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Original Article

Strengthening Finerenone Assessment: A Central Composite Design Viewpoint for Liquid Chromatographic Method Development

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ABSTRACT

Background: A RP-HPLC technique was created and proven for the measurement of Finerenone, a new medication currently being studied. The approach was improved to achieve excellent precision, accuracy, and sensitivity for use in pharmaceutical formulations. **Materials and Methods:** The separation by chromatography took place utilizing an isocratic elution method with a mobile phase of 0.01N dihydrogen of potassium orthophosphate and acetonitrile in a 65:35 (v/v) proportion. The velocity of the fluid was set at 1 mL/min, and the division happened via an Agilent C18 (250×4.6 mm, 5 µm) at 30ºC. Finerenone was detected at a wavelength of 230 nm, at a retention time of 2.306 min and the injection volume was 10 µL. **Results:** The developed method exhibited excellent linearity with an R^2 of 0.999 over the concentration range of 50% to 150%. Precision studies demonstrated good repeatability (RSD=0.4%) and day after (RSD=0.5%). The linearity range of 50% to 150% translates to a concentration range of 25 µg/mL to 75 µg/mL. The recovery of Finerenone from spiked samples was 99.47%, indicating high accuracy. The LOD and LOQ were determined to be 0.041 µg/mL and 0.123 µg/mL, respectively, suggesting high sensitivity of the method. When applied to a marketed formulation of Finerenone, the assay value obtained was 99.62%, confirming the expected concentration in the formulation. **Conclusion:** The RP-HPLC method developed in this study is precise, accurate, and highly sensitive, making it suitable for the quantification of Finerenone in pharmaceutical formulations. The method was successfully validated and can be applied in both research and quality control settings. Additionally, the incorporation of Quality by Design (QbD) and DOE principles throughout the method development process ensures its robustness and reliability.

Keywords: Central composite design, Finerenone, Perturbation, Response surface plots, Statistical analysis.

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INTRODUCTION

Finerenone in, a novel non-steroidal mineralocorticoid receptor antagonist, has demonstrated significant potential in managing Chronic Kidney Disease (CKD) and Heart Failure with reduced Ejection Fraction (HFrEF). This emerging therapeutic agent necessitates the development of robust analytical methods to ensure its quality and efficacy in pharmaceutical formulations. QbD provides a scientific framework for identifying and reducing significant sources of variability, improving the method's sensitivity, specificity, accuracy, and robustness.

Central Composite Design (CCD), a powerful statistical tool, is employed to optimize key chromatographic parameters, such as mobile phase composition, flow rate, and temperature, ensuring the development of a high-performance Reverse-Phase High-Performance Liquid Chromatography (RP-HPLC) method. The optimized method meets stringent criteria for pharmaceutical analysis, addressing limitations of previously reported methods, such as reduced sensitivity, selectivity, and robustness. By proving the method's precision, accuracy, resilience, and sensitivity under varied conditions, the validation process establishes its reliability for research applications and routine quality control in pharmaceutical formulations. Numerous studies have documented RP-HPLC methods for Finerenone analysis, with varying degrees of success in achieving precision and stability under different conditions.[1-9] Incorporating QbD and CCD significantly enhances method reliability compared to traditional

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approaches, ensuring comprehensive coverage of critical method parameters. The novel method integrates advanced analytical tools and principles to achieve superior detection limits, quantification accuracy, and consistent results in stability-indicating and formulation-specific assessments.[10-12] This research is guided by international regulatory standards and methodologies, such as those outlined by the International Council for Harmonisation (ICH) Q2(R1), to ensure global acceptance.[13-17] Recent advancements in chromatographic techniques, as highlighted in contemporary literature, further inform this development process, ensuring alignment with cutting-edge practices.[18-22] The method's validation underscores its reliability and application in both research and quality control, facilitating regulatory compliance and safeguarding patient safety.[28-39] Ultimately, this QbD-driven RP-HPLC method represents a significant advancement in pharmaceutical analytical practices, offering a robust, efficient, and reliable tool for the analysis of Finerenone in various formulations. This approach exemplifies the integration of innovative methodologies into pharmaceutical sciences, ensuring the development of therapeutic agents with un compromised quality and efficacy.

MATERIALS AND METHODS

Chemicals and Reagents

Finerenone, Methanol HPLC Grade (RANKEM), Acetonitrile HPLC Grade (RANKEM), HPLC grade Water (RANKEM), Glacial Acetic acid, 0.1 N Hydrochloric acid, 0.1 N Sodium hydroxide, 0.1% Hydrogen peroxide, Trifluoroacetic Page acid-HPLC grade.

Devices

HPLC instrument used was of WATERS HPLC 2965 SYSTEM with Auto Injector and PDA Detector. Software used is Empower, UV-VIS spectrophotometer PG Instruments T60 with special bandwidth of 2 mm and 10 mm and matched quartz was used for measuring absorbance for Finerenone solutions, Sonicator (Ultrasonic sonicator), PH meter (Thermo Scientific), Microbalance (Sartorius), Vacuum filter pump.

Diluent

Based on the drug's solubility, the diluent was chosen as a mixture of acetonitrile and water in a 50:50 ratios.

Conventional inventory Solution

5 mg of Finerenone active grade was considered and deposited to a 50 mL volumetric flask. 30 mL of diluent was put in and the entire mix underwent ultrasound treatment for 5 min. To create a conventional inventory solution with a level of 100 mcg, the container size has been raised to 50 mL using the diluting agent.

Functional inventory solution (100% solution)

Introduced 1 mL of the conventional inventory solution into a 10 mL volumetric flask and attenuated to the marking to accomplish the final level of 10 mcg of Finerenone.

Specimen Solution

Five tablets were weighed and pulverized to a fine powder. A portion of powdered corresponding to the typical mass of the pills was put to a volumetric flask measuring 100 mL. To generate a 100 mcg Finerenone solution, immersed the dried powder in diluting agent, vibrated for 15 min, and subsequently reduced to the desired volume.

Functional Specimen Solutions (100% solution)

To experience an ultimate level of 60 µg/mL of Finerenone, moved 1 mL of the filtered sampling inventory solution to a 10 mL volumetric flask and reduced with moderator.

Inventory for performing Degradation study

A single serving of pulverized pellets that equalled an average value has been placed in a 50 mL volumetric flask. Then, 30 mL of moderator was incorporated, and the whole thing underwent ultrasound for 15 min. To produce a 100 µg/mL Finerenone approach tweaked the total amount to 50 mL and added medium.

Preparing the buffering agent

1.36 w/w of KH_2PO_4 was precisely counted and put to a 1000 mL volumetric flask. To degas, incorporated around 900 mL of Milli-Q water and sonicated it. The final volume was modified with water, and the pH was set to 4.0 using 0.1% v/v OPA (1 mL of it in 1000 mL of HPLC equal water).

Handling

The separation through chromatography occurred utilizing isocratic resolution with a 0.01N potassium dihydrogen orthophosphate and acetonitrile in a 65:35 v/v ration. The fluid velocity was placed as 1 mL/min. An Agilent C18 analytical column (250×4.6 mm, 5 µm) was used as the stagnant phase, and the column heat was maintained at 30°C. Finerenone showed up at 230 nm with a 10 µL injector volume. Finerenone's peak shape was satisfactory, and the system adaptability indicators were within reasonable limits.

Development involving QbD

Central Composite Design (CCD) is a powerful statistical technique used in response surface methodology for optimizing processes and experiments. It allows researchers to efficiently explore the relationships between multiple factors and their interactions by systematically varying conditions. By employing CCD, one can derive a comprehensive understanding of how

different variables affect a response variable, facilitating more informed decision-making. This approach is particularly valuable in fields such as pharmaceuticals, where precise method optimization is essential for ensuring quality and efficacy.

Preliminary studies for factor selection

An initial investigation took place into choose variables listed in Table 1 for QbD. The essential variables for the HPLC method were chosen based on the risk estimation matrix and preliminary experiments, together with their levels, to screen and research designed to optimize the HPLC method, allowing Finerenone to be separated in a short run time. The preliminary trial findings show in Table 2 that the amount of organic solvent in the mobile phase is crucial for estimating both medicines. As a result, the fraction of organic solvent in the mobile phase (X1) was chosen as one of the essential parameters to investigate further. The velocity of flow (X2) and warmth of the column have been shown to possess an influence on the principal system fit challenges, whereas Retaining Duration of Finerenone, tailing component, and Number of Conceptual Panels were chosen as response variables.

Statistical analysis

Statistical analysis was utilized to build second-order quadratic equations that best match the experimental data while also predicting responses. Quadratic models with straightforward, connection, and exponential terms were created across all response parameters deploying design expert 12.0 program. The model with the greatest fit was chosen following an evaluation of all the data points included in Design Expert 12.0. Furthermore, an ANOVA, as shown in Table 3, was performed to determine the main contribution of relevant components to model predictions. The Friedman test and its p-value have been calculated using Design Expert 12.0 tools.

RESULTS

System suitability

Six repeat shots of the operational solution in question (Finerenone at 10 µg/mL) served to figure out system appropriateness. The average, standard deviation, and % standard relative deviation have been measured. To ensure efficient separation and system performance, the analysis implied a low tailing factor of 1.2%, elevated number of theoretical plates (>8000), constant retention times.

The degree of specificity

To judge the uniqueness, HPLC was used to examine a blank, a standard and test of Finerenone at 10 mcg strength. The chromatograms for all of them are provided.

Precision

Six mimic infusions of the active stock solution (Finerenone 10 µg/mL) were used. The norm, deviation from the average, and percentage comparative variance were arrived. To ensure technique precision (assay repeatability), six mimic infusions of the norm as well as test (Finerenone 10 mcg) were conducted in triplicate and looked at. Intermediate preciseness was scored in an analogous way on several days.

Linearity

In order to verify a straight line, reductions of Finerenone (100 µg/mL) from 2.5 µg/mL to 15 µg/mL have been generated and fed into the HPLC.

Accuracy

The exactitude of what was done came to light using the usual additional method. The percentage returned was arrived at in threefold at three different tiers (50%, 100%, and 150%). The norm, deviation from the average, and percentage comparative variance arrived.

LOD and LOQ

The linearity curve was used to calculate the Limits of Detection (LOD) and Quantification (LOQ) as deviation from the standard method.

Robustness

To figure out, purposeful changes were made to the chromatographic settings, Adjust the mobile phase composition (±2 mL organic mix), velocity of circulation (±0.2 cc/min), and short waves (±2 nm).

Table 1: Uncorrelated data points (factors) and their consequences.

Independencies	Variability			Depencies	Hit point
	low (-1)	medium (0)	high (+1)	Retention time of Finerenone RT1; Number of theoretical plates (NTP)	Lowering retaining time while optimizing closure.
Velocity of moving phase, mL/min	0.2495	0.3000	0.3505		
Acetonitrile ration	31.59	40.00	48.41		
Temperature, °C	24.95	30.00	35.05		

Table 2: Total runs with actual values for three factors and two responses.

Run	Type	F1	F2	F3	R1	R2
1	Factorial	0.249546	40	30	2.165	3652
2	Axial	0.27	38	28	2.151	3300
3	Factorial	0.27	38	32	2.017	3560
4	Center	0.27	42	28	2.023	4190
5	Center	0.27	42	32	1.937	3526
6	Axial	0.3	36.6364	30	1.93	3035
7	Factorial	0.3	40	26.6364	2.004	4027
8	Center	0.3	40	30	1.874	4384
9	Center	0.3	40	30	1.87	4375
10	Factorial	0.3	40	30	1.871	4498
11	Axial	0.3	40	30	1.871	4387
12	Factorial	0.3	40	30	1.874	4418
13	Axial	0.3	40	30	1.872	4334
14	Axial	0.3	40	33.3636	1.784	4190
15	Factorial	0.3	43.3636	30	1.813	2975
16	Center	0.33	38	28	1.829	3514
17	Factorial	0.33	38	32	1.733	3475
18	Center	0.33	42	32	1.749	3240
19	Factorial	0.33	40	28	1.655	2845
20	Axial	0.350454	42	42	1.638	3672

F1-Flow rate; F2-Acetonitrile; F3-Temperature, °C R1-Retention time of Finerenone; R2-NTP-Number of Theoretical Plates.

Forced degradation

Acid deterioration

1 mL of the stock solution was sent to a 10 mL graduation container. To this, 1 mL of 2N HCl was added up, container was let to remain at room heat for 20 min. Following this interval, 1 mL of 0.1 N NaOH was put in to neutralize the contents and the volume was corrected to the target using filtered water. The mix was then placed into an HPLC vial and inserted into the HPLC apparatus.

Alkaline deterioration

1 mL of the stock solution was drained into a 10mL graduation container. Upon that, 1mL of 0.1 N NaOH was poured to the container, which was then left at room heat for 20 min. Following this interval, 1mL of 0.1 N HCl was put in to neutralize the contents and the volume was corrected to the target using filtered water. The mix was then placed into an HPLC vial and inserted into an HPLC vial and inserted into the HPLC apparatus.

Oxidative deterioration

1 mL of the stock solution was funnelled into a 10 mL flask that was volumetric. After applying 1 mL of 0.1% H_2O_2, the flask was left at room temperature for 20 min. Following that, a diluent was used to increase the volume to the desired level. The mix was then placed into an HPLC vial and inserted into the HPLC apparatus.

Thermal deterioration

The normal medication mix was heated in an oven at 105°C for 6 hr. The way forward was diluted to 10 µg/mL. Following diluted form, the mixture was then moved to an HPLC vial and put forward into the HPLC apparatus.

Photolytic deterioration

Expose a 100 µg/mL remedy to UV light. The pitcher was set in a UV chamber for 7 days in a photo stability environment. The solution was diminished to 10 µg/mL. Then set in an HPLC vial and run.

Neutral deterioration

Stress simulations under neutral settings involved refluxing the medication in water for 6 hr at 60°C.The mix was diminished to 10 µg/mL. Then set in an HPLC vial and run.

DISCUSSION

Statistical analysis for optimization

A full factorial framework with 20 testing circumstances was used, with nine requirements drawn up by the computational

plan and six repeats at the domain's center. The coded polynomial equations for each model were as follows:

$$\text{Finerenone RT1}=+1.88-0.15\times A-0.04\times B-0.04\times C+0.0062\times AB+0.02\times AC+0.02\times BC \quad (1)$$

$$\text{NTP}=+4402.05-108.98\times A-12.37\times B+15.09\times C-217.50\times AB+97.5\times AC-58.75\times BC-278.43A2-510.72B2-120.57C2$$

Where A is the flow rate, B is the mobile phase content (percentage), and C is column temperature. Figure 1 shows the main impact terms, A, B, and C, as well as the interaction effects of AB, AC, and BC. A visual inspection of the actual vs. expected plot was performed to ascertain perhaps the device itself was suitable. The residuals were regularly distributed along an uninterrupted line, with negligible disperse reflecting a satisfactory fit to the data. The expression written as a set of specified variables allows you to foresee the reaction for given levels of all of them.

The effect of variables on retention Time of Finerenone (Y1).

$$\text{RT1 of Finerenone}=+1.88-0.15\times A-0.04\times B-0.04\times C+0.0062\times AB+0.02\times AC+0.02\times BC \quad (1)$$

The impact of variables on the number of theoretical plates (Y2).

$$\text{NTP}=+4402.05-108.98\times A-12.37\times B+15.09\times C-217.50\times AB+97.5\times AC-58.75\times BC-278.43A2-510.72B2-120.57C2 \quad (2)$$

Using exactly the same chromatographic facts, the study of Finerenone revealed permitted system suitability traits. The chromatograms articulated low tailing (1.2%) and an enormous number of theoretical plates (>8000). The analysis was done productively in 10 min.

Exterior charts for interference, 2D contouring, and 3D responder

The contour plot generated using Design-Expert® software illustrates the impact of two critical factors, percentage of the organic fraction (B) and column heat (C), on the appearance Time (RT1) of the analyte. The gradient of the color-coded regions reflected in Figure 2 variations in retention time, with lighter shades indicating higher RT1 values and darker shades corresponding to shorter retention times. The plot reveals a clear interaction between the percentage of the organic phase and column temperature. Upon an increase in percentage of the organic phase, a noticeable reduction in retention time is observed, suggesting enhanced elution efficiency. Similarly, at elevated column temperatures, retention time decreases further, likely due to reduced viscosity of the mobile phase and increased analyte diffusion rates. The design points marked on the plot indicated the experimental conditions tested, with optimal retention time observed near the central region of the plot. The 3D response surface plot in Figures 3 and 4 demonstrates the relationship between two critical factors-flow rate (A) and organic phase percentage (B)-on the Retention Time (RT1) of the analyte as well as on Number of Theoretical Plates (NTP). The z-axis represents retention time, while the x-axis and y-axis depict variations in flow rate and organic phase percentage, respectively. Upon decrease in retention time the flow rate increased, indicating faster elution of the analyte due to the reduced interaction time between the analyte and stationary phase. Similarly, increasing the percentage of organic portion also resulted in reduced retention time, suggesting improved elution efficiency attributed to increased solvent strength. The combined effect of these two factors is evident from the plot, as regions with

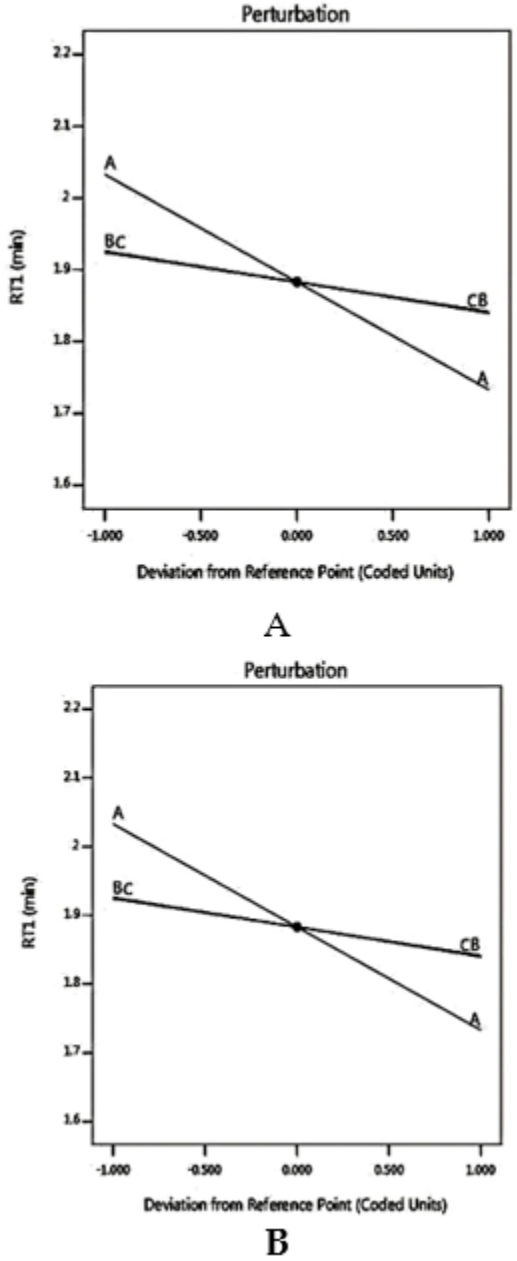


Figure 1: Perturbation plots showing effect of Velocity(A), moving phase fraction(B) and Column heat (C) on Responses: RT1(Y1) NTP(Y2).

higher flow rates and higher organic phase percentages exhibit the shortest retention times.

Validation of constructed Approach

System suitability

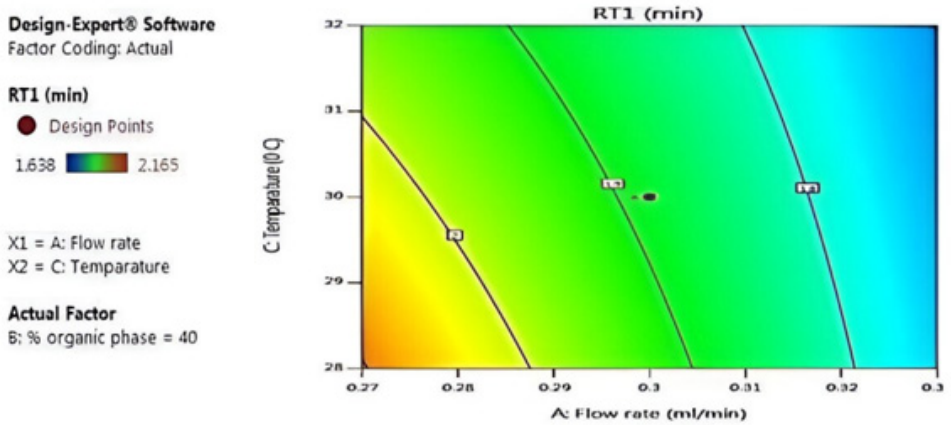
The system suitability results focus the chromatographic system is reliable and robust for the separation and analysis of the substances of interest. The average signal intensity values for the analytes were constant, with Relative Standard Deviations (RSD) of less than 5%, indicating high precision and reproducibility. The retention duration for each analyte was also uniform, with very little change between injections. The Theoretical Plate number (HETP) values were low, indicating effective separation and peak resolution. The asymmetrical factors were near to one, indicating symmetric peak forms. These characteristics are crucial for accurately quantifying and identifying analytes.

Specificity

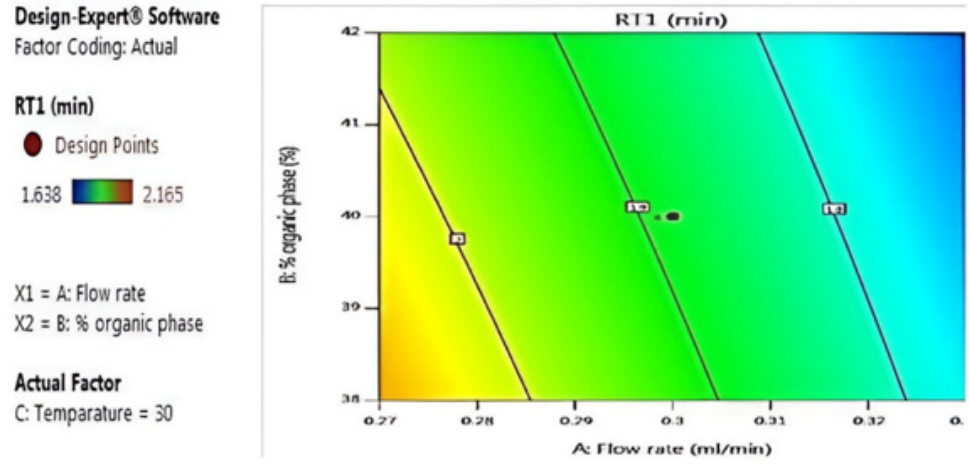
Particularity was examined by placing an empty specimen and the point in time coinciding to the elementary signal was verified. No major impact was noted, supporting the method's specificity.

Linearity

To make sure the assay system's linearity, six conventional solutions were held from 2.5 ppm to 15 ppm were inserted. Calculated inclination seemed 417399, the y-intercept was



A



B

Figure 2: 2D contoured layer visualizations exhibiting the impact of CMPs: (A) fluid velocity; (B) organic fraction (%) on appearance time of Finerenone (RT1).

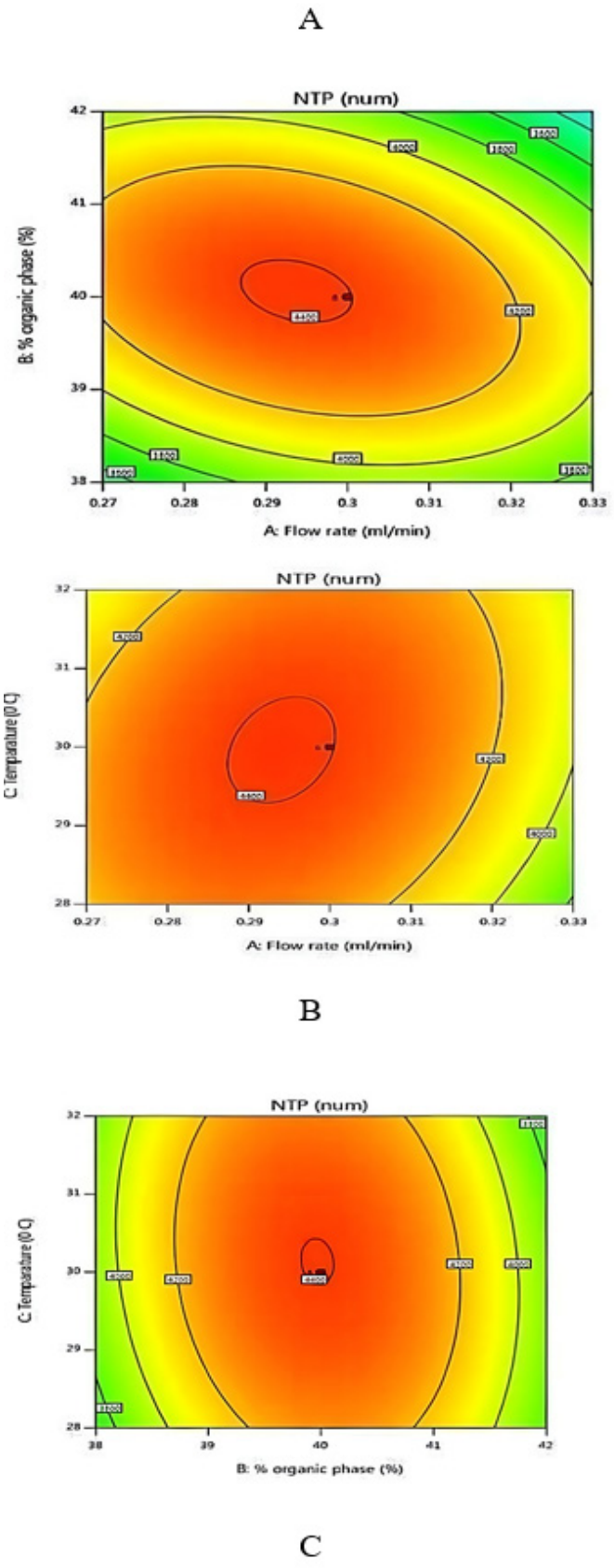


Figure 3: 2D contoured layer visualizations exhibiting the impact of CMPs:(A) fluid velocity; (B) organic fraction (%) and (C) column heat; on Number of Theoretical plates (NTP).

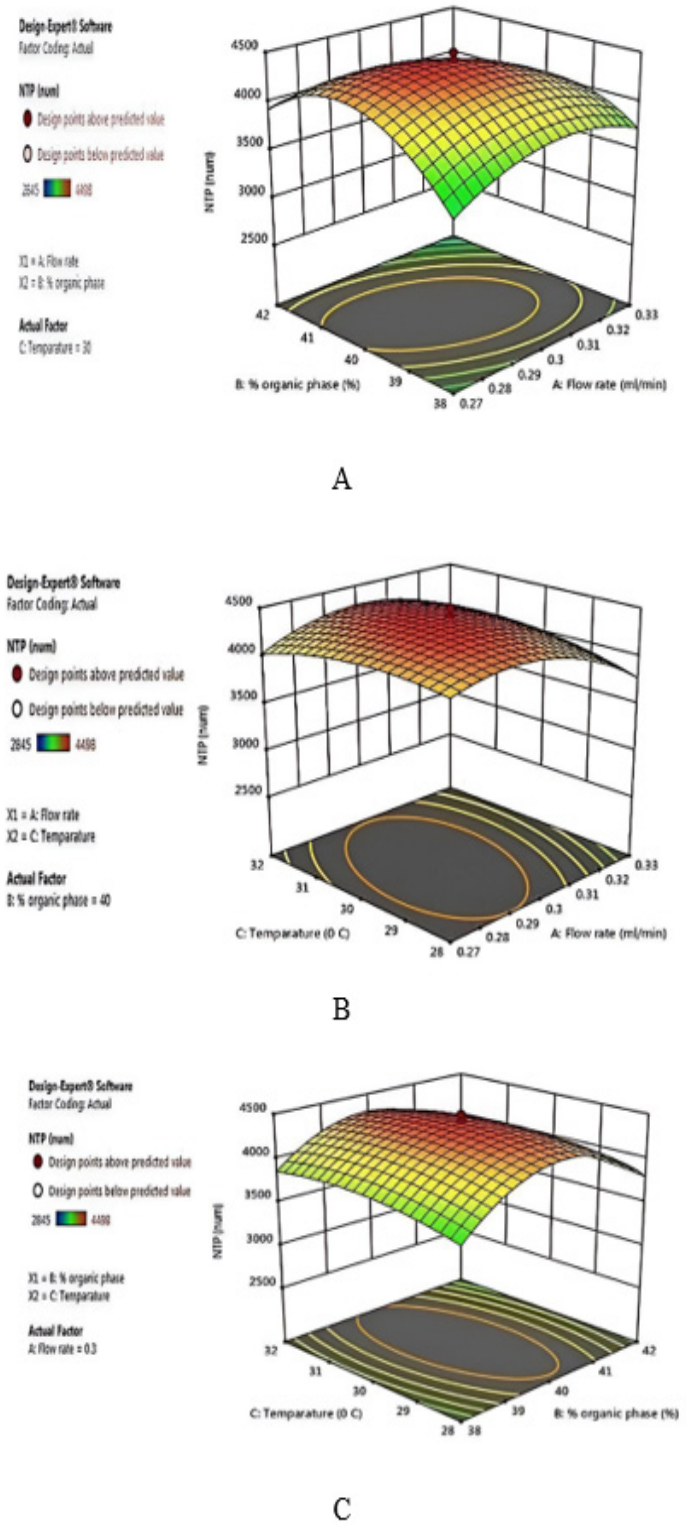


Figure 4: 3D contoured layer visualizations exhibiting the impact of CMPs.: (A) fluid velocity; (B) organic fraction (%) and (C) column heat; on Number of Theoretical plates (NTP).

Table 3: An ANOVA for the responses of 2FI and quadratic models.

Source	Degrees of freedom		*f*-value	
	@	NTP	$	NTP
Model	6	9	79.29	14.43
A	1	1	395.65	4.05
B	1	1	29.87	0.0522
C	1	1	33.05	0.0777
AB	1	1	0.4025	9.45
AC	1	1	7.65	1.90
BC	1	1	9.12	0.6898
A^2		1		27.91
B^2		1		93.90
C^2		1		5.23

@:RT of F- Retention time of Finerenone;$: NTP-Number of Theoretical Plates.

Table 4: Linearity metrics of Finerenone.

Concentration (ppm)	Area
0	0
0.25	109220
0.5	213566
0.75	315302
1.0	426476
1.25	523312
1.5	626882
Linearity Range	2.5-15.0#
Slope	417399
Intercept	3350.3
Correlation Coefficient R^2	0.999
LOD	0.041#
LOQ	0.123#

in µg/mL.

3350.3, and the correlation value was 0.999. The metrics can be observed in Table 4.

Precision

The data set reflects signal intensity measurements over two consecutive days, focusing on absolute and relative deviations from standard values. On the day itself, the signal intensities appear higher (426089) compared to the subsequent day (418300), suggesting a general trend of decline. This pattern is further supported by the averages (a1=426056 vs. a2=418184), indicating consistently higher values on the first day. The deviations from the standard are notable, with b1 (1627.3) on the first day being smaller than b2 (2255.6) on the second day, implying greater variability in signal intensity the day after. Additionally, the percentage deviations (c1=0.4%, c2=0.5%) highlight relatively minor but consistent increases in variability on the following day. This analysis suggests that the signal may stabilize or peak on the day of observation and become more variable over time, potentially influenced by environmental or systemic factors.

Accuracy

The proof of concept brought about in Table 5 portrays a high level of accuracy and consistency in the recovery of the target compound, with recovery percentages hovering around 99%. The consistency across all three sets (5^x, 10^y, and 15^z) reinforces the reliability of the technique employed. However, slight variations in recovery (99.23% in the 15^z set) should be monitored to ensure long-term precision and to identify any potential sources of error, although these variations are minor and well within acceptable limits for most scientific procedures.

LOD and LOQ

Identification, measurement thresholds for drug of choice in this approach seemed 0.041 µg/mL and 0.123 µg/mL, respective.

Robustness

Switching the organic mix of the portable phase led in an RSD of 0.23%, arguing strong repeatability with little sensitivity to chemical makeup variations. Differences in the pace of flow culminated 0.95%, demonstrating the method's capacity to maintain precise despite slight fluctuations. Changing the detecting spectrum ended in 0.67%, demonstrating the method's dependability even with minor changes.

Forced degradation

The drug's breakdown was investigated under various stress situations, revealing distinct levels of degradation and purity profiles. The maximum degradation, 4.77%, occurred in acidic circumstances, with a purity angle of 0.236 falling below the threshold (0.372), indicating considerable degradation and purity loss. Alkali degradation resulted in a mild 3.15% degradation,

Table 5: Trueness data for Finerenone.

	Target reached (µg/mL)	%Reacquired	On a whole %Reacquired
5[x]	4.98	99.61	99.41%
	4.97	99.38	99.51%
	4.96	99.23	99.49%
10[y]	9.94	99.41	
	9.93	99.32	
	9.98	99.79	
15[z]	14.92	99.45	
	14.97	99.78	
	14.88	99.23	

[x] Half Quantity of API spiked in µg/mL; [y]Full quantity Spiked in µg/mL; [z]three-forth quantity spiked in µg/mL.

with a purity angle of 0.187, significantly less than the threshold (0.380), indicating a moderate effect on drug integrity. Oxidative degradation produced 2.21% degradation and a purity angle of 0.306, which is less than the purity threshold (0.376), indicating some deterioration, albeit less severe. Thermal deterioration had the least influence, with only 1.26% degradation and a purity angle of 0.173, which is less than the criterion (0.370), indicating minor consequences under heat stress. UV degradation caused the least degradation (0.90%), with a purity angle of 0.176, somewhat lower than the threshold (0.374), demonstrating the drug's resistance to photochemical degradation. Water degradation was 1.22%, with a purity angle of 0.174, which is slightly lower than the threshold (0.376), indicating mild hydrolytic breakdown. Overall, the medicine was most sensitive to acidic circumstances and least sensitive to UV radiation exposure, indicating that it is very stable under most settings, except for acidic environments. These findings provide critical information about the drug's stability, which influences storage settings and shelf-life considerations.

CONCLUSION

The perfected chromatographic technique for Finerenone shows high system ability to adapt, linearity, precision, and accuracy, rendering it appropriate for routine analysis. The approach accomplished a high R^2 value of 0.999 for linearity between 25% and 150% levels, with a recovery rate of 99.47%. The precision, as measured by method precision (0.4) and intermediate precision (0.5), was adequate. Furthermore, the approach demonstrated robustness in degradation studies where purity thresholds exceeded purity angles. The assay of the marketed product yielded 99.62% Finerenone, demonstrating the method's reliability and efficiency. Incorporating Quality by Design (QbD) principles enhanced operational efficiency, product integrity, legal compliance, and drug safety.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

mL: Millilitre; **g:** Gram; **mg:** Milligram; **µg:** Microgram; **NMT:** Not More Than; **RSD:** Relative Standard Deviation; **VF:** Volumetric Flask; **mcg:** Microgram.

SUMMARY

The study tackles the substantial need for accurate and reliable analytical tools to assure Finerenone's efficacy and safety. The method involves employing a portable phase of 0.01N potassium dihydrogen orthophosphate, acetonitrile (65:35 ratio) when flow velocity was 1 mL/min, with recognition at 230 nm on an Agilent C18 column that operates beneath isocratic settings. CCD optimization resulted in a Method Operable Design Region (MODR), which was validated using International Council for Harmonisation (ICH) requirements for linearity, precision, accuracy, robustness, and sensitivity. The findings disclosed exceptional linearity (R^2=0.999) spanning an amount range of 2.5-15.0 µg/mL. Rates of recuperation went from 99.23% to 99.79%, with predictable RSD readings of 0.4% (day itself) and 0.5% (day after). deterioration experiments revealed highest under acidic circumstances (4.77%) and lowest over UV contact (0.90%), indicating substantial durability. The validated think about made sure precision, sensitivity, and robustness, making it worthy of ongoing testing of Finerenone in pharmaceutical formulations, as well as research and quality control applications. Bringing together QbD principles increased operational efficiency

and product integrity while also assuring legal compliance and improved protection of medicines. Effect on elution efficiency was confirmed by the 3D plot, retention duration lead to raise in flow rates and organic phase percentages.

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