EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 8
Issue: 12
261-270
Year: 2021

DEVELOPMENT AND VALIDATION OF STABILITY INDICATING RP-HPLC METHOD FOR THE SIMULTANEOUS ESTIMATION OF MONTELUKAST AND BAMBUTEROL

CTNR Rajeswari[1], Dr. C. Parthiban*[2] and Dr. M. Sudhakar[3]

[1]M.Pharmacy, Department of Pharmaceutical Analysis, Malla Reddy College of Pharmacy, Dhulapally, Hyderabad - 500014, Telangana.
[2]Professor and HOD, Department of Pharmaceutical Analysis, Malla Reddy College of Pharmacy, Dhulapally, Hyderabad -500014, Telangana.
[3]Principal and Professor, Department of Pharmaceutical Analysis, Malla Reddy College of Pharmacy, Dhulapally, Hyderabad -500014, Telangana.

***Corresponding Author: Dr. C. Parthiban**
Professor and HOD, Department of Pharmaceutical Analysis, Malla Reddy College of Pharmacy, Dhulapally, Hyderabad -500014, Telangana.

Article Received on 08/10/2021 **Article Revised on 29/10/2021** **Article Accepted on 19/11/2021**

ABSTRACT
A successful attempt has been made to develop simple, accurate, economic and rapid methods for the estimation of Montelukast and bambuterol in tablet Formulation by RP-HPLC and to validate the methods, as per the guidelines given by ICH requirements. Disodium hydrogen phosphate and Acetonirile were used as mobile phase and C18 columnn and UV detector were employed. Run time was found out to be 5 minutes; injection volume was 10µL. Montelukast and Bambuterol were eluted at 2.154min and 3.057 min respectively with good resolution.

KEYWORDS: Montelukast, Bambuterol, RP-HPLC,ICH and Validation.

INTRODUCTION

Montelukast

Montelukast is chemically known as 2-[1-[[(1R)-1-[3-[(E)-2-(7-chloroquinolin-2 yl) ethenyl] phenyl]-3-[2-(2-hydroxypropan-2yl) phenyl] propyl] sulfanylmethyl] cyclopropyl] acetic acid. Chemically Montelukast belongs to the class of organic compounds known as 1,3-diarylpropanoids (linear). Being Leukotriene receptor antagonist, Montelukast is used as an alternative to anti-inflammatory medications in the management and chronic treatment of asthma and exercise-induced bronchospasm (EIB). The chemical structure of MON is shown below

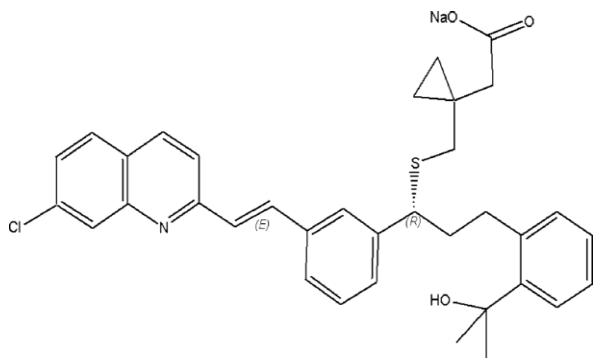

Fig. 1: Structure of Montelukast.

Bambuterol

Bambuterol is chemically known as 3-[2-(tert-butylamino)-1-hydroxyethyloxy] phenyl N,Ndimethyl carbamtae. Bambuterol is used or the prevention and reversal of bronchospasm in patients 12 years of age and older with asthma and reversible bronchospasm associated with bronchitis and emphysema. 3-[2-(tert-butylamino)-1-hydroxyethyl]-[(dimethylcarbamoyl)oxy]phenylN,Ndimethylcarbamae

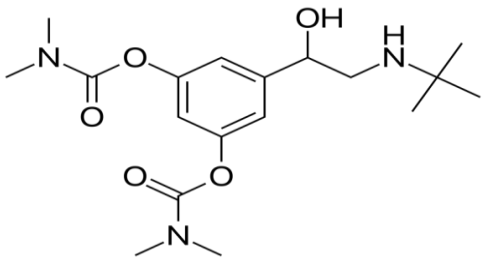

Fig. 2: Structure of Bambuterol.

The wide variety of challenges is encountered while developing the methods for different drugs depending on its nature and properties. This along with the importance of achieving the selectivity, speed, cost, simplicity, sensitivity, reproducibility and accuracy of results gives an opportunity for researchers to come out with solution to address the challenges in getting the new methods of

analysis to be adopted by the pharmaceutical industry and chemical laboratories. Different physico-chemical methods are used to study the physical phenomenon that occurs as a result of chemical reactions. Among the physico-chemical methods, the most important are optical (refractometry, polarimetry, emission and fluorescence methods of analysis), photometry (photocolorimetry and spectrophotometry covering UV-Visible, IR Spectroscopy and nepheloturbidimetry) and chromatographic (column, paper, thin layer, gas liquid and high performance liquid chromatography) methods. Methods such as nuclear magnetic resonance (NMR) and para magnetic resonance (PMR) are becoming more and more popular. The combination of mass spectroscopy (MS) with gas chromatography is one of the most powerful tools available. The chemical methods include the gravimetric and volumetric procedures which are based on complex formation; acid-base, precipitation and redox reactions. Titrations in non-aqueous media and complexometric have also been used in pharmaceutical analysis. The number of new drugs is constantly growing. This requires new methods for controlling their quality. The selected method should be precise and selective.

METHOD DEVELOPMENT

Materials and Methods

Materials

- Montelukast and Bambuterol pure drugs (API) received from Aurobindo pharma ltd.
- Combination Montelukast and Bambuterol (Montek Plus Tab).
- Distilled water, Acetonitrile, Phosphate buffer, Methanol, Potassium dihydrogen ortho phosphate buffer, Ortho-phosphoric acid. All the above chemicals and solvents are from Rankem.

Equipment Used

- Electronics Balance-Denver
- pH meter -BVK enterprises, India
- Ultrasonicator-BVK enterprises
- Waters HPLC System series with Binary pumps, Photo Diode array detector and manual sampler integrated with empower software
- Lab india UV double beam spectrophotometer with UV win5 software was used for measuring absorbances of Montelukast and Bambuterol solutions.

Diluent: Based up on the solubility of the drugs, Diluent was selected, Acetonitrile and Water taken in the ratio of 50:50

Preparation of Standard stock solutions: Accurately weighed 5mg of Montelukast and 5mg of Bambuterol and transferred to 50ml volumetric flask. And ¾ th of diluents was added to these flasks and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (100µg/ml of Montelukast and 100µg/ml of Bambuterol).

Preparation of Standard working solutions (100% solution): 1ml from each stock solution was pipetted out and taken into a 10ml volumetric flask and made up with diluent. (10µg/ml Montelukast of and 10µg/ml of Bambuterol).

Preparation of Sample stock solutions: 5 equivalent tablets weighed and transferred to 100 ml volumetric flask, to this 5 ml of acetonitrile was added and sonicated. Volume was made upto 100 ml with diluents and filtered through 0.45 µm or finer porosity membrane filter (100µg/ml of Montelukast and 100µg/ml of Bambuterol)

Preparation of Sample working solutions (100% solution): 1ml of filtered sample stock solution was transferred to 10ml volumetric flask and made up with diluent. (10µg/ml of Montelukast and 10µg/ml of Bambuterol).

Preparation of buffer

Buffer: 0.01N Sodium hydrogen phosphate

Accurately weighed 1.42gm of Sodium hydrogen phosphate in a 1000ml of Volumetric flask add about 900ml of milli-Q water added and degas to sonicate and finally make up the volume with water then added 1ml of Triethylamine then PH adjusted to 3.5 with dil. Orthophosphoric acid solution.

Optimized Chromatographic conditions

Mobile phase	: 60% 0.01N Na_2HPO_4: 35% Acetonitrile
Flow rate	**:** 1ml/min
Column	**:** C18 (4.6 x 150mm, 5µm)
Detector wave length	**:** 225nm
Column temperature	**:** 30°C
Injection volume	**:** 10µL
Run time	**:** 3.5min
Diluent	**:** Water and Acetonitrile in the ratio 50:50
Results	**:** Both peaks have good resolution, tailing Factor, theoretical plate count and resolution.

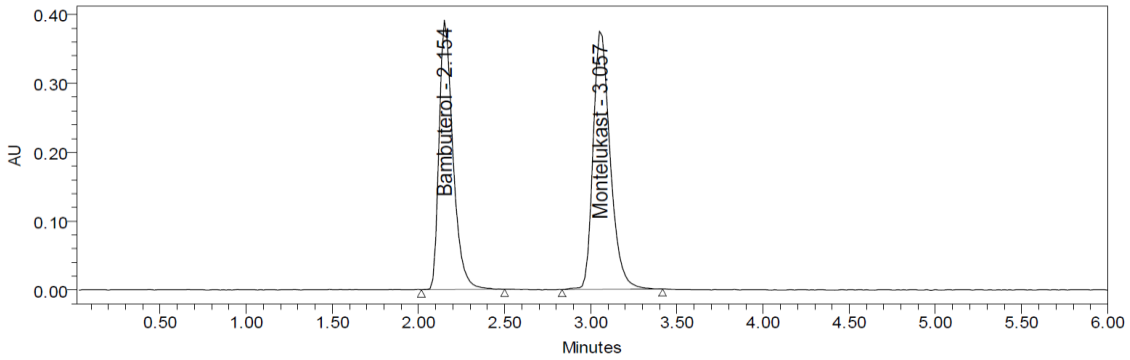

Fig. 3: Chromatogram of Montelukast and Bambuterol.

METHOD VALIDATION

System suitability: The system suitability parameters were determined by preparing standard solutions of Montelukast (10ppm) and Bambuterol (10ppm) and the solutions were injected six times and the parameters like peak tailing, resolution and USP plate count were determined. The % RSD for the area of six standard injections results should not be more than 2%.

Table 1: System suitability of Montelukast and Bambuterol.

S no	Montelukast			Bambuterol			
Inj	RT(min)	USP Plate Count	Tailing	RT(min)	USP Plate Count	Tailing	Resolution
1	2.149	3364	1.39	3.038	4727	1.31	5.5
2	2.154	3508	1.40	3.056	4659	1.31	5.5
3	2.155	3465	1.40	3.057	4555	1.31	5.4
4	2.155	3524	1.40	3.057	4610	1.30	5.4
5	2.155	3541	1.40	3.058	4566	1.31	5.4
6	2.156	3521	1.40	3.059	4665	1.31	5.4

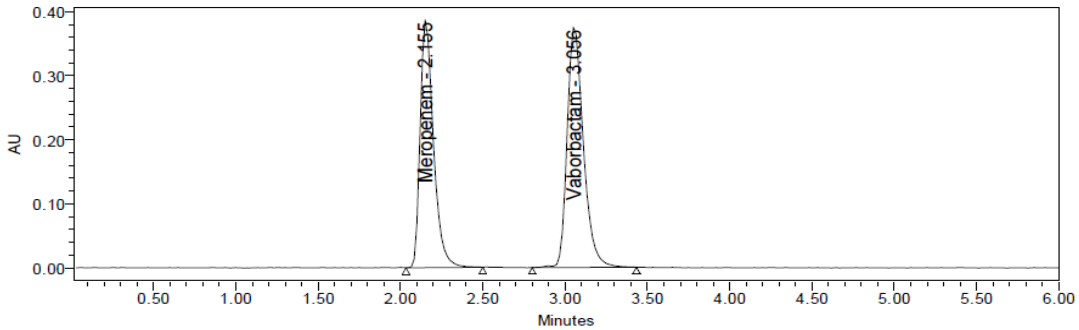

Fig 4: System suitability Chromatogram of Montelukast and Bambuterol.

Result: According to ICH guidelines plate count should be more than 2000, tailing factor should be less than 2 and resolution must be more than 2. All the system suitable parameters were passed and were within the limits.

Specificity: Checking of the interference in the optimized method. We should not find interfering peaks in blank and placebo at retention times of these drugs in this method. So this method was said to be specific.

Result: Retention times of Montelukast and Bambuterol were 2.154min and 3.057 min respectively. We did not found and interfering peaks in blank and placebo at retention times of these drugs in this method. So this method was said to be specific.

Linearity

Preparation of Standard stock solutions: Accurately weighed 5mg of Montelukast and 5mg of Bambuterol and transferred to 50ml volumetric flask. and 3/4 th of diluents was added to these flasks and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (100µg/ml of Montelukast and 100µg/ml of Bambuterol).

25% Standard solution: 0.25ml each from two standard stock solutions was pipetted out and made up to 10ml. (2.5µg/ml of Montelukast and 2.5µg/ml of Bambuterol).

50% Standard solution: 0.5ml each from two standard stock solutions was pipetted out and made up to 10ml. (5µg/ml of Montelukast and 5µg/ml of Bambuterol).

Table 2: Linearity of Montelukast and Bambuterol.

Montelukast		Bambuterol	
Conc (µg/mL)	Peak area	Conc (µg/mL)	Peak area
0	0	0	0
2.5	552741	2.5	620304
5	1048792	5	1190020
7.5	1605740	7.5	1813023
10	2191362	10	2441403
12.5	2709731	12.5	3025427
15	3168313	15	3555329

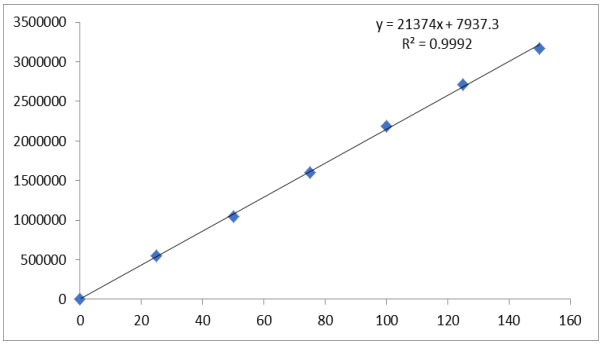

Fig. 5: Calibration curve of Montelukast.

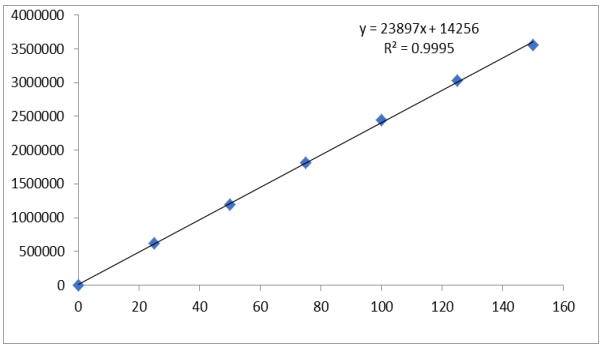

Fig. 6: Calibration curve of Bambuterol.

Result: Six linear concentrations of Montelukast (2.5-15µg/ml) and Bambuterol (2.5-15µg/ml) were injected in a duplicate manner. Average areas were mentioned above and linearity equations obtained for Montelukast was y =21374x + 7937 and of Bambuterol was y = 23897x + 14256 Correlation coefficient obtained was 0.999 for the two drugs.

Accuracy

Preparation of Standard stock solutions: Accurately weighed 5mg of Montelukast and 5mg of Bambuterol and transferred to 50ml volumetric flask. And ¾ th of diluents was added to these flasks and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (100µg/ml of Montelukast and 100µg/ml of Bambuterol).

Preparation of 50% Spiked Solution: 0.5ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out, and made up to the mark with diluent.

Preparation of 100% Spiked Solution: 1.0ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out, and made up to the mark with diluent.

Preparation of 150% Spiked Solution: 1.5ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out, and made up to the mark with diluent.

Table 3: Accuracy of Montelukast.

% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean %Recovery
50%	5	4.931	98.62	99.71%
	5	4.971	99.42	
	5	5.036	100.73	
100%	10	10.089	100.89	
	10	10.017	100.17	
	10	9.942	99.42	
150%	15	14.953	99.69	
	15	14.701	98.01	
	15	15.064	100.43	

Table 4: Accuracy of Bambuterol.

% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean %Recovery
50%	5	5.0477599	100.96	100.10%
	5	4.9544538	99.09	
	5	4.9884921	99.77	
100%	10	10.035792	100.36	
	10	10.092277	100.92	

	10	10.031862	100.32	
150%	15	14.970368	99.80	
	15	14.913519	99.42	
	15	15.036524	100.24	

Discussion: Three levels of Accuracy samples were prepared by standard addition method. Triplicate injections were given for each level of accuracy and mean %Recovery was obtained as 99.71% and 100.10% for Montelukast and Bambuterol respectively.

Acceptance Criteria

The % Recovery for each level should be between 98.0 to 102.

Precision

Table 5: System precision of Montelukast and Bambuterol.

S. No	Area of Montelukast	Area of Bambuterol
1.	2199254	2599926
2.	2185405	2566687
3.	2196543	2593478
4.	2165566	2581870
5.	2165540	2596740
6.	2193056	2577451
Mean	2184227	2586025
S.D	15193.9	12871.1
%RSD	0.7	0.5

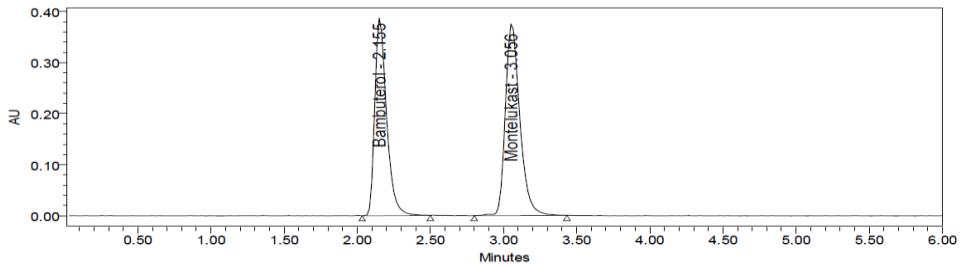

Fig. 7: System precision chromatogram of Montelukast and Bambuterol.

Discussion: From a single volumetric flask of working standard solution six injections were given and the obtained areas were mentioned above. Average area, standard deviation and % RSD were calculated for two drugs. % RSD obtained as 0.7% and 0.5% respectively for Montelukast and Bambuterol. As the limit of Precision was less than "2" the system precision was passed in this method.

Repeatability

Table 6: Repeatability of Montelukast and Bambuterol.

S. No	Area of Montelukast	Area of Bambuterol
1.	2184077	2596345
2.	2196970	2596957
3.	2165409	2592741
4.	2207827	2565390
5.	2181436	2551818
6.	2166003	2593274
Mean	2183620	2582754
S.D	16799.9	19263.4
%RSD	0.8	0.7

Discussion: Multiple sampling from a sample stock solution was done and six working sample solutions of same concentrations were prepared, each injection from each working sample solution was given and obtained areas were mentioned in the above table. Average area, standard deviation and % RSD were calculated for two drugs and obtained as 0.8% and 0.7% respectively for Montelukast and Bambuterol. As the limit of Precision was less than "2" the system precision was passed in this method.

Robustness: Small deliberate changes in method like Flow rate, mobile phase ratio, and temperature are made but there was no recognized change in the result and are within range as per ICH Guide lines.

Robustness conditions like Flow minus (0.9ml/min), Flow plus (1.1ml/min), mobile phase minus, mobile phase plus, temperature minus (25°C) and temperature plus(35°C) was maintained and samples were injected in duplicate manner. System suitability parameters were not much effected and all the parameters were passed. %RSD was within the limit.

Robustness

Table 7: Robustness data for Montelukast and Bambuterol.

S.no	Condition	%RSD of Montelukast	%RSD of Bambuterol
1	Flow rate (Decrease in flow rate) 0.9ml/min	0.1	0.7
2	Flow rate (Increase in flow rate) 1.1ml/min	0.1	0.1
3	Mobile phase (Decrease in flow rate) 59B:36A	0.7	0.8
4	Mobile phase (Increase in flow rate) 61B:34A	0.2	0.1
5	Temperature (Decrease in flow rate) 25°C	0.3	0.4
6	Temperature (Increase in flow rate) 35°C	0.4	0.6

DISCUSSION

Robustness conditions like Decrease in flow rate (0.9ml/min), Increase in flow rate (1.1ml/min), mobile phase Decrease in flow rate (65B:35A), mobile phase Increase in flow rate (55B:45A), temperature Decrease in flow rate (25°C) and temperature Increase in flow rate (35°C) was maintained and samples were injected in duplicate manner. System suitability parameters were not much affected and all the parameters were passed. %RSD was within the limit.

LOD sample Preparation: 0.25ml each from two standard stock solutions was pipetted out and transferred to two separate 10ml volumetric flasks and made up with diluents. From the above solutions 0.1ml each of Montelukast and Bambuterol solutions respectively were transferred to 10ml volumetric flasks and made up with the same diluents

LOQ sample Preparation: 0.25ml each from two standard stock solutions was pipetted out and transferred to two separate 10ml volumetric flask and made up with diluent. From the above solutions 0.3ml each of Montelukast and Bambuterol solutions respectively were transferred to 10ml volumetric flasks and made up with the same diluent.

Table 8: Sensitivity table of Montelukast and Bambuterol.

Molecule	LOD	LOQ
Montelukast	0.09	0.26
Bambuterol	0.05	0.16

Assay

Montek Plus Tab, bearing the label claim containing Montelukast 10mg + Bambuterol 10mg.Assay was performed with the above formulation. Average % Assay for Montelukast and Bambuterol obtained was 99.87 and 99.77% respectively.

Table 9: Assay Data of Montelukast.

S.no	Standard Area	Sample area	% Assay
1	2199254	2184077	99.89
2	2185405	2196970	100.48
3	2196543	2165409	99.04
4	2165566	2207827	100.98
5	2165540	2181436	99.77
6	2193056	2166003	99.07
Avg	2184227	2183620	99.87
Stdev	15193.9	16799.9	0.77
%RSD	0.7	0.8	0.8

Table 10: Assay Data of Bambuterol.

S.no	Standard Area	Sample area	% Assay
1	2599926	2596345	100.30
2	2566687	2596957	100.32
3	2593478	2592741	100.16
4	2581870	2565390	99.10
5	2596740	2551818	98.58
6	2577451	2593274	100.18
Avg	2586025	2582754	99.77
Stdev	12871.1	19263.4	0.7442
%RSD	0.5	0.7	0.7

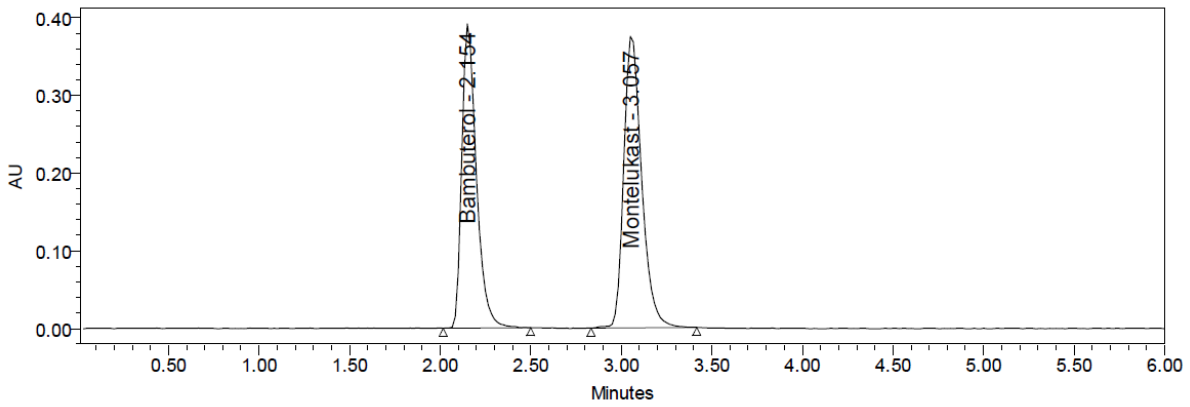

Fig. 8: Chromatogram of working standard solution of Montelukast and Bambuterol.

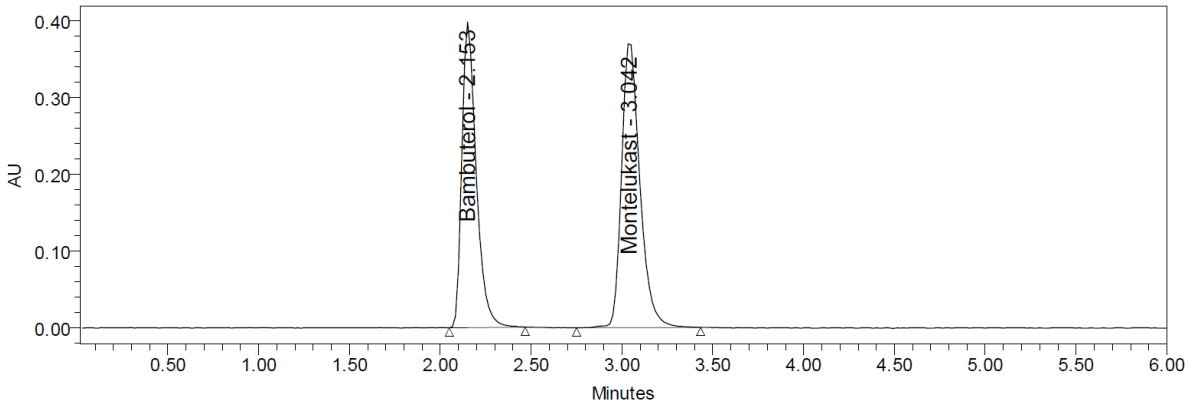

Fig. 9: Chromatogram of working sample solution of Montelukast and Bambuterol.

Degradation studies

Oxidation

To 1 ml of stock solution of Montelukast and Bambuterol, 1 ml of 20% hydrogen peroxide (H2O2) was added separately. The solutions were kept for 30 min at 600c. For HPLC study, the resultant solution was diluted to obtain 10µg/ml&10µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Acid Degradation Studies

To 1 ml of stock s solution Montelukast and Bambuterol, 1ml of 2N Hydrochloric acid was added and refluxed for 30mins at 600c. The resultant solution was diluted to obtain 10µg/ml&10µg/ml solution and 10 µl solutions were injected into the system and the chromatograms were recorded to assess the stability of sample.

Alkali Degradation Studies

To 1 ml of stock solution Montelukast and Bambuterol, 1 ml of 2N sodium hydroxide was added and refluxed for 30mins at 600c. The resultant solution was diluted to obtain 10µg/ml&10µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Dry Heat Degradation Studies

The standard drug solution was placed in oven at 105°C for 1 h to study dry heat degradation. For HPLC study, the resultant solution was diluted to 10µg/ml&10µg/ml solution and 10µl were injected into the system and the chromatograms were recorded to assess the stability of the sample**.**

Photo Stability studies

The photochemical stability of the drug was also studied by exposing the 100µg/ml&100µg/ml solution to UV Light by keeping the beaker in UV Chamber for 1days or 200-Watt hours/m^2 in photo stability chamber. For HPLC study, the resultant solution was diluted to obtain 10µg/ml&10µg/ml solutions and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Neutral Degradation Studies

Stress testing under neutral conditions was studied by refluxing the drug in water for 1hrs at a temperature of 60º. For HPLC study, the resultant solution was diluted to 100µg/ml&100µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of the sample**.**

Table 11: Degradation of Montelukast and Bambuterol.

Type of degradation	Montelukast		Bambuterol	
	% RECOVERED	% DEGRADED	% RECOVERED	% DEGRADED
Acid	93.93	6.07	93.81	6.19
Base	95.91	4.09	95.17	4.83
Peroxide	95.34	4.66	95.11	4.89
Thermal	97.24	2.76	97.27	2.73
Uv	99.09	0.91	99.14	0.86
Water	99.72	0.28	99.86	0.14

Degradation chromatograms

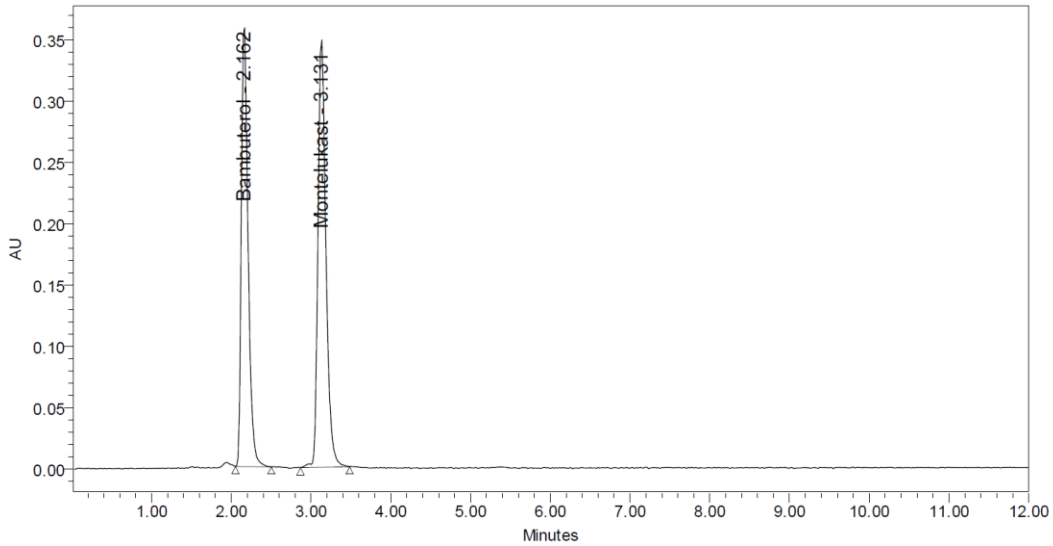

Fig. 10: Acid degradation chromatogram of Montelukast and Bambuterol.

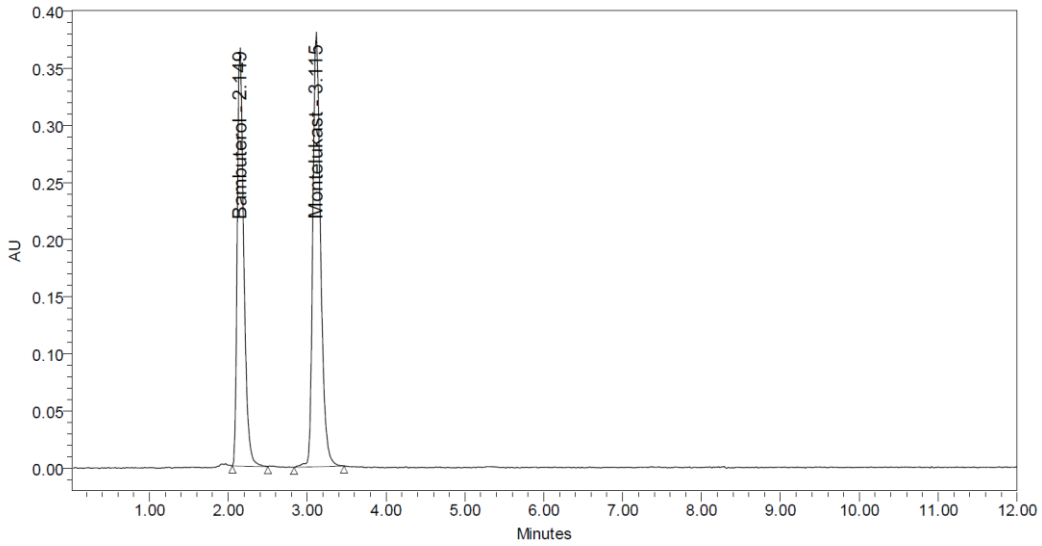

Fig. 11: Base degradation chromatogram of Montelukast and Bambuterol.

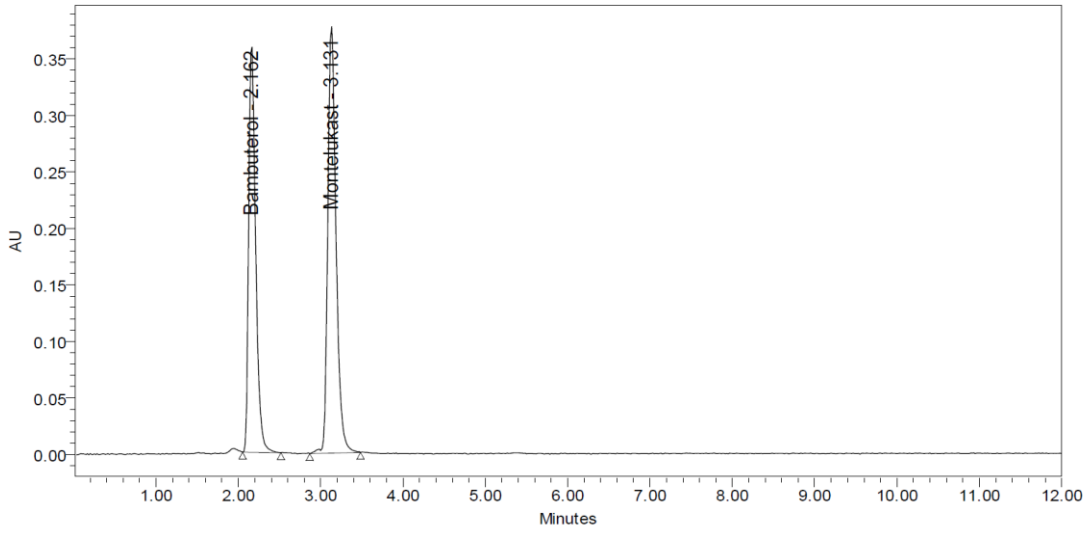

Fig. 12: Peroxide degradation chromatogram of Montelukast and Bambuterol.

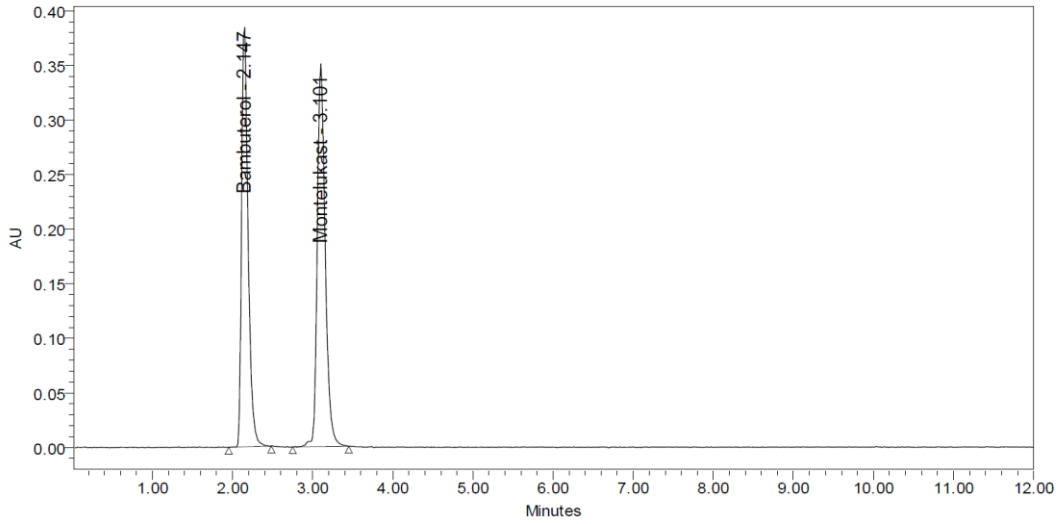

Fig. 13: Thermal degradation chromatogram of Montelukast and Bambuterol.

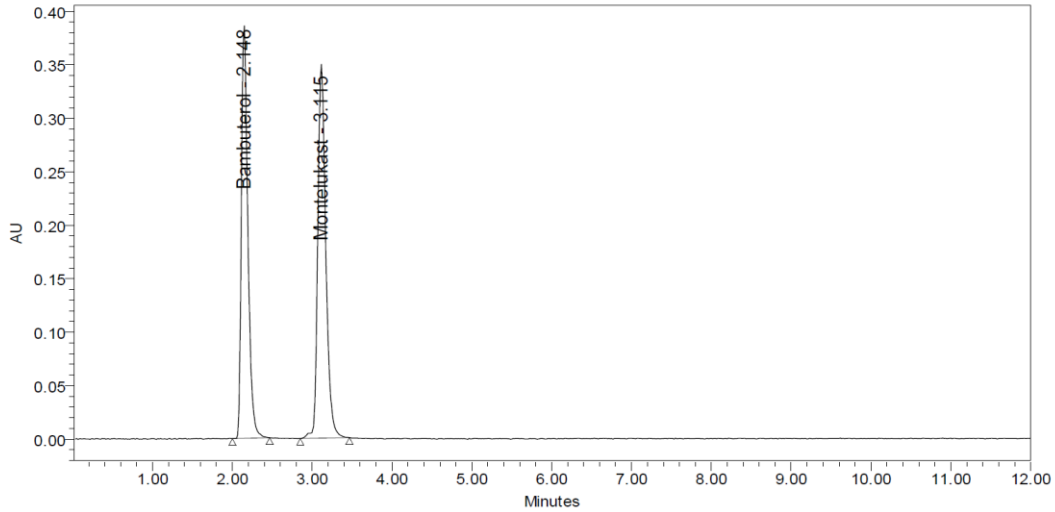

Fig. 14: UV degradation chromatogram of Montelukast and Bambuterol.

SUMMARY AND CONCLUSION

A simple, Accurate, precise method was developed for the simultaneous estimation of the Montelukast and Bambuterol in injection dosage form. Retention time of Montelukast and Bambuterol were found to be 2.154 min and 3.057 min. %RSD of the Montelukast and Bambuterol were and found to be 0.7 and 0.5 respectively. %Recovery was obtained as 99.71% and 100.10% for Montelukast and Bambuterol respectively. LOD, LOQ values obtained from regression equations of Montelukast and Bambuterol were 0.09, 0.26µg/ml and 0.05, 0.16µg/ml respectively. Regression equation of Montelukast isy =21373x + 7937, and y = 23896x + 14256 of Bambuterol. Retention times were decreased and that run time was decreased, so the method developed was simple and economical that can be adopted in regular Quality control test in Industries.

REFERENCES

1. Sreelakshmi. Ma, RP- HPLC Method for Simultaneous Estimation of Montelukast and Bambuterol in Bulk Samples. International Journal of Medical Science and Innovative Research (IJMSIR), 2017; 361–367.
2. Mojgan Sabet, Activity of Simulated Human Dosage Regimens of Montelukast and Bambuterol against Carbapenem-resistant *Enterobacteriaceae* in an In Vitro Hollow Fiber Model., Copyright © American Society for Microbiology. Accepted manuscript posted online13, doi:10.1128/AAC.01969-17, 2017; 781–783.
3. Olga Lomovskaya, Bambuterol: Spectrum of Beta-Lactamase Inhibition and Impact of Resistance Mechanisms on Activity in Enterobacteriaceae American Society for Microbiology. Accepted manuscript posted online28 doi:10.1128/AAC.01443, 2017; 48-52.
4. Przemys Aw Zalewski, Development and validation of stability-indicating Hplc method for simultaneous determination of Montelukast and potassium clavulanate. Acta Poloniae Pharmaceutica ñ Drug Research, 2014; 71(2): 255-260.
5. GregoriCasals[a], Development and validation of a UHPLC diode array detector method for Montelukast

quantification in human plasma The Canadian Society of Clinical Chemists. Published by Elsevier Inc., 2014; 47(16–17): 223-227.
6. Ping CHANG, Determination of Montelukast in Human Plasma by HPLC: Validation and its Application to Pharmacokinetic Study, Latin American Journal of Pharmacy, 2014; 870.
7. Zalewski P, Development and validation of stability-indicating HPLC method for simultaneous determination of Montelukast and potassium clavulanate. Acta Pol Pharm, 2014; 71(2): 255-60.
8. Ramona khanum, development and validation of a rp-hplc method for the detection of Montelukast as a pure compound, in a pharmaceutical dosage form and post thermal induced degradation**.** International Journal of Pharmacy and Pharmaceutical Sciences, 2014; 4(2): 78-84.
9. "International Nonproprietary Names for Pharmaceutical Substances (INN). Recommended International Nonproprietary Names: List 75"(PDF). World Health Organization, 161–2.
10. Rashmin, An introduction to analytical Method Development for Pharmaceutical formulations. Indoglobal Journal of Pharmaceutical Sciences, 2012; 2(2): 191-196.
11. Ashok Kumar, Lalith Kishore, navpreet Kaur, Anroop Nair. Method Development and Validation for Pharmaceutical Analysis. International Pharmaceutica Sciencia, 2012; 2(3): 43-57.
12. Vibha Gupta, Ajay Deep Kumar Jain, N.S. Gill, Kapil, Development and Validation of HPLC method. International Research Journal of Pharmaceutical and Applied Sciences, 2012; 2(4): 97-101.
13. L.Venkateswara Rao, Reverse Phase HPLC and Visible Spectrophotometric Methods for the Determination of Montelukast in Pure and Pharmaceutical Dosage Form. International Journal of PharmTech Research, 2012; 4(3): 957-962.
14. Guan yang LIN, Determination of Montelukast in Rabbit Plasma by LC–MS/MS. Latin American Journal of Pharmacy, 2011; 1895-1900.
15. Malvia R, Bansal V, Pal O.P and Sharma P.K, A Review of High-Performance Liquid Chromatography. Journal of Global Pharma technology, 2010; 3(4): 105-110.
16. Indian Pharmacopoeia, Indian pharmacopeial Commission, Controller of Publication, Government of India, Ministry of health and Family Welfare, Ghaziabad, India, 2010; 2: 1657-1658.
17. Nicolau, David P., et al., Pharmacokinetic and pharmacodynamic properties of Montelukast. Clinical infectious diseases: an official publication of the Infectious Diseases Society of America, 2008; 47(1): S32-40. PMID: 18713048, 187–192.
18. Lindholm.J, Development and Validation of HPLC Method for Analytical and Preparative purpose. Acta Universitatis Upsaliensis, 2004; 13-14.
19. Cottagnoud, P., et al., Cellular and molecular aspects of drugs of the future: Montelukast. Cellular and molecular life sciences: CMLS., 2002; 59(11): 1928-33. PMID: 12530523
20. Green JM, APracticle guide to analytical method validation, Anal Chem, 1996; 305A-309A.
21. ICH, Validation of analytical procedures: Text and Methodology. International Conference on Harmonization, IFPMA, Geneva, 1996.