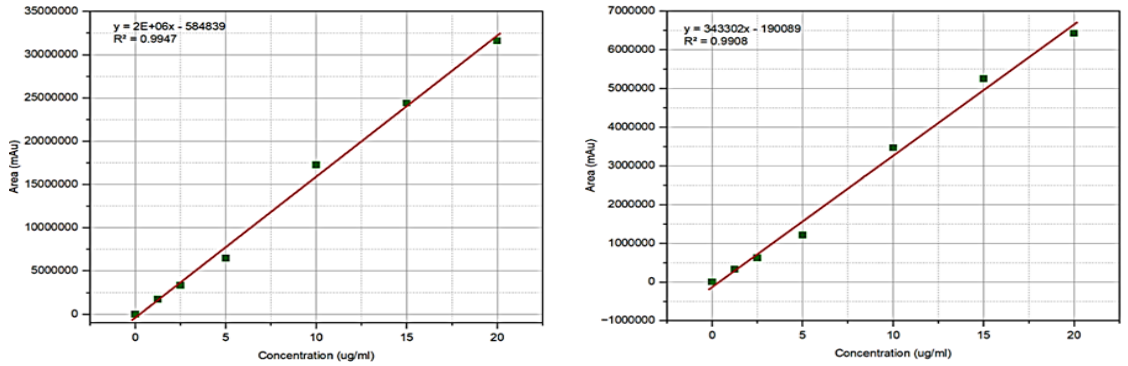

Fig. 8. Calibration graphs of Metformin and Sesamol

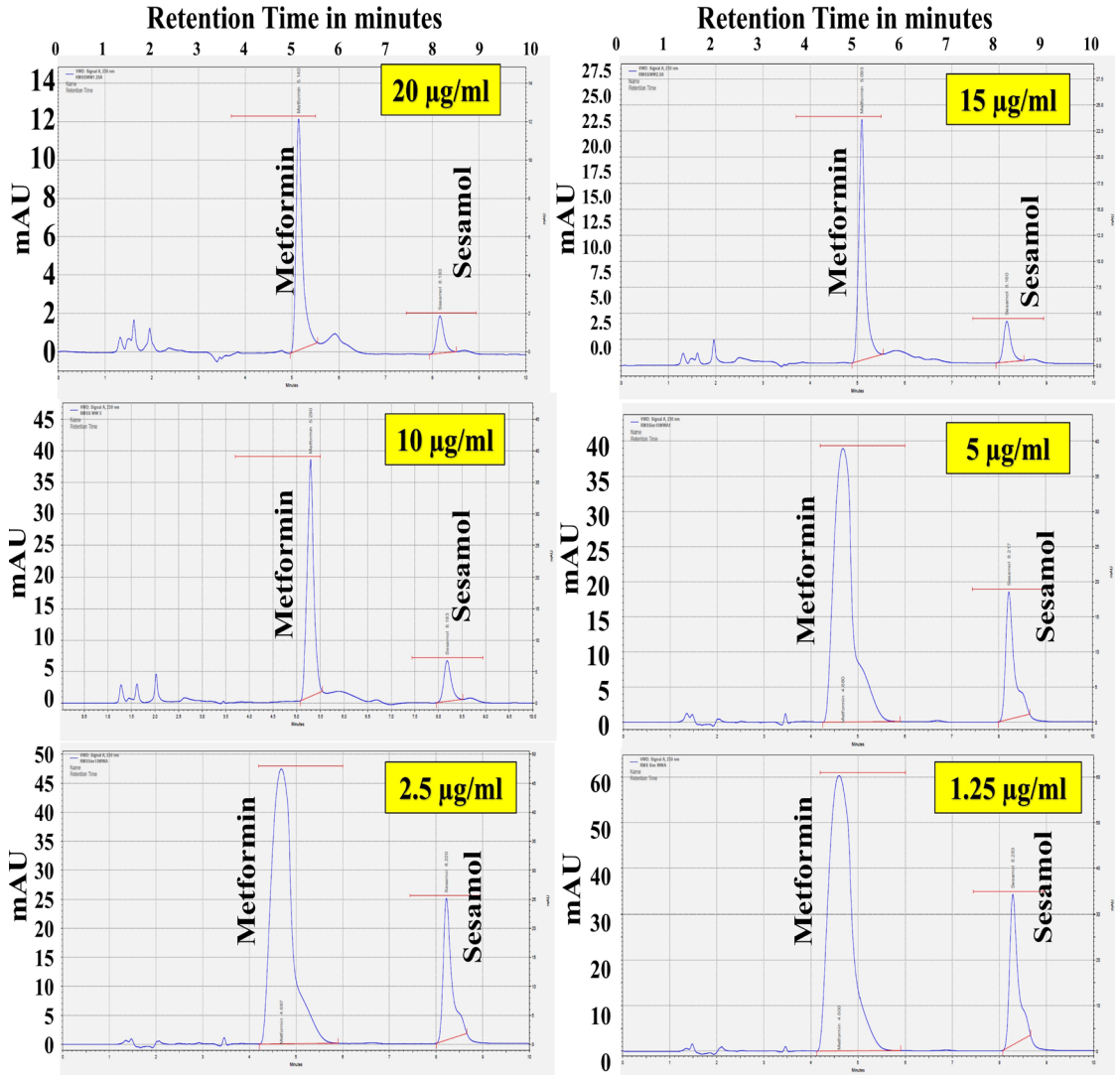

Fig. 9. Chromatogram obtained at 20, 15, 10, 5, 2.5, 1.25 µg/mL concentration of Metformin and Sesamol for linearity

Robustness

The suggested method was tested under normal operating settings, and its robustness was verified by adjusting the wavelength, organic solvent concentration, and flow rate of the HPLC pump (Table 9, 10). In spite of the fact that the %RSD was less than

2, the technique displayed robustness. Metformin's resistance was evaluated by modifying three parameters: the wavelength (with a tolerance of ±3 nm), flow rate (with a tolerance of ±0.2 min), and the mobile phase concentration (with a tolerance of ±2% organic solvent). An illustration of the chromatograms that are relevant to the robustness studies may be observed in the (Figure 10, 11, and 12).

Table 9: Robustness of Metformin

Sr. No	Parameter	Condition	Metformin Mean RT	SD	%RSD	Mean Peak Area	SD	%RSD	Mean %RSD
1	Flow rate	0.8	6.225	0.015	0.237	3278877	55711.73388	1.699	0.573
		1.2	3.95	0.04	1.011	3402712.333	44574.43463	0.013	
		1	4.513	0.015	0.338	3398865	20652.2209	0.006	
2	Mobile Phase	ACN-28	4.785	0.031	0.652	3344347.333	2324.36321	0.001	0.008
		ACN-30	4.5	0.082	1.819	3225397	64105.85018	0.02	
		ACN-32	4.992	0.03	0.591	3253065	13106.56923	0.004	
3	Wave length	227nm	4.72	0.026	0.561	3255785	5534.758622	0.002	0.002
		230nm	4.481	0.012	0.264	3381526.667	4035.897092	0.001	
		233nm	4.738	0.006	0.12	3489777.333	12048.77763	0.003	

Table 10: Robustness of Sesamol

Sr. No	Parameter	Condition	Sesamol Mean RT	SD	% RSD	Mean	SD	% RSD	Mean %RSD
1	Flow rate	0.8	10.478	0.0062	0.0596	975306.667	15802.15	1.62	1.142
		1.2	6.9077	0.004	0.0585	872968.333	10304.142	1.18	
		1	8.3647	0.0329	0.3929	982742.667	6136.046	0.624	
2	Mobile Phase	ACN-28	9.1463	0.017	0.186	857696.667	9842.55	1.148	1.057
		ACN-30	8.3243	0.0125	0.1502	807816.667	6020.713	0.745	
		ACN-32	7.6647	0.0391	0.5107	938225	11991.952	1.278	
3	Wave	227nm	8.282	0.0653	0.7882	948480.333	19599.655	2.066	1.288
	length	230nm	8.3713	0.0297	0.3552	898645	6685.845	0.744	
	233nm	8.32	0.0265	0.318	930823	9793.353	1.052		

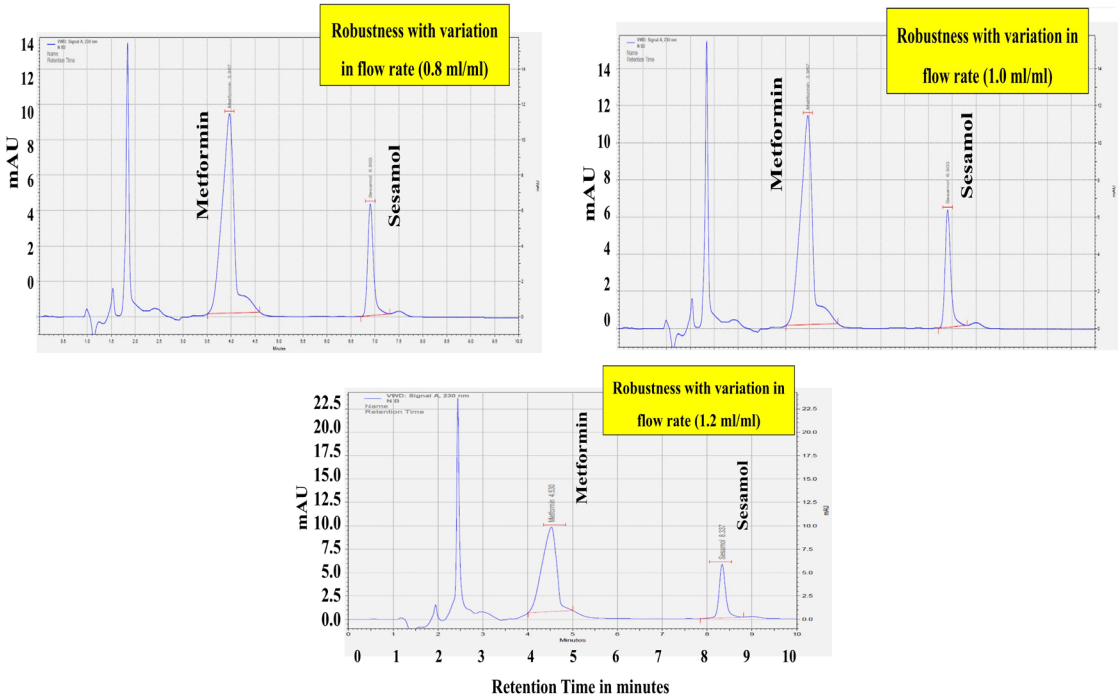

Fig. 10. Chromatogram obtained of Robustness with variation in flow rate (0.8, 1, 1.2 mL/min) for simultaneous determination of Metformin and Sesamol

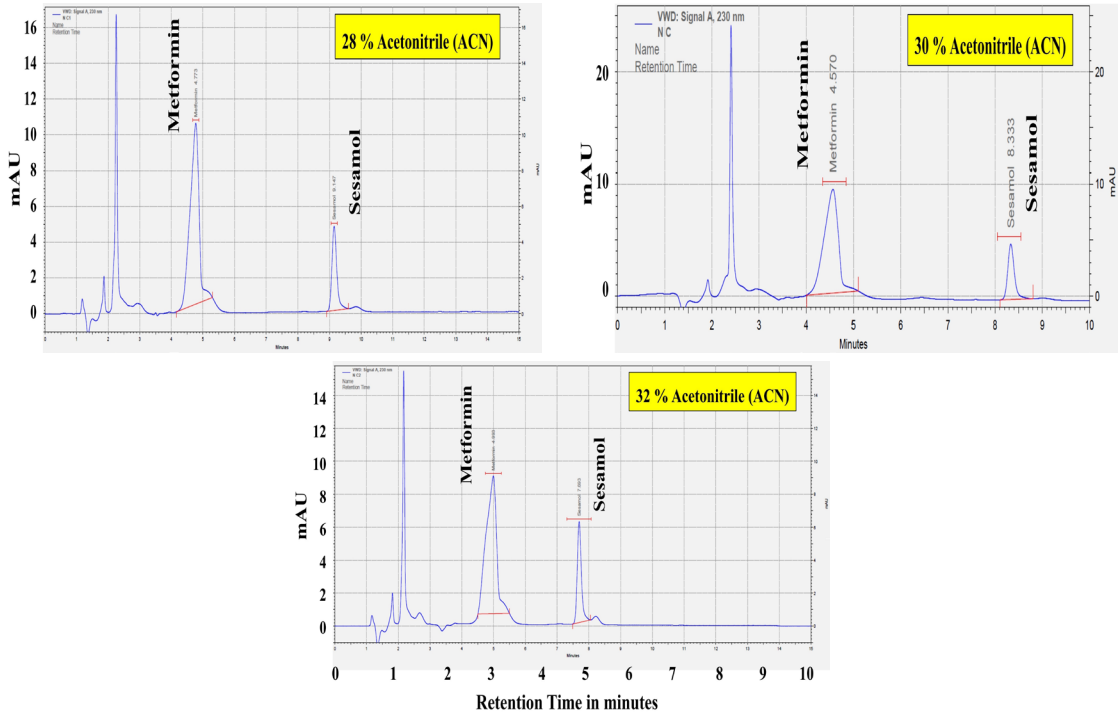

Fig. 11. Chromatogram obtained of Robustness with variation in ACN (28, 30, 32%) for simultaneous determination of Metformin and Sesamol

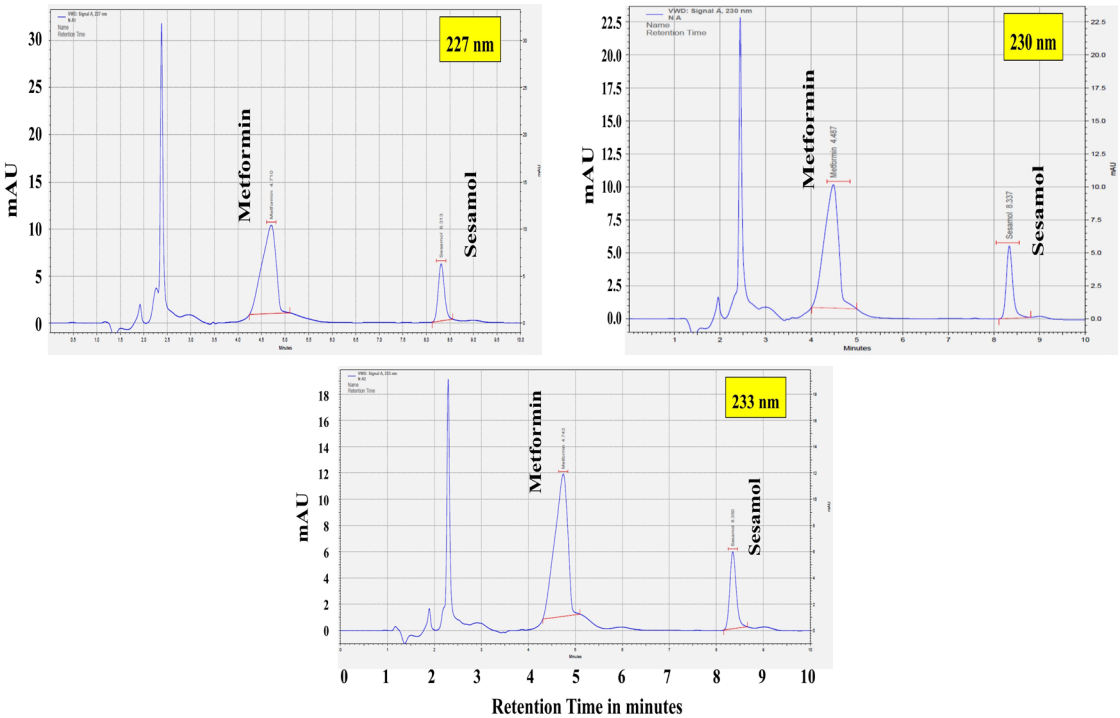

Fig. 12. Chromatogram obtained of Robustness with variation in wavelength (227, 230, and 233 nm) for simultaneous determination of Metformin and Sesamol

LOD and LOQ

For metformin, LOD & LOQ determined to be 0.8947 & 2.7114 µg/mL, respectively, whereas for sesamol, 1.2754 and 3.8648 µg/mL, respectively.

As a result, the developed method may precisely measure the lowest levels of metformin and sesamol, which would be very advantageous for determining the amounts of these two medications in the final product or any other formulations.

Utilization of Designed and Verification of Simultaneous Method towards Drug-loaded Liposomes

PDI & particle size of drug-loaded formulations found to be 0.040±0.011 and 389.1±2.17 nm, respectively. It was discovered that size of formulation was within nano range, and zeta potential of drug-loaded liposomes was found to be -14.9 mV. The suggested and proven method was used in a liposome formulation that included a number of different excipients in order to concurrently measure the levels of metformin and sesamol content. In no manner did the drugs and the excipients interact with one another. The drug-loaded liposomes were found to contain 98.364% metformin and 96.891% sesamol, respectively, according to the findings.

CONCLUSION

Pharmaceutical analysis has made considerable progress with the development and subsequent validation of a robust HPLC method for the simultaneous assessment of metformin and sesamol. This methodology offers a method that is both reliable and effective for measuring these compounds in bulk and formulated items, so ensuring that quality control and regulatory compliance are maintained. Validated high-performance liquid chromatography (HPLC) technology demonstrated great specificity, precision, accuracy, and linearity, which made it an efficient instrument for routine analysis. Its versatility and potential for use in pharmaceutical research and development activities are shown by the fact that it was utilized in the assessment of nanostructured formulations *(liposomes)*. By making it feasible to conduct accurate measurements of metformin and sesamol in complex matrices, this technology will be of great assistance in the development of novel drug delivery methods, as well as in our understanding of the pharmacokinetics and therapeutic efficacy of these drugs. Taking everything into consideration, the findings of this study highlight how important it is to develop robust analytical methods to keep up with the rapidly changing environment of pharmaceutical formulations.

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Conflict of Interest

The author declare that we have no conflict of interest.

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