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A new stability-indicating specific and accurate HPLC method for determination of process-related impurities in bulk and formulation samples of quetiapine hemifumarate: An antipsychotic drug

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ABSTRACT

A simple reversed-phase liquid chromatographic method was developed for the related substances determination in quetiapine hemifumarate, an antipsychotic drug. Forced degradation studies were performed on bulk sample of quetiapine hemifumarate using acid, base, oxidative hydrolysis, thermal stress and photolytic degradation. Considerable degradation of the drug substance was observed during oxidative and acid hydrolysis. The chromatographic method was fine tuned using the samples generated from forced degradation studies and eight process related impurities (Imp-1 to Imp-8). Good resolution between the peaks corresponds to synthetic impurities and degradation products from the analyte were achieved on Zorbax eclipsed XDB C18 column. The stressed test solutions were assayed against the qualified working standard of quetiapine hemifumarate and the mass balance in each case was close to 99.9%, indicating that the developed method was stability-indicating. Validation of the developed method was carried out as per ICH requirements. The proposed RP-HPLC method was successfully applied for the routine evaluation of the quality of bulk drug samples and detection of impurities in pharmaceutical formulations.

KEYWORDS

Quetiapine hemifumarate;
Related substances;
Degradation;
RSD and validation;
Stability indicating.

INTRODUCTION

Quetiapine hemifumarate **(1)**, a dibenzothiazepine derivative is described chemically as 2-[2-(4-dibenzo [b,f] [1,4] thiazepin-11-yl-1-piperazinyl) ethoxy]-ethanol fumarate (2:1) has international approvals for the treatment of schizophrenia as well as for the treatment of acute manic episodes associated with bipolar I disorder as either monotherapy or adjunct therapy to lithium or divalproex[1-3]. Analytical method reported in the literature for this drug include, a separation method for the determination of impurities by HPLC, a stability indicating reverse phase high-performance liquid chromatographic (RP-HPLC) method for the determination of quetiapine, and for the quantitative determination of quetiapine in biological samples[4-11]. None of

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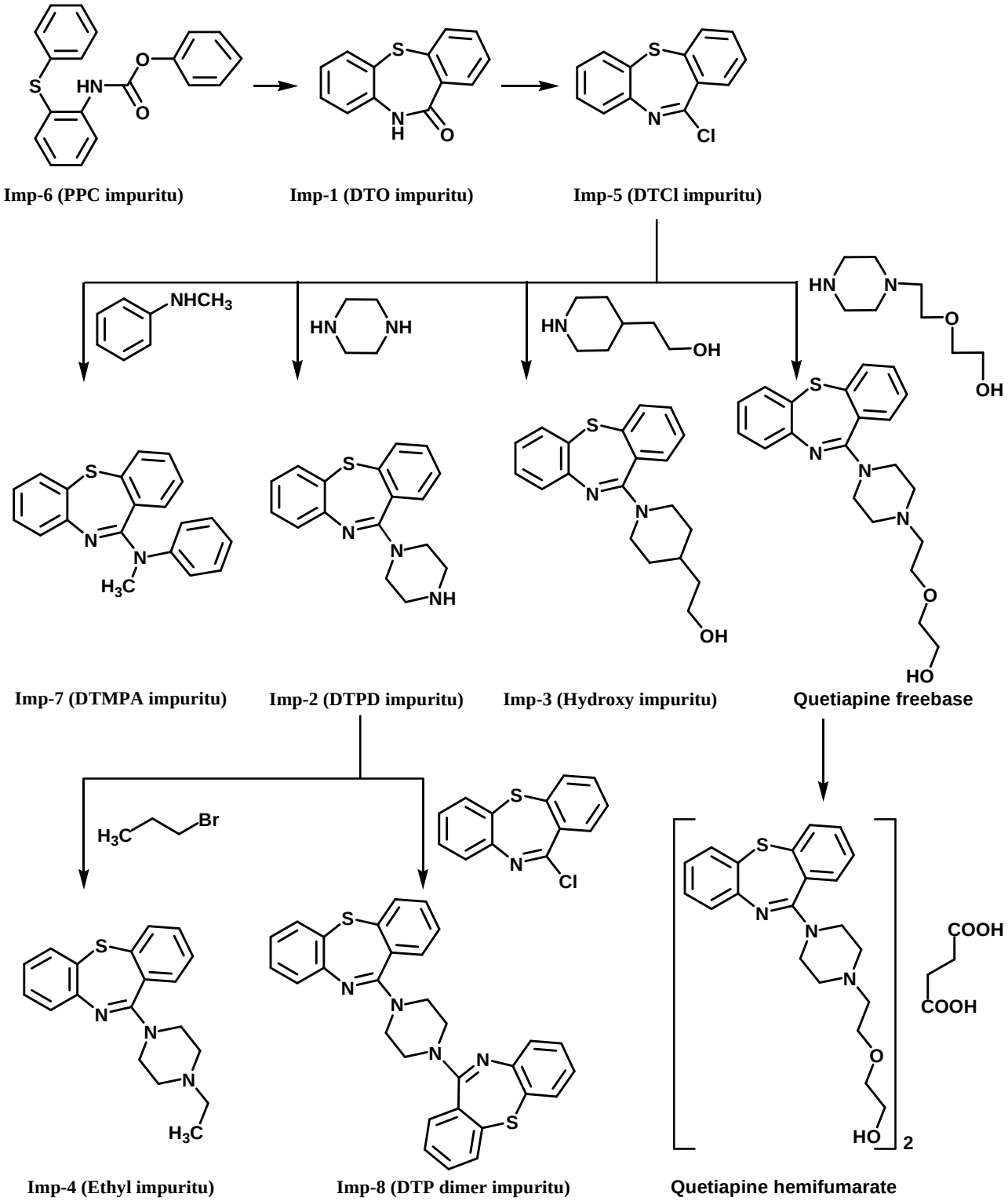

Figure 1 : Synthetic scheme for quetiapine hemifumarate with formation of its process related impurities

these reported methods were found to specific and stability indicating to all the eight impurities and hence a need of such a method aroused.

An impurity is a characteristic of the process and behaves as a fingerprint for the product. Development of an accurate analytical method eluting all process related impurities with good peak separation followed by its validation is a very critical activity during the devel-

opment. Moreover the method should be stability indicating to monitor the integrity of the sample throughout its lifecycle. During the process development activity for quetiapine in our laboratory[12], the analysis of crude quetiapine hemifumarate generated has shown eight process related impurities viz., dibenzo [b,f] [1,4] thiazepin-11(10H)-one **(Imp-1)**, 11-piperazin-1-yldibenzo [b,f] [1,4] thiazepine **(Imp-2)**, 2-(4-dibenzo [b,f] [1,4] thiazepin-11-ylpiperazin-1-yl)ethanol **(Imp-3)**, 11-(4-ethylpiperazin-1-yl)dibenzo [b,f] [1,4] thiazepine **(Imp-4)**, 11-chlorodibenzo [b,f] [1,4] thiazepine **(Imp-5)**, phenyl [2-(phenylthio)phenyl] carbamate **(Imp-6)**, N-methyl-N-phenyldibenzo [b,f] [1,4] thiazepin-11-amine **(Imp-7)**, and 1,4-bis[dibenzo [b,f] [1,4] thiazepine-11-yl] piperazine **(Imp-8)**, whose area percentage ranged from 0.05 to 0.2% consistently. All these impurities are identified, synthesized and characterized with the help of different spectroscopic techniques. Isocratic reverse phase liquid chromatography method developed initially was not able to elute Imp-8 due to its non-polar nature and to the best of our knowledge **(Imp-7)** is not reported anywhere. Also the literature methods[9-11] were found to be limited for separation and elution of some of these impurities in total. In the present study, attempts were made to develop a simple, more precise and accurate stability-indicating HPLC method, which can separate and elute all the eight potential impurities **(Imp-1 to Imp-8)** generated during the synthesis and degradation impurities formed during the forced decomposition studies of quetiapine hemifumarate. The developed method was validated to ensure the compliance in accordance with USP and ICH guideline[13,14].

TABLE 1 : Summary of forced degradation results

Stress condition	Time	Percentage assay of active substance	Mass balance[a]	Remarks
Acid hydrolysis (5 N HCl, refluxed in oil bath for 24 h)	1 day	89.38	99.40	Unknown degradation products formed
Base hydrolysis (5 N NaOH, refluxed in oil bath for 24 h)	1 day	99.93	99.93	No degradation products formed
Oxidative hydrolysis (30% H_2O_2 at RT)	2 h	85.03	99.30	Unknown degradation products formed
Thermal at 105°C	8 days	99.34	99.34	No degradation products formed
Phololytic degradation	8 days	99.63	99.90	No degradation products formed

[a]**Mass balance= % assay + % sum of all impurities + % sum of all degradants**

TABLE 2a : Precision results of Imp-1, Imp-2, Imp-3 and Imp-4 in spiked test preparations

Test preparation	Precision for percent impurities in spike test preparation							
	MP	IP	MP	IP	MP	IP	MP	IP
	Imp-1	Imp-1	Imp-2	Imp-2	Imp-3	Imp-3	Imp-4	Imp-4
1	0.187	0.204	0.179	0.179	0.169	0.188	0.155	0.162
2	0.186	0.170	0.176	0.157	0.171	0.164	0.154	0.141
3	0.184	0.178	0.176	0.177	0.170	0.172	0.153	0.154
4	0.185	0.182	0.178	0.167	0.169	0.176	0.154	0.153
5	0.188	0.180	0.180	0.173	0.171	0.172	0.156	0.155
6	0.185	0.182	0.176	0.169	0.170	0.172	0.153	0.155
Mean $\bar{x}$ (n=6)[a]	0.186	0.183	0.178	0.170	0.170	0.174	0.154	0.153
± S.D. (n=6)	0.001	0.011	0.002	0.008	0.001	0.008	0.001	0.007
% RSD (n=6)	0.54	6.01	1.12	4.71	0.59	4.60	0.65	4.58
Mean $\bar{x}$ (n=12)	0.184		0.174		0.172		0.154	
± S.D. (n=12)	0.008		0.007		0.006		0.005	
Overall % RSD(n=12)	4.35		4.02		3.49		3.25	

EXPERIMENTAL

Chemicals

Sample of Quetiapine hemifumarate and its eight potential process related impurities (Figure 1) were received from synthetic laboratory of Megafine Pharma (P) Ltd, Nashik, India. HPLC grade acetonitrile was purchased from Qualigen fine chemicals, Mumbai, India. Potassium dihydrogen orthophosphate was purchased from Merck, Mumbai, India. 1-Pentane sulphonic acid sodium salt was purchased from Merck, Mumbai, India. Potassium hydroxide was purchased from Merck, Mumbai, India. High pure water was prepared by using Millipore Milli Q plus purification system.

Equipment

The HPLC system used for method development, forced degradation studies and method validation was Agilent 1200 series (manufactured by Agilent technologies, 76337 Waldbronn Germany) with an Agilent photodiode array detector (PDA) and variable wavelength detector (VWD). The output signal was monitored and processed using Ezchrome Elite software version 3.2.1. The Zorbax Eclipse XDB C18 (250 mm length×4.6 mm ID, 5µm particle size) column has been procured

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from Agilent technologies made in USA and used for the method development, forced degradation and method validation studies.

Chromatographic conditions

(a) Mobile phase preparation

The mobile phase solvent-A consists of a mixture of 0.025M potassium dihydrogen orthophosphate and 0.01M 1-pentane sulphonic acid sodium salt in water (pH adjusted to 6.6 with 10% potassium hydroxide solution). The mobile phase solvent-B consists of a mixture of acetonitrile, methanol and water in the ratio of 45:45:10 v/v.

(b) Chromatographic procedure

The flow rate of the mobile phase was kept at 1.0 mL min^{-1}. The HPLC gradient was set as: T/%B: 0/45, 5/45, 40/90, 55/90, 58/45 and 65/45. The column temperature was maintained at 40°C and the detection wavelength was 250 nm. The test concentration was about 1.0 mg mL^{-1} (i.e. 1000 µg mL^{-1}) and the injection volume was 20 µL for related substances determination. A degassed mixture of water and acetonitrile (20:80 v/v) was used as diluent during the standard and test sample preparation.

Preparation of solutions

A working standard solution of 1000 µg mL^{-1} and test sample solution of 1000 µg mL^{-1} was prepared for the determination of related substances analysis. A stock solution of impurity (mixture of **(Imp-1)** to **(Imp-8)**) at 150 µg mL^{-1} was also prepared in diluent.

Analytical method validation protocol

Specificity

Specificity is the ability of the method to measure the analyte response in the presence of its potential impurities and degradation products. The specificity of the developed HPLC method for quetiapine hemifumarate was carried out in the presence of its eight impurities viz., **(Imp-1)** to **(Imp-8)** as shown in figure 1.

Stress degradation studies were performed on bulk drug substance to provide an indication of the stability-indicating property and specificity of the established method. Intentional stress conditions, such as photolytic degradation, thermal degradation (drug substance exposed in solid state at 105°C), acid hydrolysis (using 5M hydrochloric acid, sample was refluxed in oil bath for 1 days), base hydrolysis (using 5M sodium hydroxide, sample was refluxed in oil bath for 1 days) and oxidative degradation (using 30% hydrogen peroxide

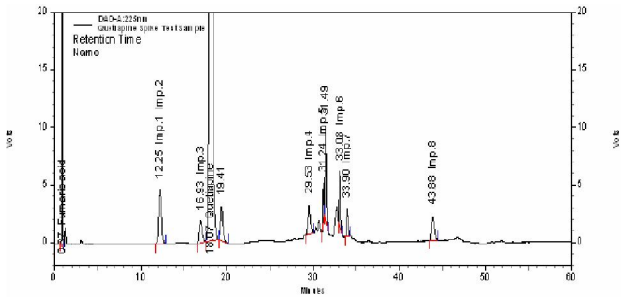


Figure 2 : HPLC chromatograms of sample solution spiked with impurities (conditions: Mobile phase-A: mix. of Phosphate buffer / Acetonitrile 90:10v/v, pH 6.7 with OPA, Mobile phase-B: Acetonitrile, Gradient elution, flow 1.5 ml/min, 45°C, 225 nm, YMC Pack-C8, 150×4.6 mm, 5µm, run time 60 min)

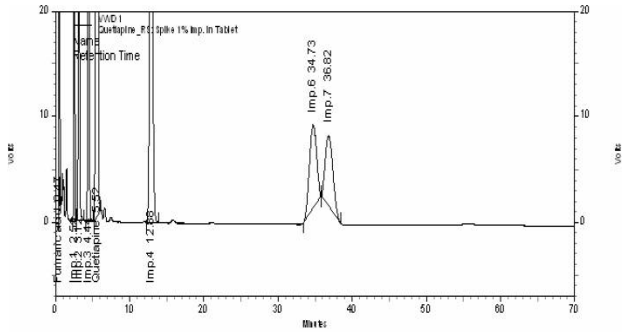


Figure 3 : HPLC chromatograms of standard solution (conditions: 0.01M KH2PO4/MeOH/ Acetonitrile, 500:400:100 v/v/v, buffer pH 7.0 with OPA, flow 1.0 ml/min, 45°C, 225 nm, Zorbax XDB , 50mm×4.6 mm, 1.8µm, run time 70 min)

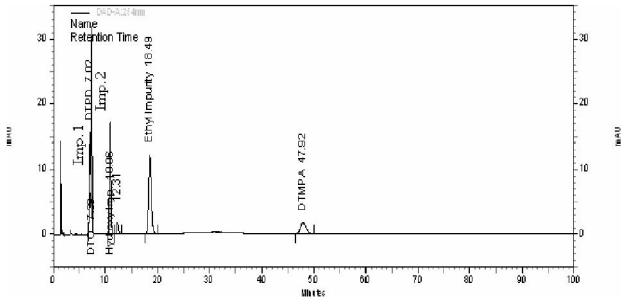
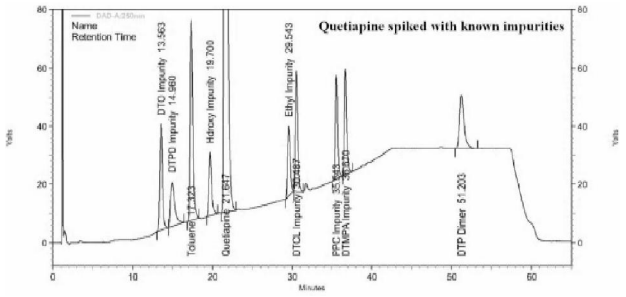
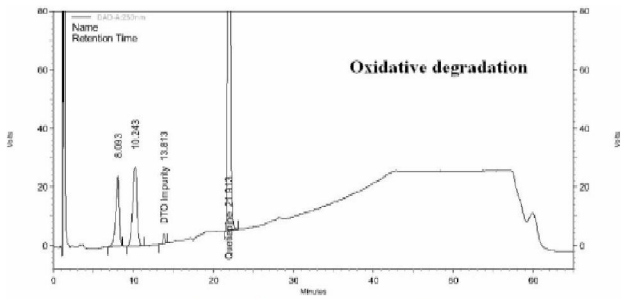


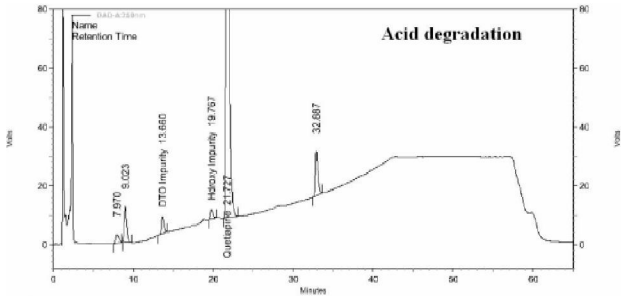
Figure 4 : HPLC chromatograms of standard solution (conditions: MeOH/Water/Acetonitrile/ Triethylamine, 500:400:100:0.4 v/v/v, buffer pH 7.0 with OPA, flow 1.5 ml/min, 25°C, 254 nm, C8-Novapak , 250mm×4.6 mm, 5µm, run time 100 min)



(a) Chromatogram of quetieapine spiked with known impurities



(b) Chromatogram of quetiapine test sample in oxidative degradation



(c) Chromatogram of quetiapine test sample in acid degradation

Figure 5 : HPLC chromatograms of; a) quetiapine spiked with known impurities (Imp-1 to Imp-8), b) oxidative degradation, c) acid degradation

at RT) to evaluate the ability of the proposed method to separate quetiapine from its degradation products were applied as per ICH recommendations[14]. The period for acid hydrolysis, base hydrolysis and oxidative degradation studies was 1 day, 1 day and 2 h respectively, where as for photolytic and thermal degradation, the period was 8 days. Photodiode array detector was employed to check and ensure the homogeneity and purity of quetiapine peak in all the stressed sample solutions. Assessment of mass balance in the degraded samples was carried out to confirm the amount of impurities detected in stressed samples matches with the amount present before the stress was applied[15]. Quantitative determination of quetiapine was carried out in all the stressed samples against the qualified working standard and the mass balance (% assay + % sum of all impurities + % sum of all degradation products) was tabulated (TABLE 1).

TABLE 2b : Precision results of Imp-5, Imp-6, Imp-7 and Imp-8 in spiked test preparations

Test preparation	Precision for percent impurities in spike test preparation							
	MP	IP	MP	IP	MP	IP	MP	IP
	Imp-5	Imp-5	Imp-6	Imp-6	Imp-7	Imp-7	Imp-8	Imp-8
1	0.142	0.155	0.170	0.189	0.158	0.168	0.143	0.157
2	0.141	0.131	0.169	0.159	0.157	0.147	0.139	0.152
3	0.140	0.142	0.168	0.172	0.156	0.157	0.140	0.165
4	0.142	0.142	0.171	0.171	0.157	0.163	0.145	0.16
5	0.143	0.143	0.172	0.173	0.159	0.158	0.148	0.162
6	0.141	0.143	0.169	0.173	0.157	0.157	0.148	0.164
Mean x^- (n=6)	0.142	0.143	0.170	0.173	0.157	0.158	0.144	0.160
± S.D. (n=6)	0.001	0.008	0.001	0.010	0.001	0.007	0.004	0.005
% RSD (n=6)	0.70	5.59	0.59	5.78	0.64	4.43	2.78	3.13
Mean x^- (n=12)	0.142		0.171		0.158		0.152	
± S.D. (n=12)	0.005		0.007		0.005		0.009	
Overall % RSD (n=12)	3.52		4.09		3.16		5.92	

[a]**Number of individual results, MP = Method Precision, IP= Intermediate precision**

TABLE 3 : Reports of Linearity results in the form of correlation coefficient intercept and slope

Sr. no.	Name of Impurities	correlation coefficient (r)	Slope	Intercept
1	Imp-1	0.99991	63654.46	619.27
2	Imp-2	0.99928	47461.60	286.73
3	Imp-3	0.99996	39532.38	-182.65
4	Imp-4	0.99994	51259.54	-522.94
5	Imp-5	0.99997	101227.63	-372.39
6	Imp-6	0.99958	50368.69	2485.29
7	Imp-7	0.99996	49992.03	-63.67
8	Imp-8	0.99941	48163.65	3273.93

Precision

The precision of the related substance method was checked by injecting six individual preparations of quetiapine hemifumarate spiked with 0.15% of all the eight impurities **(Imp-1)** to **(Imp-8)** with respect to target analyte concentration (i.e. 1.0 mg mL^{-1}). The percentage of added impurities in six different spiked test

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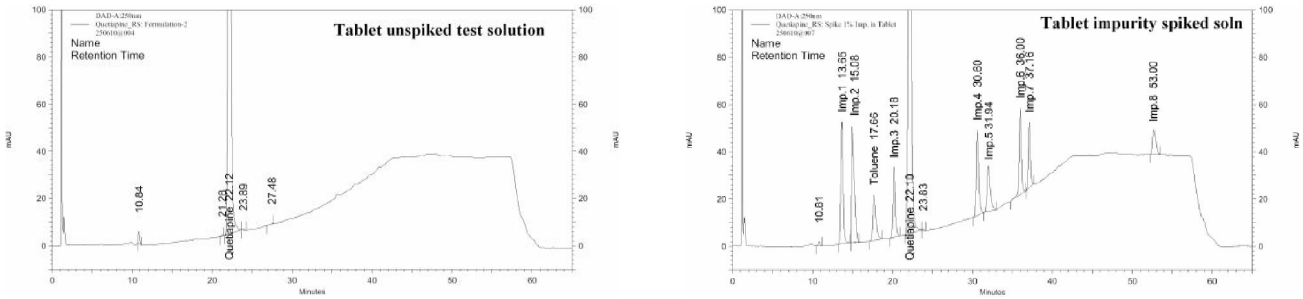

Figure 6 : HPLC chromatograms of formulation sample; a) Tablet solution unspiked, b) Tablet spiked with known impurities (Imp-1 to Imp-8)

TABLE 4 : Results of formulated tablet analysis and bulk drug batches sample analysis

Sample source	Formulation product analysis results (%)		Bulk drug batch analysis results (%)		
	Formulation-1	Formulation-2	Batch-1	Batch-2	Batch-3
Imp.1	ND	ND	0.01	0.01	0.02
Imp.2	ND	ND	0.01	0.01	0.02
Imp.3	ND	ND	0.02	0.02	0.02
Imp.4	ND	ND	ND	ND	ND
Imp.5	ND	ND	ND	ND	ND
Imp.6	ND	ND	ND	ND	ND
Imp.7	ND	ND	ND	ND	ND
Imp.8	ND	ND	ND	ND	ND
SMUI	0.06	0.08	0.02	0.09	0.01
Total impurities	0.08	0.11	0.06	0.24	0.07

ND = Not detected. SMUI= Single maximum unknown impurity

preparations was calculated. The RSD was calculated for percentage of each impurity **(Imp-1)**, **(Imp-2)**, **(Imp-3)**, **(Imp-4)**, **(Imp-5)**, **(Imp-6)**, **(Imp-7)** and **(Imp-8)** in each spiked test preparation.

The intermediate precision of the method was also verified using different analyst, different day and different instrument number in the same laboratory. The results of method precision and intermediate precision were expressed in terms of percentage of impurities in six spiked test preparation. The percentage RSD for results of intermediate precision and results of method precision was calculated and compared with each other.

Limit of detection (LOD) and limit of quantification (LOQ)

The LOD and LOQ of all the eight impurities were determined using calibration curve method according to ICH Q2R1 recommendations by establishing the lowest concentration that can be measured. Precision study was also carried out at the LOQ level by injecting six individual preparations of all the impurities and calculating RSD of the area.

Linearity

The linearity of an analytical procedure is its ability (within a given range) to obtain test results, which are directly proportional to the concentration of the analyte in the sample. Linearity test solutions for related substance method were prepared by diluting the impurity stock solution (as described above) to the required concentrations. The solutions were prepared at six concentration levels from LOQ to 250% with respect to the impurities specification level of 0.15% (i.e. LOQ. 0.075, 0.150. 0.225, 0.300 and 0.375%). The calibration curve was drawn by plotting the peak areas of each impurity versus its respective concentration. The correlation coefficient, intercept and slope of the calibration curve were calculated & reported.

Accuracy

The accuracy of an analytical procedure expresses the closeness of agreement between the value, which is accepted either as a conventional true value or an accepted reference value and the value found. The recovery experiments were conducted to determine accuracy of the related substance method for the quantification of all eight impurities in bulk drug samples. The study was carried out in triplicate at four concentration levels i.e. LOQ, 0.075, 0.15 and 0.225 % of the analyte concentration (1000 $\mu g\ mL^{-1}$). The percentage recoveries were calculated by using formula eq. (A1):

$$\% \text{ Recovery} = \frac{\text{Amount recovered}}{\text{Amount added}} \times 100 \qquad \text{(A1)}$$

Robustness

To evaluate the robustness of the developed method,

the chromatographic conditions were deliberately altered and the resolution between closely eluting impurities i.e. **(Imp-1)** and **(Imp-2)**, **(Imp-4)** and **(Imp-5)**, was evaluated. The flow rate of the mobile phase was 1.0 mL min^{-1}. To study the effect of flow rate on the resolution, the same was altered by 0.1 units i.e. from 0.9 to 1.1 mL min^{-1}. The effect of column temperature on resolution was studied at 37 and 43°C instead of 40°C, also the effect of buffer pH ± 0.1 (6.5 and 6.7 instead of 6.6) on resolution was studied. All the other mobile phase components were held constant as described above.

RESULTS AND DISCUSSION

Development of the HPLC method

Before starting the development of new method, we checked the literature methods for their efficiency in eluting all the eight impurities. According to method[9], co-elution of **(Imp-1)** with **(Imp-2)** is observed as shown in figure 2, while another method[10] was not able to elute the **(Imp-8)** within the 70 minute run time and resolution between **(Imp-6)** and **(Imp-7)** was less than 1.1 as shown in figure 3. Also the Indian Pharmacopeia's method[11] was found to be limited to separate the **(Imp-1)** and **(Imp-2)** (resolution less than 1.2) and the method was not capable to elute **(Imp-8)** within the 100 minute run time (Figure 4). The new method developed in our laboratory showed all the eight impurities of quetiapine hemifumarate satisfactorily (Figure 5).

The main target of developing the chromatographic method is to detect and determine the potential impurity **(Imp-8)** and a new impurity **(Imp-7)** present in bulk samples produced by Megafine Pharma (P) Ltd., and to achieve the separation of all eight impurities (Figure 1) along with the degradation products generated during stress studies from the analyte peak. During the column selection, we observed the co-elution of impurities when different stationary phases like C18, C8, Cyano and Phenyl and different mobile phases containing buffers like phosphate, sulphate and acetate with different pH (3.0-7.0) and various organic modifiers like acetonitrile, methanol and tetrahydrofuran in the mobile phase with different ratios are used. Apart from the co-elution of impurities poor peak shapes for some impurities and quetiapine were also noticed during the development phase. Satisfactory chromatographic separation was achieved on Zorbax XDB C18 column using mobile phase consisting a mixture of 0.025M potassium dihydrogen orthophosphate and 0.01M 1-pentane sulphonic acid sodium salt dihydrate solution in water (pH adjusted to 6.6 with 10% potassium hydroxide solution) as solvent A and a mixture of acetonitrile, methanol and water in the ratio of 45:45:10 v/v as solvent B. The **(Imp-2)** is critical to the pH of mobile phase solvent-A; it was co-eluted with **(Imp-1)** when pH of mobile phase solvent-A was below 6.4 and it was co-eluted with an **(Imp-3)** when pH of mobile phase solvent-A was above 6.8 and hence the pH range of 6.4 to 6.8 was essential to maintain the separation of **(Imp-2)** peak. The flow rate of the mobile phase was kept at 1.0 mL min^{-1}. The HPLC gradient of mobile phase-B was kept as: T/%B: 0/45, 5/45, 40/90, 55/90, 58/45 and 65/45. In the optimized conditions the quetiapine and all the eight impurities were well separated with a resolution greater than 1.5. The typical retention times of **(Imp-1)**, **(Imp-2)**, **(Imp-3)**, quetiapine, **(Imp-4)**, **(Imp-5)**, **(Imp-6)**, **(Imp-7)**, and **(Imp-8)** were about 13.6, 15.0, 19.7, 21.7, 29.5, 30.5, 35.5, 36.7and 51.2 min respectively (Figure 5a), and the developed HPLC method was found to be specific for quetiapine and its eight impurities, namely **(Imp-1)**, **(Imp-2)**, **(Imp-3)**, quetiapine, **(Imp-4)**, **(Imp-5)**, **(Imp-6)**, **(Imp-7)**, and **(Imp-8)**. Separation of all the eight impurities in spiked formulations is shown in figure 6. An unknown impurity which eluted close to the impurity **(Imp-1)** was well separated under the optimized conditions confirming specificity and selectivity of the developed method.

Results of forced degradation studies

Degradation was not observed in quetiapine hemifumarate bulk samples under stress conditions such as photolytic stress, thermal stress and base hydrolysis whereas considerable degradation of the drug substance was observed under acid and oxidative hydrolysis that leads to the formation of some unknown degradation peaks (Figure 5b and Figure 5c). Peak purity test results obtained from PDA confirms that the quetiapine peak is homogeneous and pure in all the stress samples analyzed. The mass balance is a process of adding together the assay value and the levels of degradation

products to see how closely these add up to 100% of initial value with due consideration of the margin of analytical error[16]. The mass balance of stressed samples was close to 99.9% (TABLE 1). The percentage of quetiapine was unaffected by the presence of eight impurities (Figure 1), thus confirms the stability-indicating strength of the developed method.

Results of method validation experiments

(1) Precision

The RSD percentage of content of **(Imp-1)** to **(Imp-8)** in method precision study was within 2.8% and in intermediate precision study was within 5.8%. The results of method precision and intermediate precision are compared with each other. The overall RSD (n=12)[17] for percentage of impurities i.e. **(Imp-1)** to **(Imp-8)** were found within the range of 3.16-5.92. The precision results are reported in TABLE 2, confirming the good precision of the developed method.

(2) LOD and LOQ

The LOD of **(Imp-1)**, **(Imp-2)**, **(Imp-3)**, **(Imp-4)**, **(Imp-5)**, **(Imp-6)**, **(Imp-7)** and **(Imp-8)** were 0.002, 0.003, 0.001, 0.001, 0.002, 0.003, 0.002 and 0.002% respectively (of analyte concentration i.e. 1000 μg mL^{-1}). The LOQ of **(Imp-1)**, **(Imp-2)**, **(Imp-3)**, **(Imp-4)**, **(Imp-5)**, **(Imp-6)**, **(Imp-7)** and **(Imp-8)** were 0.005, 0.010, 0.004, 0.004, 0.007, 0.008, 0.007 and 0.006% respectively (of analyte concentration i.e. 1000 μg mL^{-1}). The LOQ precision for all impurities at LOQ level was below 10 % RSD.

(3) Linearity

Linear calibration plot for the related substance method was obtained over the calibration ranges tested, i.e. LOQ to 0.375% for **(Imp-1)** to **(Imp-8)**. The correlation coefficient obtained was greater than 0.999. The results demonstrate that an excellent correlation existed between the peak area and concentration of **(Imp-1)** to **(Imp-8)**. The correlation coefficient, intercept and slope for each impurity are shown in TABLE 3.

(4) Accuracy

The percentage recovery of Imp-1 to Imp-8 in bulk drugs samples ranged from 96.32 to 107.68%. HPLC chromatogram of all eight impurities spiked in quetiapine bulk drug sample was shown in figure 2a.

(5) Robustness

In all the deliberate varied chromatographic conditions (flow rate, column temperature and pH of buffer) the resolution between **(Imp-1)** and **(Imp-2)**, and **(Imp-4)** and **(Imp-5)** was greater than 1.5, while other impurities were greater than 2.0 illustrating the robustness of the method.

(6) Application of method

The results of the commercial formulation sample and bulk drug sample analysis are shown in TABLE 4. The values of impurities detected indicate that the method is capable of detecting known and unknown impurities in routine analysis of bulk drug and formulation sample.

CONCLUSIONS

The method presented in this communication describes the development of specific and accurate HPLC method that separates eight potential impurities **(Imp-1)** to **(Imp-8)** generated during the chemical synthesis and forced degradation studies of quetiapine hemifumarate. The developed method was validated to ensure the compliance in accordance with ICH guideline. The behavior of quetiapine hemifumarate under various stress conditions was studied. The developed method could be useful for monitoring quality of bulk samples and as well employed to check the quality during stability studies.

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