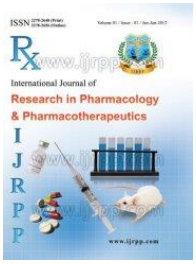
International Journal of Research in Pharmacology & Pharmacotherapeutics



ISSN Print: 2278-2648 IJRPP |Vol.7 | Issue 3 | July - Sep - 2018
ISSN Online: 2278-2656 Journal Home page: www.ijrpp.com

Research article Open Access

Quality by design approach to analytical method development for simultaneous estimation of ibuprofen and famotidine in their combined dosage form by RP-HPLC method

Jayaprakash J*, Vijay Amirtharaj R and Senthil Kumar N

J.K.K.MMRF, Annai JKK Sampoorani Ammal College of Pharmacy, Komarapalayam. Tamil Nadu, India

***Corresponding author: Jayaprakash J**

ABSTRACT

This article aims to explain the steps for application of quality by design (QbD) concept to analytical method development and validation, by using an example of simultaneous determination of Famotidine and Ibuprofen in its pharmaceutical dosage form by RP-HPLC. By using QbD tools, enable earlier understanding and identification of variables affecting the method performance. Fractional design and Central composite design were used for screening the variables and optimization of chromatographic conditions with building the design space employing a three factor three level Box– Behnken design (BBD) using ANOVA software. A QbD guide is described from identification of analytical target profile to definition of control strategy. The optimized chromatographic method was performed using 0.01M ammonium acetate buffer (pH 4): methanol: acetonitrile (50:50, 60:40, 40:60, 35:65 % v/v) as mobile phase at a flow rate 1.0mL/ min and UV detection at 225 nm.

Keywords: Ibuprofen; Famotidine; Quality by design; Design space; Central composite design; RP- HPLC; Validation

INTRODUCTION

Quality by Design (QbD) approach has been introduced by FDA for the pharmaceutical development to ensure a predefined product quality. Application of Quality by Design concept to the analytical method development leads to a more robust method. ICH guidelines Q8 (R2) defines QbD as "A systematic approach to development that begins with predefined objectives and emphasizes product and process control, based on sound science and quality risk management". In this approach potential method variables that affects the overall quality of method are defined, their interactions are studied, control strategy is implemented and finally the method is continually monitored [1-2].

Several HPLC methods for the individual estimation of Famotidine and Ibuprofen are available in the literature. [3-6] Although, there are scanty number of works describing the methods for the simultaneous determination of these drugs in combination with other drugs in pharmaceutical mixtures. However, there seems to be no reports

concerning methods for the determination of Famotidine and Ibuprofen using experimental design. Therefore, in the present work, a new HPLC method was developed, optimized and validated for the determination of these drugs and its impurity in formulations.

MATERIALS AND METHODS

Selection of wavelength

The sensitivity of HPLC depends upon proper selection of wavelength of detection. PDA detector was used to determine the proper wavelength of Ibuprofen and Famotidine 225nm wavelength was found. It was selected for the analysis.

Preparation of working standard solution

About 3.32mg of Famotidine and 100mg of Ibuprofen were separately accurately weighed and transferred into a 100ml volumetric flask. 10 ml of methanol was added and sonicate for 15 minutes. The volume was made 100ml with methanol to obtain final concentration of 33.2µg/ml for Famotidine and 1000µg / ml for Ibuprofen. From this, the 5 ml of the solution was pipetted out and transferred into 25 ml volumetric flask. Then it was diluted to 25 ml with methanol as diluent. The final concentrations of Famotidine and Ibuprofen were 6.64µg/ml and 200µg/ml.

Preparation of Sample solution

Ten tablets Duexis (Ibuprofen 800mg and famotidine 26.6 mg) were accurately weighed and crushed into fine powder. An amount of tablet powder equivalent to 100 mg of ibuprofen (3.32mg of Famotidine) was transferred into a 100ml volumetric flask. 10 ml of methanol was added, shaken for 5 minutes on a rotatory shaker and then sonicated for 10 min with intermediate shaking. After that the volume was finally made upto the mark with methanol. Then it was filtered through a whatmann filter paper No 41. So the concentrations were 33.2µg/ml for Famotidine and 1000µg /ml for Ibuprofen. 5ml of the above solution was pipetted out and it was transferred into 25ml volumetric flask and diluted to 25 ml with methanol as diluent. Then the final concentrations of Famotidine and Ibuprofen were 6.64µg/ml and 200µg/ml.

Method optimization

The optimization of mobile phase condition was performed as per the experimental design employing a three factor three level Box– Behnken design (BBD) using ANOVA software (Stat-Ease Inc., Minneapolis, USA) by selecting the methanol volume (A), buffer strength (B) and buffer pH (C) as independent variables, while the Capacity factor (K_1), Resolution ($Rs_{1,2}$) and retention time (tR_1) as responses. Response surface analyses were carried out to identify the effect of different independent variables on the observed responses.

Table 1 illustrates total 20 experimental runs obtained from Box Behnken design with their observed responses and predicted responses. The responses were statistically evaluated using the ANOVA procedure. Further, the optimum condition was selected by the numerical optimization procedure using the desirability function. BBD has the advantage of optimization for experiments by using 3k-factorial design (where k=1, 2, 3 . . .) having at least three dependent variables or factors and more than one response as compared to other experimental designs such as central composite design (CCD) and fractional factorial design (FFD) [7 -9]. For an experimental design with the three factors, including linear, quadratic and cross terms, the model can be expressed as

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \beta_{11} X_1^2 + \beta_{22} X_2^2 + \beta_{33} X_3^2$$

Where Y is the response to be modeled, β is the regression coefficient β0 is constant; β1, β2, β3 are linear coefficients, β12, β13, β23 are interaction coefficients between the three factors, β11, β22, β33 are quadratic coefficients computed from the observed experimental values of Y from experimental runs and A, B and C are the coded levels of independent variables high (+), low (−) and center point (0). Statistical parameters obtained from ANOVA for the reduced models are given in table 2.

Method validation [10]

Developed method was validated according to ICH Q2 (R1) guidelines.

System suitability

System suitability tests are an integral part of any chromatographic analysis method which is used to verify reproducibility of the chromatographic system.

Five replicate injections of the drug solution at the concentration of 6.64µg/ml for Famotidine and 200µg/ml for Ibuprofen were used. 20µl of standard and sample solutions were injected and the chromatograms were recorded.

Specificity and selectivity

The specificity of the method was checked by comparing the chromatograms obtained from standard, sample and the corresponding placebo. The retention time of the standard and the drugs from sample were identical. This confirmed the specificity of the method.

Linearity

From standard solution of Famotidine 3.32 – 9.97 µg/ml and Ibuprofen 100-300 µg / ml were prepared. Regression equation, Correlation coefficient, Slope and Intercept were calculated. **LOD and LOQ**

LOD and LOQ of developed methods were calculated from the equations given by ICH guidelines.

LOD= 3.3*σ/S

LOQ=10*σ/S

Where, σ= standard deviation of intercept

S=slope of calibration curve

Content Estimation (Assay)

6.64µg/ml for Famotidine and 200µg/ml for ibuprofen of standard and sample solutions were prepared separately from the standard and sample stock solutions (33.2µg/ml for Famotidine and 1000µg/ml for Ibuprofen). 20µl of each standard and sample solution were injected and the chromatograms were recorded.

Precision

Precision study was conducted by injecting standard solutions of Ibuprofen and Famotidine five times at a concentration of 6.64µg/ml for Famotidine and 200µg/ml for Ibuprofen. The peak area and the chromatograms were recorded. The percentage relative standard deviation (%RSD) values of pure drugs were calculated.

Accuracy

Accuracy of method was assessed by recovery study from formulation at three level of standard addition (50%, 75% and 100%) in triplicate. % recovery within 98-102% with low standard deviation justified the accuracy of developed method.

Robustness

Robustness of method was determined in the form of Standard Deviation of retention time by small deliberate changes in flow rate, pH of buffer solution and detection wavelength.

Table 1: Central composite arrangement and responses

Run	Type	MeOH concentration A (%v/v)	Buffer strength B (mM)	Buffer (pH) C	Capacity factor (K_1)	Resolution ($Rs_{1,2}$)	Retention time (tR_1)
1	Fact	30.00	25.00	4.50	0.91	11.68	8.63
4	Fact	30.00	23.00	3.50	0.92	8.86	6.56
5	Fact	40.00	27.00	3.50	0.96	8.98	4.30
9	Fact	30.00	27.00	3.50	0.82	10.88	6.56
12	Fact	40.00	27.00	4.50	0.80	6.94	4.30
13	Fact	40.00	23.00	4.50	0.80	6.98	4.25
15	Fact	40.00	23.00	4.50	0.88	8.12	4.25
17	Fact	30.00	27.00	4.50	0.76	10.86	6.56
3	Axial	35.00	25.00	3.16	0.78	6.90	4.25
7	Axial	35.00	25.00	4.84	0.78	10.46	4.10
14	Axial	26.59	25.00	4.00	0.96	4.68	3.55
18	Axial	35.00	28.36	4.00	0.86	11.46	4.36
19	Axial	35.00	21.64	4.00	0.85	14.60	4.45
17	Axial	43.41	25.00	4.00	0.94	4.92	8.50
2	Center	35.00	25.00	4.00	1.06	11.68	4.36

6	Center	35.00	25.00	4.00	1.04	11.65	4.25
8	Center	35.00	25.00	4.00	0.99	11.69	4.33
10	Center	35.00	25.00	4.00	1.06	11.62	4.30
11	Center	35.00	25.00	4.00	1.06	11.60	4.34
16	Center	35.00	25.00	4.00	0.99	11.59	4.30

Table 2: Reduced Response Surface Models and Statistical Parameters obtained from ANOVA

Responses	Regression model	Adjusted R^2	Model p value	% C.V	Adequate Precision
K_1	+1.06-2.66E-004A-0.011B-0.023 C+0.041AB-	0.9110	<0.0001	3.61	14.975
$Rs_{1,2}$	$0.037A^2$-$0.071B^2$-$0.098C^2$	0.7209	<0.0001	5.19	11.601
tR_1	+12.14-0.79A+0.41C-2.39A^2-1.01 C^2 +4.42+1.47A-0.16B+0.27AB+0.74 A^2+0.17B^2- 2.87 AB^2	0.8455	<0.0001	8.52	15.082

Table 3: System suitability parameters

Parameters	Compound	
	Famotidine	Ibuprofen
Capacity factor (K')	1.14	1.03
Retention time (Rt) in min	4.45	8.68
Theoretical plates (N)	10980.9	10452.7
Resolution (Rs)	-	2.90

Table 4: Data for Linearity

Parameters	Famotidine	Ibuprofen
Range (µg/ml)	3.32-9.92	100-300
Y=mx + c	Y=1583.3X+157.11	Y=15.118X+34.31
Regression coefficient	r^2= 0.9993	r^2= 0.9992
Slope (m)	1583.3	15.118
Intercept (c)	157.11	34.31
LOD (µg/ml)	0.008	0.206
LOQ (µg/ml)	0.025	0.624

Table 5: Results for Linearity Data

Concentration of Famotidine (µg/mL)	Peak Area	Concentration of Ibuprofen (µg/mL)	Peak Area
3.32	5623.1	100	1552.23
4.98	8056.23	150	2346.77
6.65	10645.23	200	3115.45
8.31	13456.12	250	3745.23
9.97	15774.01	300	4563.78
Correlation coefficient - 0.9993		Correlation coefficient - 0.9992	

Table 6: Data for content estimation

Drugs	Standard	peak area	Sample	peak area	Mean (%)	SD	RSD (%)
Famotidine							
1	10980.9		10842.9				
2	10812.7		10855.7		100.23	0.8804	0.8484
3	10632.2		10642.2				
Average	10808.6		10780.2				
Ibuprofen							
1	3019.33		3015.42				
2	3042.54		3045.43				
3	3024.23		3020.22		99.94	0.1302	0.1301
Average	3028.70		3027.02				

Table 7: Data for Precision

	Area (μV^2 sec)	
Injection	**Famotidine**	**Ibuprofen**
1	10640.7	3020.44
2	10802.1	3044.56
3	10810.7	3056.78
4	10925.2	3078.23
5	10808.4	3096.44
SD	90.78	29.459
%RSD	0.8407	0.9629

Table 8: Accuracy Results

Compound	Concentration (%)	Amount added (µg/ml)	Amount found (µg/ml)	Mean Recovery (%)	SD	RSD (%)
Famotidine	50	3.32	3.35			
	75	4.98	5.00	99.26	0.2886	0.2907
	100	6.64	6.70			
Ibuprofen	50	100.0	100.13			
	75	150.0	150.23	99.98	0.1457	0.1469
	100	200.0	200.11			

Table 9: Robustness study

Peak name	Parameter	USP plate count	Tailing factor
Famotidine	MeOH: Ammonium acetate buffer pH 3.94 (29.26: 70.74% v/v)	10642.3	1.08
	Actual (32.26: 67.74 % v/v)	10982.9	1.03
	MeOH: Ammonium acetate buffer pH 3.94 (35.26 : 64.74 %v/v)	10655.3	1.11
	Flow rate (0.8 ml/min)		
	Actual (1.0 ml/min)	10725.5	1.05
	Flow rate (1.2 ml/min	10982.9	1.03
		10654.5	0.99

Ibuprofen	MeOH: Ammonium acetate buffer pH 3.94 (29.26: 70.74% v/v)	3044.59	1.10
	Actual (32.26: 67.74 % v/v)	3056.12	1.13
	MeOH: Ammonium acetate buffer pH 3.94 (35.26 : 64.74 %v/v)	3010.45	1.08
	Flow rate (0.8 ml/min)		
	Actual (1.0 ml/min)		
	Flow rate (1.2 ml/min)	3090.75	1.11
		3076.45	1.05
		3043.10	1.12

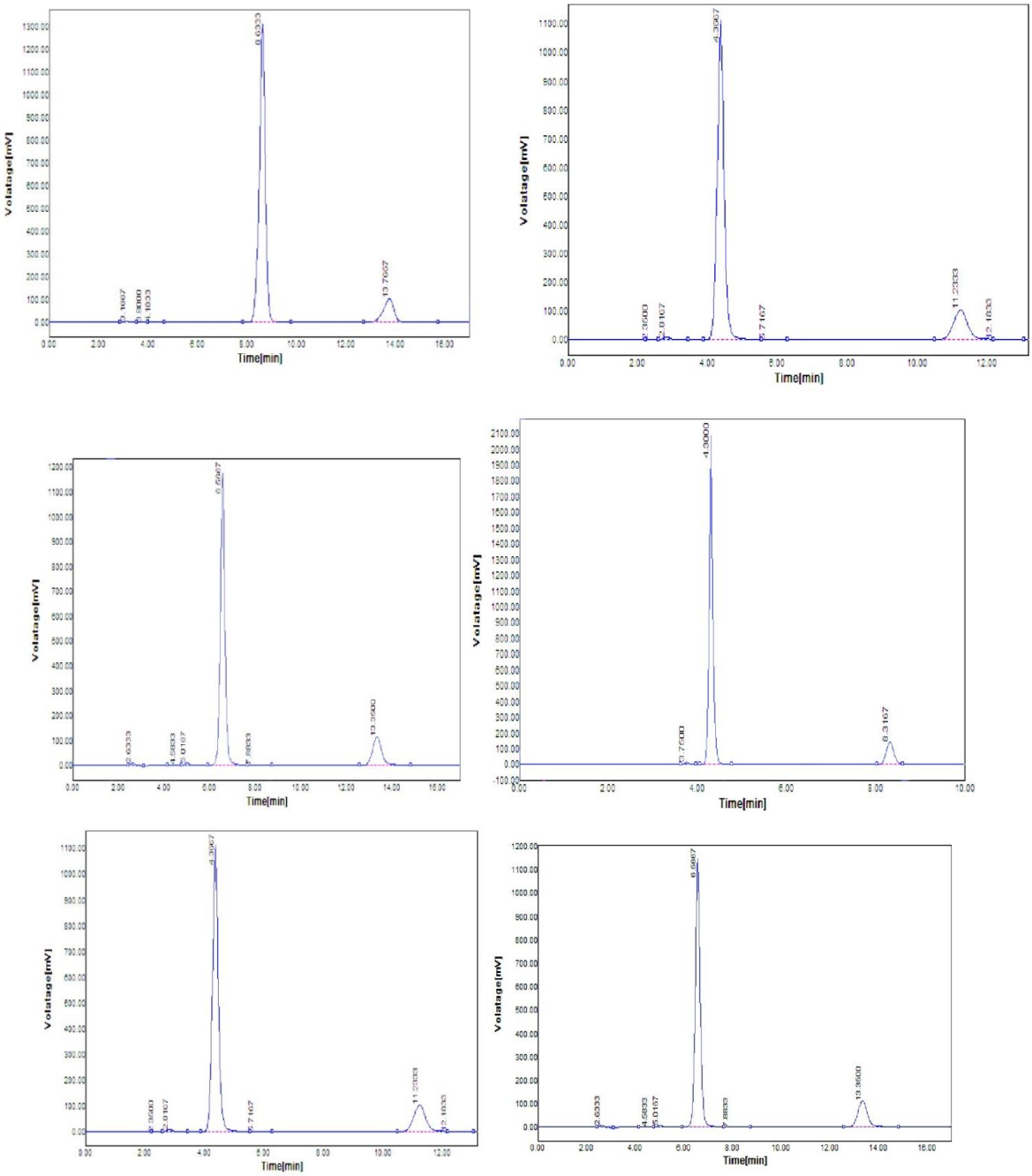


Figure 1: CCD generated trial runs and their respective chromatograms

RESULTS AND DISCUSSION

The developed method was optimized using statistical software Minitab. Three factors viz flow rate, percentage of buffer in the mobile phase and pH of the buffer were found to have an effect on the peak area, retention time, asymmetry and resolution between both the drugs. The optimized chromatographic method was performed using 0.01M ammonium acetate buffer (pH 4): methanol: acetonitrile (50:50, 60:40, 40:60, 35:65 % v/v) as mobile phase at a flow rate 1.0mL/ min and UV detection at 225 nm. Among these, the mobile phase composition ratio 35:65 % v/v resulted in a quality separation in terms of peak symmetry, optimum resolution and reasonable run time. The chromatogram recorded by running the chromatographic separation at the optimized condition is depicted in Fig. 1.

Method validation

Method was successfully validated according to ICH Q2 (R1) guidelines. Beer's law was obeyed in range of 3.32-9.97µg / ml for Famotidine, 100-300 µg / ml for Ibuprofen. It showed 0.9993 and 0.9992 r^2 values for Famotidine and Ibuprofen respectively, indicates good linearity. Intraday and inter day precision values were indicated as %RSD and %RSD below 2 showed good precision of developed method. Low LOD and LOQ values indicate sensitivity of proposed method. Accuracy of method was investigated by means of recovery study. Results obtained in range of 98 % to 102 % shows good accuracy of developed method. Recovery study data are shown in Table 8. Developed method was also applied to tablet dosage form. %Assay obtained for two drugs were 99.94 to 100.23 % for Famotidine and Ibuprofen respectively. The % RSD values were found to be 0.8484 and 0.1301 for Famotidine and Ibuprofen respectively. The results were shown in table 6. Robustness study was also performed and low SD indicates that method is robust enough that small changes in method parameter do not affect method responses (Table-9). System suitability parameters were also studied and reported in Table 3.

CONCLUSION

An innovative Quality by Design approach has been applied for development of RP-HPLC method for simultaneous estimation of Famotidine and Ibuprofen in their combined dosage form. For this central composite design which is a response surface methodology was adapted to spot out the significant impact of the independent variables such as % organic phase and the flow rate each at triplet levels on the chromatographic responses. The chromatographic responses such at the resolution, theoretical plates, tailing factor and total analysis time were simultaneously optimized with the backing of design of experiments methodology. The method was fully validated in compliance with ICH guidelines and a robustness study was performed by varying three chromatographic parameters at three levels. So, RP-HPLC method was successfully developed and validated for simultaneous estimation of Famotidine and Ibuprofen in their combined dosage form.

REFERENCES

[1]. Syed Salman Lateef and Vinayak AK. Quality-by-Design Approach to Stability Indicating Method Development for Linagliptin Drug Product. *Agilent Technologies Inc.* 2014:5991-3834 EN.

[2]. Ramalingam Peraman, Kalva Bhadraya, and Yiragamreddy Padmanabha Reddy. Analytical Quality by Design: A Tool for Regulatory Flexibility and Robust Analytics, *Int. jou. ana. chem*. 2015: Article ID 868727.

[3]. Dimal A. Shah , Dixita J. Suthar , Chirag D. Nagda , Usman K. hhalotiya, Kashyap K. Bhatt. Development and Validation of HPTLC method for estimation of ibuprofen and famotidine in pharmaceutical dosage form. *J Liq Chroma &Rel Technologies*.37(4), 2013.

[4]. HalaE.Zaazaa, EmanS.Elzanfaly, AyaT.Soudi, MaissaY.Salem. Application of the ratio difference spectrophotometry to the determination of ibuprofen and famotidine in their combined dosage form; Comparison with previously published spectrophotometric methods SpectrochimicaActa Part A: Molecular and Biomolecular Spectroscopy. 143, 2015, 251-255.

[5]. Allabasha Mumtaz, Iffath Rizwana. Simultaneous estimation of paracetamol, ibuprofen and famotidine by using RP-HPLC method. *Indo American J Pharma Res*. 5(09), 2015, 2964-2981.

[6]. Sandhyarani G, Sarangapani M. Method development and validation for the simultaneous estimation of

Ibuprofen and Famotidine. *Int J Pharma Biol Sci*. 7(2), 2017, 56-71.

[7]. Rozet E, Ziemons E, Marini RD, Boulanger B, Hubert P. Quality by design compliant analytical method validation. *Anal Chem*. 84, 2012, 106-112.

[8]. Lionberger RA, Lee SL, Lee L, Raw A, Yu LX. Quality by design: concepts for ANDAs. *AAPS J*. 10, 2008, 268-276.

[9]. Nethercote P, Ermer J. Quality by design for analytical methods: implications for method validation and transfer. *Pharmaceutical Technology*. 36, 2012, 74–79.

[10]. S. Gorinstein, D. Huang, H. Leontowicz, M. Leontowicz, K. Yamamoto, R. Soliva-Fortuny, O. Martin Belloso, A. L. Martinez Ayala, and S. Trakhtenberg, Determination of naringin and hesperidin in citrus fruits by High Performance Liquid Chromatography. The antioxidant potential of Citrus fruits, Acta chromatographica. 17, 2006.