AMERICAN JOURNAL OF PHARMTECH RESEARCH
Journal home page: http://www.ajptr.com/

Validated Stability-Indicating Isocratic RP-HPLC Method of Determination of Rosuvastatin calcium and Fenofibrate in Bulk and in Solid dosage by Vieordt's method

Rekha Rajeevkumar *[1], Dr. P.Rajeev kuma[2]
1. *School of Pharmacy, PRIST University, Manamai-Nallur, ECR, Near Mahabalipuram, Chennai, Tamilnadu -603102.*

ABSTRACT

A modest, fast, precise, and accurate, stability-indicating reversed phase high performance liquid chromatographic method was developed and validated for vierodt's method of analysis of rosuvastatin calcium(RSTC) and fenofibrate (FBT). The volume of injection sample was 20 µl and the quantification was obtained by UV-VISIBLE detector at 240nm.The chromatographic separation was achieved on X bridge C_{18}, 250 x 4.6 mm, 5 µm particle column, by an gradient mobile phase comprising of acetonitrile: 10 mM potassium dihydrogen phosphate buffer solutions of pH 5.5 in the ratio of 90:10 *v/v* at a flow rate of 1.5 ml/ min. The retention times for RSTC and FBT were found to be 4.35min and 7.75 min, respectively. The drugs were shown to thermal, photolytic, hydrolytic, and oxidative stress conditions and the stressed samples were evaluated by the suggested method. Validation of the method was approved as per International Conference on Harmonization (ICH) guidelines. Linearity was accepted for RSTC and FBT in the range of 0.040-0.120 mg/ ml and 0.016-0.048 mg/ ml, respectively. The limits of detection were 0.02 µg/ ml and 0.05 µg/ ml, respectively and the LOQ value 0.1 µg/ ml and 0.09 µg/ ml, for RSTC and FBT, respectively. The developed method was profound to be accurate, precise and stability-indicating as no interfering peaks of degradants and excipients were identified. The suggested method is hence appropriate for application in quality-control laboratories for quantitative determination of both the drugs individually and in combination, since it is simple and rapid with good accuracy and precision.

Keywords: Rosuvastatin calcium (RSTC), Fenofibrate (FBT), reversed-phase HPLC, stability-indicating assay, forced degradation studies, method validation

*Corresponding Author Email: rekhavas@gmail.com
Received 24 February 2017, Accepted 08 March 2017

Please cite this article as: Rekha R *et al*., Validated Stability-Indicating Isocratic RP-HPLC Method of Determination of Rosuvastatin calcium and Fenofibrate in Bulk and in Solid dosage by Vieordt's method. American Journal of PharmTech Research 2017.

INTRODUCTION

Rosuvastatin calcium (RSTC) is chemically bis [(E)-7 [4-(4- fluorophenyl)-6 isopropyl-2-[methyl (methylsulphonyl) amino] pyrimidin-5-yl] (3R, 5S) -3, 5-dihydroxyhept-6-enoic acid] Calcium salt **(**Figure 1). It is a lipid lowering drug. It inhibits the enzyme 3-hydroxry- 3-methyl glutaryl coenzyme A (HMG-CoA) reductase, the rate limiting enzyme that converts HMG-CoA to mevalonate; a precursor of cholesterol and thereby monitors the synthesis of cholesterol. It is significant in the treatment of hypercholesterolemia and dyslipidemia[1-5]. It is employed to lower the amounts of LDL cholesterol, VLDL cholesterol, total cholesterol, triglycerides and apolipoprotein B in the blood. RSTC also modestly raises the level of HDL cholesterol in the blood. These actions are paramount factors in reducing the hazard of astherosclerosis, which in turn can result in numerous cardiovascular problems such as heart attack, stroke and peripheral vascular disease [6].

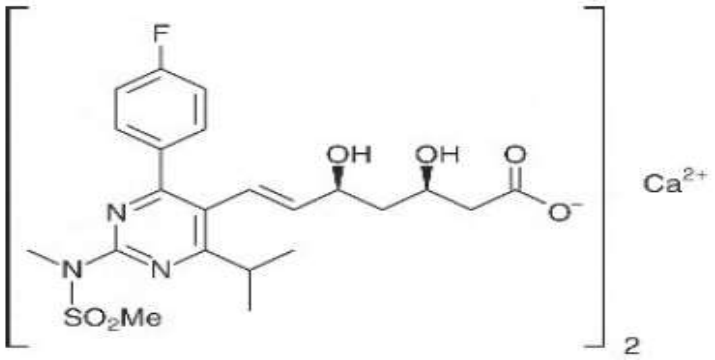

Figure 1: Chemical structure of Rosuvastatin calcium

Figure 2: Chemical structure of Fenofibrate

Fenofibrate (FBT) is a drug of the fibrate class[4]. FBT is chemically propan-2-yl 2{4-[(4-chlorophenyl) - carbonyl] phenoxy}-2-methylpropanoate **(**Figure 2).It is mainly used to lower cholesterol levels in patients at risk of cardiovascular disease. Like other fibrates, it decreases low density lipoprotein (LDL) and very low density lipoprotein (VLDL) levels, as well as reducing triglycerides (TG) level. It also elevates high density lipoprotein (HDL) levels [4-5]. It is used alone or in combination with statins in the treatment of hypercholesterolemia and hypertriglyceridemia[6] . To the best of our knowledge no method is reported in literature for simultaneous determination of RSTC and FBT by high performance liquid chromatography (HPLC) using the specified

chromatographic condition. The survey of literature showed few UV Spectrophotometric [7-9] HPLC[10-12] HPTLC[13] chromatography stability indicating method [14] micellar stability indicating method[15], stability indicating LC method[16]LC-MS method[17-20] are available for the estimation of RSTC in pharmaceutical preparation and in biological fluids. Literature survey also showed that available for the estimation of UV spectrophotometric[21] HPLC[22] and HPTLC[23-24], TLC [25] for the estimation of FBT in pharmaceutical preparation.

The aim of the present work was to develop and validate a modest, economic, rapid, precise, gradient and accurate stability-indicating method with good sensitivity for simultaneous analysis of of RSTC and FBT in as per ICH [26-28]guidelines. The suggested method was successfully employed to a tablet dosage form of RSTC and FBT was determined in presence of commonly used tablet excipients. This method can also be employed for quality control during manufacture of drug product.

MATERIALS AND METHOD

Chemicals, Reagents, and Solutions

HPLC grade acetonitrile, methanol and analytical grade potassium dihydrogen ortho phosphate,ortho phosphoric acid, triethylamine was obtained from S.D. Fine chemicals Ltd. (Mumbai, India). Hydrochloric acid, sodium hydroxide pellets and 3% v/v hydrogen peroxide solution were obtained from Ranbaxy Fine Chemicals, New Delhi (India). High purity water was prepared by Millipore Milli-Q plus purification system (Millipore, Bedford, USA).

HPLC Instrumentation and Chromatographic Conditions

The chromatographic system used to perform development and validation of this assay method was comprised of a gradient (ACME 930) pump, UV-VISIBLE [UV 730 DA]DETECTOR and a rheodyne manual injector with 20µl loop, Column C_{18}, 250 x 4.6 mm, 5 µm particle connected to a multi-instrument data acquisition and data processing system (YOUNGLIN,ACME 3000 SERIUS).The chromatographic separation was achieved on the chromatographic column used was X bridge C_{18}, 250 x 4.6 mm, 5 µm particle using an gradient mobile phase consisting of acetonitrile: 10 mM potassium dihydrogen phosphate buffer solutions of pH 5.5 in the ratio of 90:10 *v/v* at a flow rate of 1.5 ml/min. The sample injection volume was 20 µL and the quantification was achieved by UV-VISIBLE detector at 240nm.

Standard Preparation

RSTC standard stock solution was prepared by accurately weighing quantity of RSTC equivalent to 10 mg of rosuvastatin in a 100 ml volumetric flask in mobile phase. This solution is then

sonicated for 10 minutes and diluted to volume with mobile phase. Further 1ml of this stock solution is taken in a 100 ml volumetric flask and made up to the mark with mobile phase (this standard solution of 10µg/ml).FBT standard stock solution containing 16 µg/ml was prepared in a 100 ml volumetric flask by dissolving 160 mg of FBT in diluent and diluted to volume with diluent. Further 1 ml of this stock solution is taken in a 100 ml volumetric flask and make the volume up to mark with diluent (this standard solution of 16µg/ml). The concentration obtained was 10 µg/ml of RSTC and 16µg/ml of FBT. The above solution was filtered through a 0.45µ nylon membrane filtered.

Test Preparation

Twenty tablets were weighed and the average weight of tablet was determined. The tablets were powdered and accurately weighed quantity of RSTC equivalent to 10 mg of rosuvastatin and fenofibrate in a 100 ml volumetric flask in mobile phase. This solution is then sonicated for 10 minutes and diluted to volume with mobile phase. Further 1ml of this stock solution is taken in a 100 ml volumetric flask and made up to the mark with mobile phase (this 10µg/ml of rosuvastatin and 16 µg/ml of fenofibrate sample solution. The sample was filtered through 0.45µm nylon syringe filter.

Forced degradation of sample for specificity study

Degradation Study

In order to determine the analytical method and assay for the study of stability indicating method in tablets of RSTC and FBT studies under various stressed conditions to conduct forced degradation studies. The degradation samples were prepared by transferring powdered tablets, equivalent to 10 mg RSTC and 160 mg FBT into a 250 ml round bottom flask. Then prepared samples were employed for acidic, alkaline and oxidant media and also for thermal and photolytic stress conditions. After the degradation treatments were completed, the stress content solutions were allowed to equilibrate to room temperature and diluted with mobile phase to attain 0.010 mg/ml of RSTC and 0.160 mg/ml of FBT concentration. Specific degradation conditions were described. The specificity of the developed HPLC method was evaluated by forced degradation studies of RSTC and FBT sample was refluxed with aqueous 0.1N hydrochloric acid and 0.1N sodium hydroxide at 60^0C for 12 hrs separately to study the formation of degradation products under acidic and basic conditions. RSTC and FBT sample was also refluxed with 3% hydrogen peroxide solution at 60^0C for half-an hour to study the formation of degradation products under oxidative condition. Degradation products under photolytic and thermal conditions was studied by exposing RSTC and FBT sample to ultraviolet light (254 nm) for 12 hrs while other sample was

kept at 70^0C temperature for 12 hrs, respectively. Purity of RSTC and FBT peak was evaluated by using UV-visible detector in case of samples subjected for stress conditions and control sample.

Method Validation

Specificity

The specificity of the method was determined by checking the interference of placebo with analyte and the proposed method were eluted by checking the peak purity of RSTC and FBT during the force degradation study. The peak purity of the RSTC and FBT were found satisfactory under different stress condition. There was no interference of any peak of degradation product with drug peak.

Linearity

Standard solutions at nine different concentration levels ranging from LOQ to 0.15µg/mL (120% of specification limit) were prepared and analyzed in duplicate to demonstrate the linearity for RSTC and FBT respectively. The calibration curves were plotted for the all the impurities using area counts versus corresponding concentrations. The slope, Y-intercept and correlation coefficient were calculated. The substances were quantified against area count of in standard solution and multiplied with their response factors to obtain the results (%w/w).

Accuracy

Accuracy of the method was demonstrated by using drug substance spiked at three different concentration levels in triplicate. The analyses were carried out at 80%, 100% and 120% of specification limit as per ICH guide lines.

Precision

System precision of the method was evaluated by injecting RSTC and FBT standard solution six times and calculated the percent relative standard deviation (%RSD) for area of RSTC and FBT peak. The precision of the method for the determination of RSTC and FBT was studied for repeatability and intermediate precision.

Limit of detection (LOD) and Limit of quantitation (LOQ)

The limit of detection and limit of quantification were evaluated by serial dilutions of RSTC and FBT stock solution in order to obtain signal to noise ratio of 3:1 for LOD and 10:1 for LOQ. The limit of detection and limit of quantitation were calculated from the linearity data.

Precision at LOQ level

Precision of the method was also evaluated by injecting standard solutions of known concentration of RSTC and FBT at about the predicted LOQ levels for six times separately.

Robustness

In order to demonstrate the robustness of the method, system suitability parameters were verified by making deliberate changes in the chromatographic conditions, viz. change in flow rate by ±0.1 mL/ min, change in pH of the buffer by ±0.1 unit and change in the column temperature by ±5ºC, keeping the rest of the chromatographic conditions for each alteration study constant.

Stability of the solution

The solution stability of RSTC and FBT in test solution was carried out by analyzing sample solution (prepared by using RSTC and FBT (1mg/mL) spiked with the known amount of the substances at 1.0 %w/w level) at a time interval of 12hr for 48 hr by keeping sample solution at room temperature (at 28ºC) and at refrigerated (2-5ºC) conditions separately.

RESULTS AND DISCUSSION

The conditions in the present work, an analytical method established on HPLC using UV detection was developed and validated for assay determination of RSTC and FBT in tablet formulation. The analytical conditions were selected, keeping in mind the different chemical nature of RSTC and FBT. Suitable selection of the methods depends upon the nature of the sample (ionic or ionisable or neutral molecule) its molecular weight and solubility. RSTC and FBT is dissolved in polar solvent therefore reversed phase-HPLC was selected to estimate them. To develop a rugged and suitable HPLC method for the quantitative determination of RSTC and FBT, the analytical condition were selected after testing the different parameters such as diluents, buffer, buffer concentration, organic solvent for mobile phase and mobile phase composition and other chromatographic conditions. Our initials trials using different composition of mobile phases consisting of water with methanol did not give good peak shape. Software suggested various parameters, like optimum flow rate, injection volume, mobile phase composition, gradient program etc. The column selection has been done on the basis of backpressure, resolution, peak shape, theoretical plates and day-to-day reproducibility of the retention time and resolution between RSTC and FBT peak. After evaluating all these factors, used X bridge C_{18}, (250 x 4.6 mm, 5 µm particle) column was found to be giving satisfactory results. The selection of buffer based on chemical structure of both the drugs. The acidic pH range was found suitable for solubility, resolution, stability, theoretical plates and peak shape of both components. Best results were obtained with buffer as 0.2 M potassium dihydrogen orthophosphate in 0.1M phosphoric acid adjusted with triethylamine (0.5%) to pH 5.5 (Gradient Elution) solutions improved the peak shape of RSTC and FBT. Finally, by fixing 0.2 M potassium dihydrogen orthophosphate in 0.1M phosphoric acid adjusted with triethylamine (0.5%) to pH 5.5 and mobile phase composition

consisting of a mixture of acetonitrile: buffer pH 5.5 (90:10, v/v). Optimized mobile phase proportion provided good resolution between and also for degradation product which is generated during force degradation study. For the selection of organic constituent of mobile phase, acetonitrile was chosen to reduce the longer retention time and to attain good peak shape. The peak at R_t 4.35min and 7.75 min for RSTC and FBT was observed in the chromatogram of the drug samples extracted from tablet. The results were tabulated in Table 1 respectively. The chromatogram of mixture of RCTC and FBT (bulk preparation) is shown in Figure 1.

To validate RP-HPLC method, a series of tests were made using the most promising conditions. A calibration curve was made and concentration examined within the detection range from 0.040-0.120 mg/ml for RSTC and 0.016-0.048 mg/ml for FBT and the correlation coefficient was found to be 0.9983 and 0.9998 for RSTC and FBT respectively. The assay values shown in Table 2.The percent recovery of RSTC and FBT were determined at determined at three different concentration levels like 80, 100 and 120%. The mean recovery for RST was 99.07-100.58 % and 98.05-100.08 % for FBT. Data obtained from precision experiments are carried out for intraday and interday precision study for both RSTC and FBT. The RSD values for intra day precision study and interday precision study was < 2.0 % for RST and FBT. The result indicating that the method was accurate and confirms good precision. The results are presented under Table 3 and 4.The method was demonstrated to be robust over an acceptable working range of its HPLC operational conditions. The system suitability results within the acceptable limits and selectivity of individual substances were also not affected when subjected deliberately for varied chromatographic conditions. The result of the study confirms the robustness of the method. The result of robustness study of the developed assay method was established in Table 6 and 7 .

The stability of sample was checked by forced degradation in different stress conditions condition like acidic, alkali, thermal, oxidative degradation, photolytic and humidity conditions and % of degradation was calculated. The peak purity of the analyte was passed in all conditions (purity angle should be less than the threshold value).The following values in Table 6 indicate that any other impurity is not merging with the main peak (Figure 5).It was concluded that the test preparation solution was found stable up to 48 hr at 2 - 5 ˚C and room temperature, as during this time the result was not decrease below the minimum percentage.

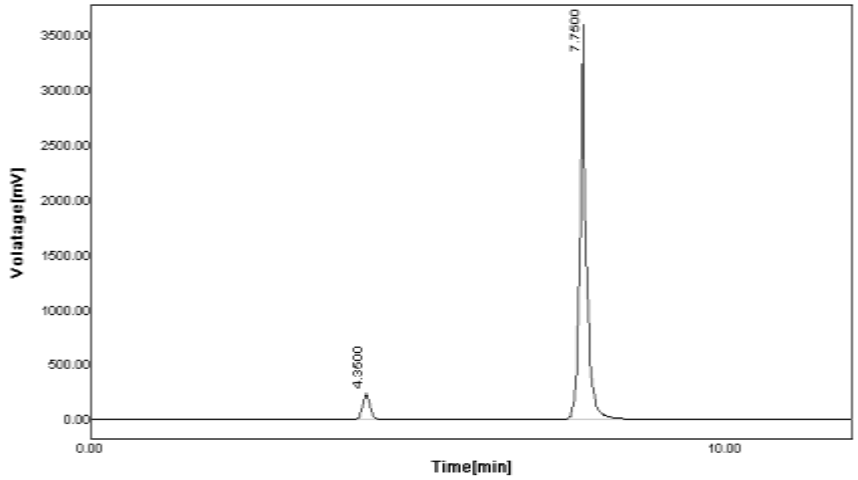

Figure 3: Chromatogram of mixture of untreated RSTC and FBT(bulk)

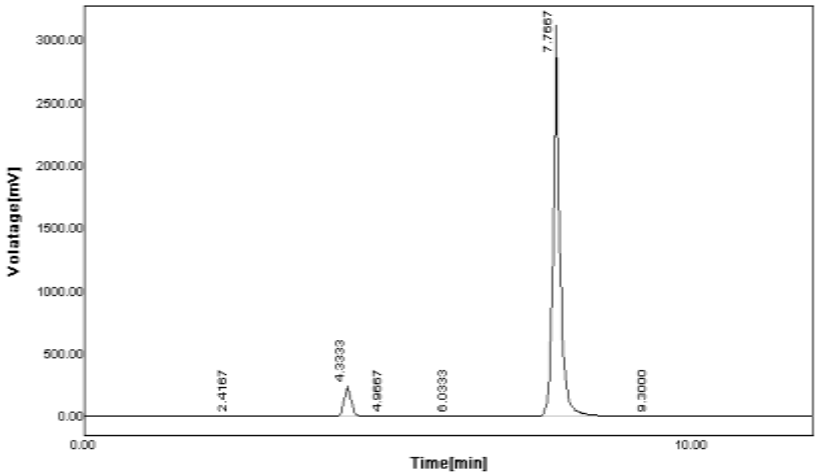

Figure 4: Chromatogram of mixture of untreated RSTC and FBT acidic forced degradation study

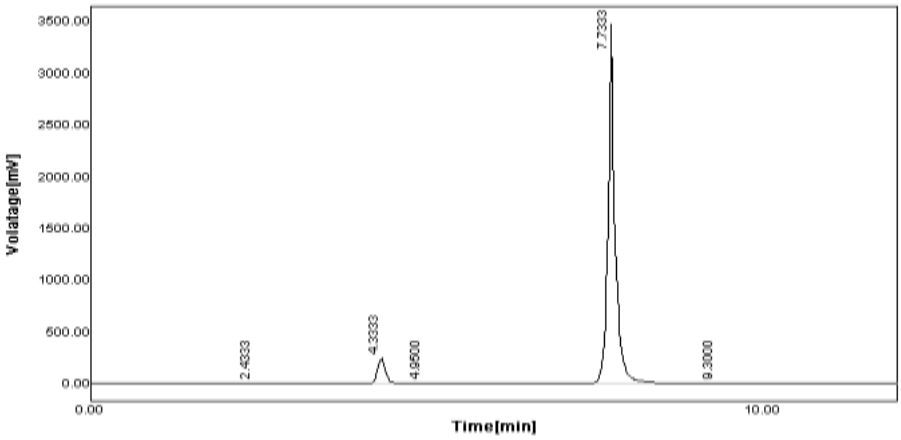

Figure 5: Chromatogram of mixture of untreated RSTC and FBT alkali forced degradation study

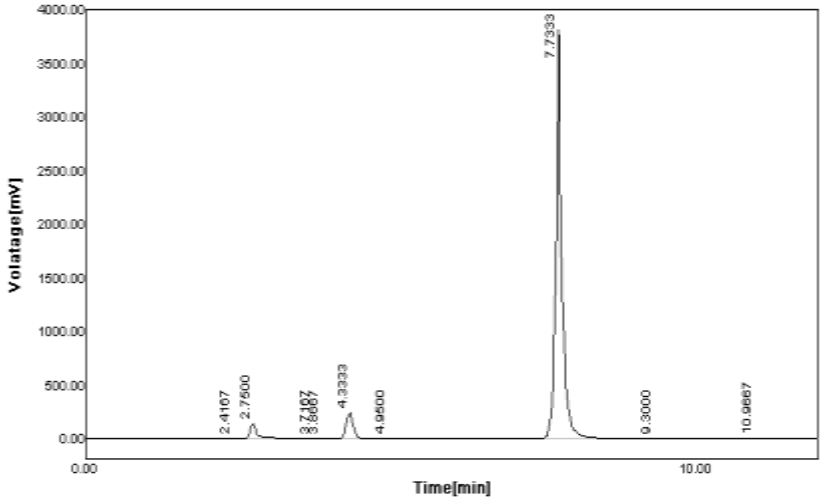

Figure 6: Chromatogram of mixture of untreated RSTC and FBT oxidative forced degradation study

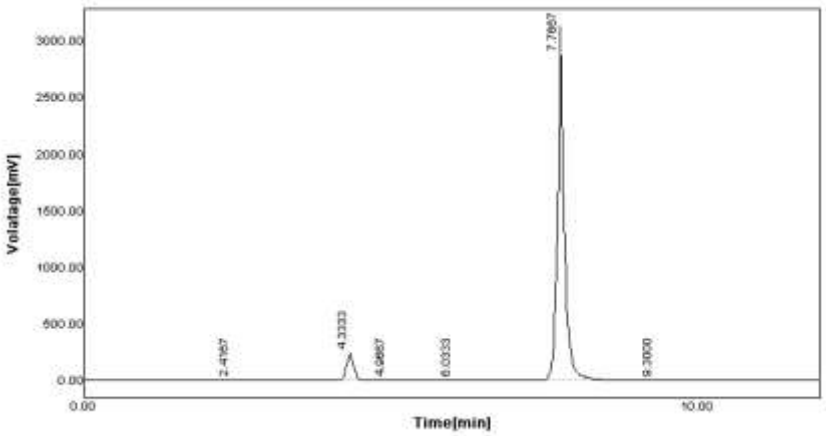

Figure 7: Chromatogram of mixture of untreated RSTC and FBT thermal degradation study

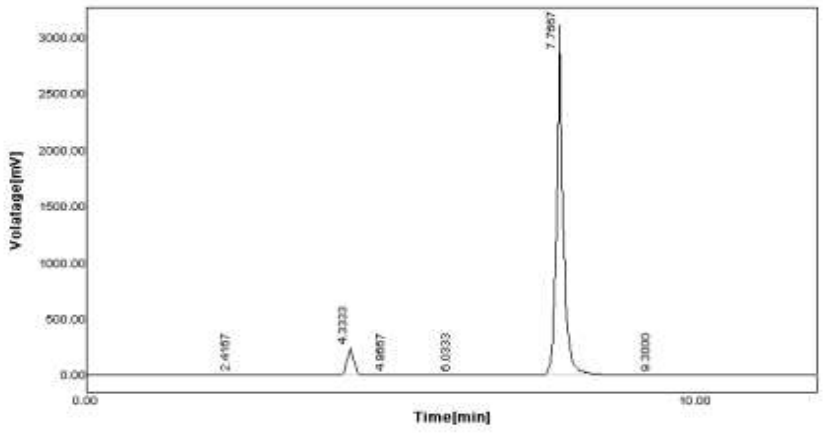

Figure 8: Chromatogram of mixture of untreated RSTC and FBT UV-light degradation study

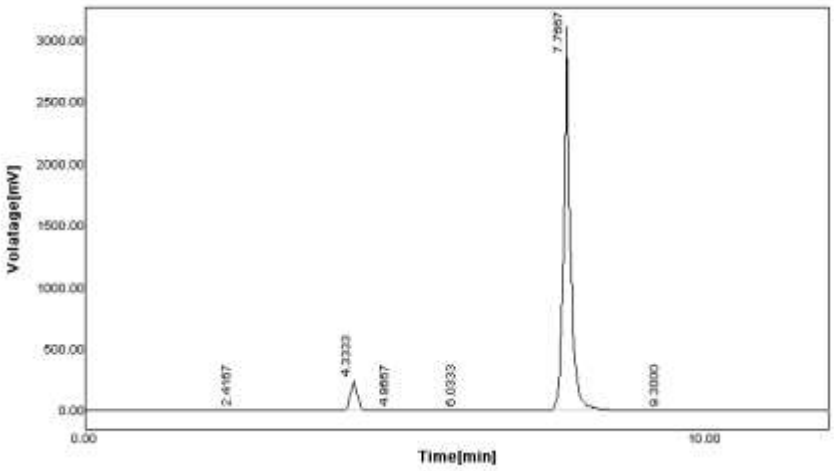

Figure 9: Chromatogram of mixture of RSTC and FBT humidity degradation study

Table 1: Method development conditions

Trial	Type of column	Mobile phase composition (Buffer: acetonitrile)	Injection volume	Flow (ml/min)	Defects
1	C_{18}, (250 x 4.6 mm), 5 µm	80: 20	20µl	1.5	Resolution was not good
2	C_{18}, (250 x 4.6 mm), 5 µm	50: 50	20µl	1.5	Resolution was not good
5	C_{18}, (250 x 4.6 mm), 5 µm	40: 60	20µl	1.5	Resolution was not good
6	C_{18}, (250 x 4.6 mm), 5 µm	10:90	20µl	1.5	Good system suitability parameters

Table 2: Assay of RSTC and FBT

Brand name	Compound	Amount found(mg)	%Assay
Fenovas	RSTC and FBT	10.10	101.4
		159.89	99.94
Novastat-TG 10	RSTC and FBT	10.16	101.6
		159.97	99.98

Table 3: Results of precision study

Set	RSTC (%Assay)		FBT (%Assay)	
	Intraday (n = 6)	Interday (n = 6)	Intraday (n = 6)	Interday (n = 6)
1	101.4	100.4	100.5	99.9
2	100.9	100.8	99.9	99.5
3	98.5	101.3	100.9	99.5
4	99.7	99.9	99.7	98.7
5	100.8	101.7	99.3	99.6
6	100.1	100.3	99.6	99.9
Mean	100.23	100.7	99.91	99.51
Standard deviation	0.93	0.69	1.09	0.23
% RSD	0.97	0.99	1.12	0.86

Table 4: Results of accuracy study

	Level (%)	Theoretical Concentration[a] (µg/ml)	Observed Concentration[a] (µg/ml)	% Recovery	% RSD
RSTC	80	41.7	41.6	98.17	0.49
	100	52.1	51.9	100.58	1.38
	120	62.9	61.9	99.07	0.32
FBT	80	638.8	639.1	100.08	1.23
	100	799.7	799.3	97.58	1.37
	120	958.5	959.2	98.05	1.68

[a] Each value corresponds to the mean of three determinations

Table 5: Forced degradation studies of control sample (1mg/mL) solution.

Type of Degradation	No of unknown impurities	Rosuvastatin calcium and fenofibrate peak area %	% Degradation
Control sample	Nil	99.95	_
Acid	Nil	99.95	_
Alkali	Nil	99.96	--
Peroxide	Nil	99.54	_
Thermal	Nil	99.96	_
Photolytic	Nil	99.89	_
Humidity	Nil	99.97	_

Table 6: Evaluation data of robustness study of RSTC

Robust conditions	% Assay	System suitability parameters	
		Theoretical plates	Asymmetry
Flow 0.9 ml/min	101.2	2122	1.65
Flow 1.5 ml/min	100.8	3500	1.34
Buffer pH 5	99.0	2800	1.54
Buffer pH 5.5	99.4	3200	1.38
Buffer-ACN (90:10,v/v)	98.7	3218	1.73
Buffer-ACN (10:90,v/v)	99.5	3308	1.45
Column change	100	3400	1.39

Table 7: Evaluation data of robustness study of FBT

Robust conditions	% Assay	System suitability parameters	
		Theoretical plates	Asymmetry
Flow 0.9 ml/min	100.4	21156	1.65
Flow 1.5 ml/min	101.5	24256	1.44
Buffer pH 5	100.9	23100	1.50
Buffer pH 5.5	99.9	24313	1.33
Buffer-ACN (90:10,v/v)	99.7	21805	1.56
Buffer-ACN (10:90,v/v)	98.9	23081	1.45
Column change	101.1	24621	1.67

CONCLUSION

A simple, rapid, accurate, and precise stability-indicating HPLC analytical method has been developed and validated for the routine analysis of RSTC and FBT in API and can be applied to formulation. The results of stress testing undertaken according to the ICH guidelines reveal that the method is specific and stability-indicating. The proposed method has the ability to separate these drugs from their degradation products, common excipients used in tablet dosage forms, and hence can be applied to the analysis of routine quality control samples and samples obtained from stability studies.

ACKNOWLEDGMENTS

The authors wish to thank School of Pharmacy, PRIST University, Chennai, Tamilnadu for providing necessary facilities and to carry over the work. The authors wish to express their gratitude to Strides research and specialty of chemicals Ltd, New Mangalore for providing the authentic sample (drug).

REFERENCES

2. Remington. The science & practice of pharmacy, 21st ed. Wolters Kluwer Health Pvt.Ltd, Vol. 2, 2007:1369.
3. Indian pharmacopoeia, The Indian Pharmacopoeia Commission, Vol.3, 2007: 1676-1678.
4. British pharmacopoeia. General medicine council.Vol.1, 2008, 891-892.
5. USP/NF, The official compendia of standards.Vol. 2, 2004, 2351-2354.
6. European Pharmacopeia, Council of Europe, 3rd Edition, Strasbourg. 1997.
7. Morrow, J.D and L.J Roberts**,** In: Goodman and Gilman's the pharmacological basis of therapeutics, 10th ed., New York.: McGraw-Hill, Medical Publishing Division, 2001:933-966.
8. Krishna MV, Sankar DG. Extractive spectrophotometric method for the determination of rosuvastatin calcium in pure form and in pharmaceutical formulation by using safranin o and methylene blue. E-J Chem 2007; 4(1): 46-49.
9. Gupta A, Mishra P, Shah K. Simple UV spectrophotometric determination of rosuvastatin calcium in pure form and in pharmaceutical formulations. J Chem 2009; 6(1): 89-92.
10. Gajjar AK, Shah VD. Simultaneous UV spectrophotometric estimation of rosuvastatin and ezetimibe in their combined dosage forms. Int J Pharm Pharma Sci 2010; 2(1): 107-110.
11. Kumar TR, Shitut NR, Kumar PK, Vinu MC, Kumar VV, Mullangi R, Srinivas NR. Determination of rosuvastatin in rat plasma by HPLC: Validation and its application to pharmacokinetic studies. Biomed Chromatogra 2006; 20(9):881.

12. Doshi N, Sheth A, Sharma A, Dave JB , Patel CN. Validated RP-HPLC method for simultaneous estimation of Rosuvastatin Calcium and Telmisartan in pharmaceutical dosage form**.** J Chem Pharm Res 2010; 2(2): 252-263.
13. Sane RT, Kamat SS, Menon SN, Inamdar SR and Mander RM. Determination of rosuvastatin calcium in its bulk drug and pharmaceutical preparations by High-Performance Thin-Layer Chromatography**.** J Planar Chromatogra 2005; 18:194.
14. Sharma MC, Sharma S, Kohli DV, Sharma AD.A validated HPTLC method for determination of simultaneous estimation of rosuvastatin calcium and ezetimibe in pharmaceutical solid dosages form. Archives of Applied Science Research 2010; 2 (1): 1-7.
15. Raj HA, Rajput SJ, Dave1 JB, Patel CN. Development and validation of two Chromatographic stability-indicating methods for Determination of rosuvastatin In pure form and pharmaceutical preparation. Int J ChemTech Res 2009;1(3):677-689.
16. Sharma S, Sharma MC, Kohli DV, Chaturvedi SC. Micellar liquid Chromatographic method development for determination of rosuvastatin calcium and ezetimibe in pharmaceutical combination dosage form. Der Pharma Chemica 2010; 2(1): 371-377.
17. Mehta TN, Patel AK, Kulkarni GM, Subbaiah G. Determination of rosuvastatin in the presence of its degradation products by a Stability-Indicating LC Method. AOAC Int 2005; 88(4):1142-1147.
18. Trivedi RK, Kallem RR, Mullangi R, Srinivas NR. Simultaneous determination of rosuvastatin and fenofibric acid in human plasma by LC–MS/MS with electro spray ionization: Assay development, validation and application to a clinical study. J Pharm and Biomed Anal 2005; 39: 661–669.
19. Budavari S editor. The Merck Index, 13th Edition, Merck and Co., Inc., Whitehouse Station,NJ:1998:4002.
20. Walker R and Edwards C. Clinical pharmacy and Therapeutics, 3rd ed, 2003: 353-373.
21. Dhabale PN, Gharge DS. Simultaneous spectrophotometric estimation of atorvastatin and fenofibrate in bulk drug and dosage form. Int J ChemTech Res 2010; 2(1): 325-328.
22. Gupta KR, Askarkar SS, Rathod PR, Wadodkar SG, Validated spectro - photometric determination of fenofibrate in formulation. Pelagia Research Library Der Pharmacia Sinica 2010;1(1):173-178.
23. Kumar SG, Prasad R. Development and validation of Reversed Phase HPLC method for simultaneous estimation of rosuvastatin and fenofibrate in tablet dosage form, Int J PharmTech Res 2010; 2(3): 2016-2021.

24. Pawar HI, Kothapalli L, Thomas A, Nanda RK and Mare S. Simultaneous RP-HPLC method for estimation of ezetimibe and fenofibrate in synthetic mixture. Res J Pharmacy Tech 2008; 1(1):45.
25. Deshpande PB, Shridharan G, Anandi L, Jadhav D, Damle MC, Gandhi SV. Validated method development for estimation of atorvastatin and fenofibrate in fixed dose combination by HPTLC.The Pharma Review 2009:151-153.
26. Shrikhedkar AA and Surana SJ. Simultaneous densiometric TLC analysis of atorvastatin and fenofibrate in bulk drug and in pharmaceutical formulation. J Planar Chromatograph 2009; 5:355-358.
27. ICH- Q2A and Q2B, Harmonised Tripartite Guideline, Text on Validation of Analytical Procedure,Geneva,Nov.1995:1-5 and 1-7.
28. ICH-Q1AR2, Harmonized Tripartite Guideline, Stability testing of New drug substances and products, In: Proceedings of the International Conferences on Harmonization,Geneva;2003.
29. International Conference on Harmonization of technical Requirements for registration of Pharmaceuticals for Human Use, ICH Harmonized Tripartite Guidelines, Validation of Analytical Procedures: Text and Methodology Q2(R1),Current Step 4 version, ICH Geneva,Nov.2005.