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A Novel Method Development and Validation by Ultra-Performance Liquid Chromatography for Assay of Asciminib in Dosage Form

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Abstract: The main objective of the present study was to develop and validate simple, precise, sensitive and accurate UPLC method for the estimation of asciminib in pure and dosage form. The UPLC method was developed by using Waters Acquity liquid chromatographic system and Zorbax Phenyl (100x1.0mm,1.7µm) column. The developed method was validated according to the international conference on harmonization (ICH) guidelines. The chromatographic separation of asciminib with good resolutions has been achieved by using the mobile phase acetonitrile: ammonium dihydrogen phosphate buffer (20:80 v/v) at a flow rate of 0.5 mL/min, injection volume of 5 µL, and at 242 nm wavelength. The validated method was found to be linear in the range of 4 - 60 µg/mL. The limit of detection (LOD) and limit of quantification (LOQ) for asciminib were found to be 0.4 and 0.12 µg/mL respectively. The % RSD was found to be less than 2 % revealing the precision of the developed method. Besides, the recovery rate was observed close to 100 % confirming the accuracy of the method. Minor alterations in the chromatographic conditions have revealed robustness and ruggedness of the developed method. The developed analytical method is simple, precise, sensitive, and reproducible which can be used for the estimation of asciminib.

Keywords: UPLC, Asciminib, Buffer, Method development, Forced degradation studies

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1. INTRODUCTION

Many modifications were made to the techniques of chromatography to overcome the shortcomings like analysis time and the range of compounds that could be detected. Application of pressure was practised by use of pumps to reduce the time of run (1). Technologies like electrochemical methods and spectroscopy were added to enhance detection. The functional efficiency of chromatographic techniques enhanced to a great extent with these developments and modifications and also the range and type of substances that could be analysed (2).

A liquid handling system that can run such columns at significantly higher pressures is now available, as well as chromatographic media with 1.7 µm particle size due to recent developments in pharmaceutical analysis. This technology, known as Ultra performance liquid chromatography (UPLC), has shown improvements in method sensitivity, resolution, and speed when compared to standard HPLC. It uses sub-2-micron particles and extremely high pressure (up to 100 MPa is achievable in UPLC system) (3). When compared to chromatographic systems using analytical columns filled with particles of 5 µm size, the UPLC system can reduce analysis time by up to nine times. Analysis time was reduced by roughly three times when compared to analytical columns packed with particles of 3 µm size (4).

There are numerous potential uses for the UPLC technique in the investigation of pharmaceutical and biological chemicals, and it is a field that is both well-established and rapidly expanding (5). UPLC technique can be used for the determination of particular biological and pharmaceutical compounds that can be determined using UPLC, together with ranges of their respective determination or detection limits and with other associated parameters (6).

Faster and greater UPLC separation can shorten the time needed for method development in research and development laboratories because most

pharmaceutical corporations want to cut expenses and time associated with research and development (7). With the introduction of UPLC, a novel liquid chromatography instrument system was created that increases throughput and analytical speed without sacrificing chromatographic performance (8).

UPLC systems reduce column re-equilibration time, which results in a significant reduction in solvent usage. Injection volume in UPLC is over 10 times less than in HPLC, leading to acceptable peak forms and little impacts linked to column diameter. With higher resolution and speed of LC analyses, UPLC boosts data quality and productivity by providing more data per unit of work (9).

The IUPAC name of asciminib was N-[4-[chloro(difluoro)methoxy] phenyl]-6-[(3R)- 3-hydroxypyrrolidin-1-yl]-5-(1H-pyrazol-5-yl) pyridine-3-carboxamide. The molecular formula of asciminib was $C_{20}H_{18}ClF_2N_5O_3$. Asciminib was a selective allosteric inhibitor of BCR/ABL1, strongly binding to the myristoyl pocket of ABL1 kinase. The GNF2 chemical asciminib had been found as having the unique ability to cause a significant bend in the domain's C-terminal helix. Asciminib inhibits downstream signalling by regaining the initial autoregulatory mechanism that caused the inactive conformation; however, it is 100 times more potent than other GNF2 drugs (10). More than 60 distinct kinases, including SRC kinases, G-protein receptors, ion channels, nuclear receptors, and transporters, were shown to be inactive against the medication in several tests (11). The National Comprehensive Cancer Network introduced asciminib as a new therapy option in 2021 after the US FDA granted it approval. The recommended dosage was 40 mg BID or 80 mg QD (12).

A comprehensive review of the literature on asciminib revealed no methods like HPLC, UPLC or LC-MS for determining asciminib in pharmaceutical formulations and bulk drugs. The goal of this study was to create and evaluate a stability-indicating RP-UPLC technique for determining asciminib in bulk and pharmaceutical dosage forms.

2. EXPERIMENTAL

2.1. Reagents and Chemicals

Asciminib (Figure 1) pure bulk drug (API), Asciminib dosage form tablets (Scemblix), acetonitrile, ortho-phosphoric acid, and ammonium dihydrogen phosphate were used. Merck provided all of the HPLC grade solvents. Whatman filter (0.22 µ) were employed to filter all of the solvents and solutions used. The asciminib drug sample was obtained from Shree Icon labs, Vijayawada, India as gift sample.

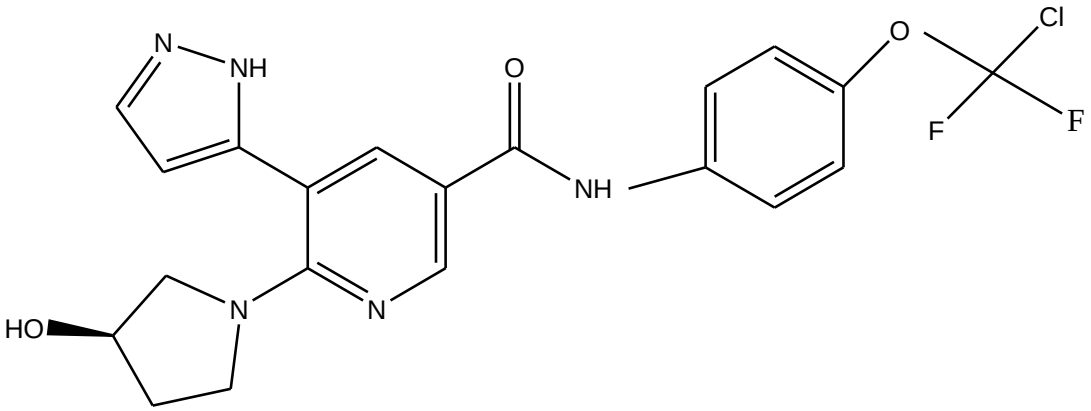

Figure 1: Structure of Asciminib.

2.2. Instrumentation and Conditions for Chromatography

The chemicals were weighed on an electronic balance-Denver. The pHs of the buffers were measured by means of pH meter -BVK enterprises. The solutions were sonicated or degassed using ultrasonicator-BVK enterprises. The chromatography analysis was performed using Waters Acquity liquid chromatographic system and the signal was detected using PDA detector with empower 2 software. UPLC method development and validation was carried out on a Phenyl (100x1.0mm,1.7µm) column, using a mobile phase of acetonitrile: ammonium dihydrogen phosphate buffer (20:80 v/v) with a flow rate of 0.5 mL/min. The sample injection volume was 5 µL. The column temperature was ambient. From the UV spectrum of asciminib, 242 nm wavelength was chosen which displays the maximum absorbance. The eluted compounds were monitored at 242 nm.

2.3. Preparation of Solutions

2.3.1. Buffer- Ammonium dihydrogen phosphate

Precisely weighed 1.15 grams of ammonium dihydrogen phosphate was transferred into thousand millilitres volumetric flask. Nine hundred millilitres HPLC grade water was added and sonicated. Lastly, orthophosphoric acid solution was used to correct the pH to 3.0 and the volume was made up with water.

2.3.2. Mobile phase

Mobile phase used for elution was acetonitrile: ammonium dihydrogen phosphate buffer in the ratio of 20:80 v/v. A 0.22 µ filter was used to filter the mobile phase. The diluent was chosen based on the drugs' solubility. Ethanol was used as diluent.

2.3.3. Preparation of standard stock solution

Precisely weighed 40 mg of asciminib was taken into a 100mL volumetric flask. 75% of the dilutants were put in the flask. Then, the flask was sonicated for ten minutes. The flask was then filled with diluent.

2.3.4. Preparation of standard working solution
Standard stock solution of 5 mL was transferred to a volumetric flask of 50 mL. 75% of the dilutants were put in the flask. Then, the diluent was used to make up to the volume.

2.3.5. Preparation of sample stock solution
The average weight of ten pills was determined. The weight comparable to 1 pill was weighed after the tablets were crushed into powder. The powder was then placed to a hundred millilitres volumetric flask. Diluents were used to make up to 100 mL of volume, which was then filtered using a 0.22 µ syringe filter.

2.3.6. Preparation of sample working solution
Filtrated sample stock solution of five millilitres was moved into a volumetric flask with a capacity of 50 mL. Then, the solution was prepared with made up with diluent.

2.4. Method Development and Validation Parameters
Mobile phase was pumped for around thirty minutes to saturate the column and the base line was adjusted. Various mobile phase ratios, buffers, and other parameters were changed to develop the method.

2.4.1. Linearity
Appropriate volumes of asciminib standard working solution were utilized to prepare seven standard drug concentrations covering the calibration range of 4 - 60 µg/mL. Each solution was injected into the instrument and peaks were recorded. The calibration curves were obtained by plotting peak area versus concentration. To statistically assess the linearity of the results, the correlation coefficient (R) and the linear regression equation were used.

2.4.2. Accuracy
Recovery studies have been used to verify the method's accuracy at 50%, 100%, and 150% level. A known amount of asciminib drug was spiked discretely to pre-analysed samples of the stated levels. The percent recovery of each level was calculated after each spiked level was administered into the UPLC system.

2.4.3. Precision
Method precision was executed by spiking the sample with asciminib at hundred percent of the quantified limit concerning the sample concentration in 6 preparations. Six homogenous replicates were injected. Then, the amount of asciminib was calculated to determine the percent relative standard deviation.

In six preparations, the intermediate precision was achieved by spiking the sample with asciminib at hundred percent of the prescribed limit in terms of sample concentration. The intermediate precision investigation was conducted on various days with various analysts.

2.4.4. LOD and LOQ
The limit of detection (LOD) and the limit of quantification (LOQ) were derived by means of the subsequent equations based on the slope of the calibration curve and the SD of linearity responses.

LOD = 3.3 × Standard deviation (SD) / slope
LOQ = 10 × Standard deviation (SD) / slope

2.4.5. System suitability parameters
Six replicates of asciminib working standard samples were injected to assess system suitability, and metrics such as tailing factor (K), relative retention time, plate number (N), resolution, and peak asymmetry of samples were investigated.

2.4.6. Robustness
Small adjustments in chromatographic settings such as flow (-) (0.45 mL/min), flow (+) (0.55 mL/min), organic phase (-) (18 O:82), and organic phase (+) (22 O:78) were used to test the method's robustness.

2.4.7. Specificity and selectivity
The absence of adjuvant interference during the application of the planned approach to the study of pharmaceutical formulations demonstrated its selectivity. The method's specificity was assessed in terms of interference caused by the occurrence of any additional placebos. Two dissimilar samples were administered and compared to their placebo counterparts.

2.5. Forced Degradation Studies
2.5.1. Peroxide degradation
The probable peaks and rates of asciminib degradation was assessed by weighing stock solution of 5 mL and transferring it into a volumetric flask of volume 50 mL. Then, it was subjected to forced degradation by adding 1mL of 30% H_2O_2; heating at 60 °C, and cooling to room temperature. After cooling, sample was diluted to volume with diluent and mixed. The samples were withdrawn at specific times (0, 6, 12, 18, and 24 hrs) intervals and subjected to UPLC runs.

2.5.2. Reduction degradation
Asciminib was tested for rate of degradation and potential degradation peaks. 5 mL of the stock solution was transferred into a fifty millilitres volumetric flask. Then, it was forcedly degraded by adding 1 mL of 30 % sodium bi sulphate and heated at 60 °C. Sample was diluted with diluent to volume after cooling, then mixed. Samples were taken out and processed through an UPLC at certain time intervals (0, 6, 12, 18, and 24 hours).

2.5.3. Acid degradation
Stock solution of 5 mL was weighed and put into a volumetric flask of 50 mL. Then, 1 millilitre of 1N HCl was added and the flask was heated for six hours and cooled to room temperature. After cooling, 1 mL of NaOH was added to neutralise the solution and sample was diluted with diluent and mixed. Samples were taken out and processed through an UPLC at certain time intervals (0, 6, 12, 18, and 24 hours).

2.5.4. Base degradation
The possible degradation peaks and rate of degradation of asciminib were assessed by weighing stock solution of 5 mL and transferring into a fifty

millilitres volumetric flask. Then, it was subjected to forced degradation. 1mL of 1N NaOH was added and heated at 60 °C and cooled to room temperature. After cooling, 1 mL of 1N HCl was added and diluted to volume with diluent. Samples were withdrawn at specific time (0, 6, 12, 18, and 24 hrs) intervals and subjected to UPLC runs.

2.5.5. Hydrolytic degradation

Asciminib was tested for rate of degradation and potential degradation peaks. 5 mL of the stock solution was transferred into a 50 mL volumetric flask. Then, it was forcedly degraded by adding 3 mL of HPLC grade water and heated at 60 °C for three hours. Sample was diluted with diluent to volume after cooling, then mixed. Samples were taken out and processed through an UPLC at certain time points (0, 6, 12, 18, and 24 hours).

2.5.6. Thermal degradation

Stock solution of 5 mL was weighed and shifted into a 50 millilitres volumetric flask. Diluents were added to this and sonicated for 15 min to dissolve the contents. This solution was heated for 48 hours. Samples were withdrawn at specific time (0, 6, 12, 18, and 24 hrs) points and subjected to UPLC runs to identify probable degradation chromatograms.

2.5.7. Photo stability degradation

The rate of degradation and possible degradation peaks of asciminib were assessed by weighing 250 mg of sample. The sample was placed in a photo stability chamber for 72 lux hours. 192 mg of the above sample was weighed and transferred to a 100 mL volumetric flask. Diluents were added to this and sonicated for 15 min to dissolve the contents. Further, from the above solution 5 mL was pipetted out into a volumetric flask of volume 50 mL and diluted to volume with diluent. Samples were withdrawn at specific time (0, 6, 12, 18, and 24 hrs) points and subjected to UPLC runs.

2.6. Assay of Asciminib

A total of 20 pills were weighed and ground to powder. Volumetric flask of volume 100 mL was filled with powder corresponding to 40 mg asciminib. 70 mL diluents were added, dissolved by sonication, diluted to volume using dilutants, and filtered by means of a 0.45 µm porosity membrane filter.

3. RESULTS AND DISCUSSION

3.1. Chromatographic Optimization

After a series of trials, the mobile phase of acetonitrile: ammonium dihydrogen phosphate buffer in the proportion of 20:80 had shown peak with good theoretical plate count, resolution, tailing factor. Hence this method was optimized and validated. Waters Acquity UPLC auto sampler enabled the elution, method development and validation of asciminib. The optimized chromatogram was shown in Figure 2.

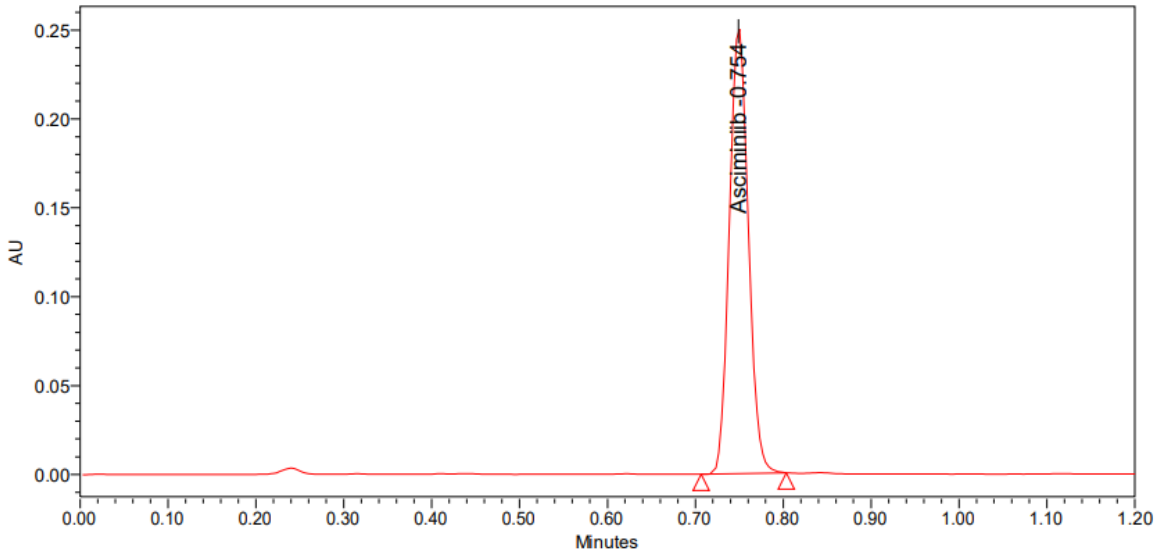

Figure 2: Optimized chromatogram of asciminib.

3.2. Method's Validation

3.2.1. Linearity

The analytical method's linearity was its capacity to produce test findings within a specified range that were directly proportional to the concentration of the analyte in the test sample (13). The regression line of analysis shows the relationship between concentration and peak area of asciminib. As a result, the findings revealed that the peak area and analyte concentration showed a strong correlation. The R high value indicated good linearity. The linearity of the analytical method was determined by seven concentration levels in the range of 4-60 µg/mL. The regression coefficient, y-intercept, and slope of regression line were calculated. The observed corelation coefficient value was 0.99982. The results were shown in Tables 1-2 and Figure 3.

3.2.2. Accuracy

The results of accuracy showed percentage recovery at all three levels in the range of 99.7–101.3%, and % RSD was 0.15 % as shown in Table 3. The percentage recovery and percent RSD values fell within the acceptable ranges of 98.0% to 102.0% and not more than 2.0%, respectively, demonstrating the method's suitability for routine drug analysis.

Table 1: Linearity studies of asciminib.

S. No	Concentration (µg/mL)	Peak area
1	4	326971
2	10	731459
3	20	1347935
4	30	2034756
5	40	2619390
6	50	3263458
7	60	3946340

Table 2: Optical characteristics of asciminib.

Parameters	Asciminib
Linearity (µg/mL)	4-60 µg/mL
Regression equation	y = 64712.07x + 46933.12
Slope	64712.07
Intercept	46933.12
Correlation coefficient (R)	0.99982

Table 3: Recovery studies of asciminib.

Recovery level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery
50%	20	19.93	99.7
	20	20.26	101.3
	20	20.09	100.5
100%	40	40.09	100.2
	40	40.00	100.0
	40	40.26	100.7
150%	60	60.27	100.5
	60	60.27	100.5
	60	60.57	101.0
Mean			100.5
SD			0.2
RSD %			0.15

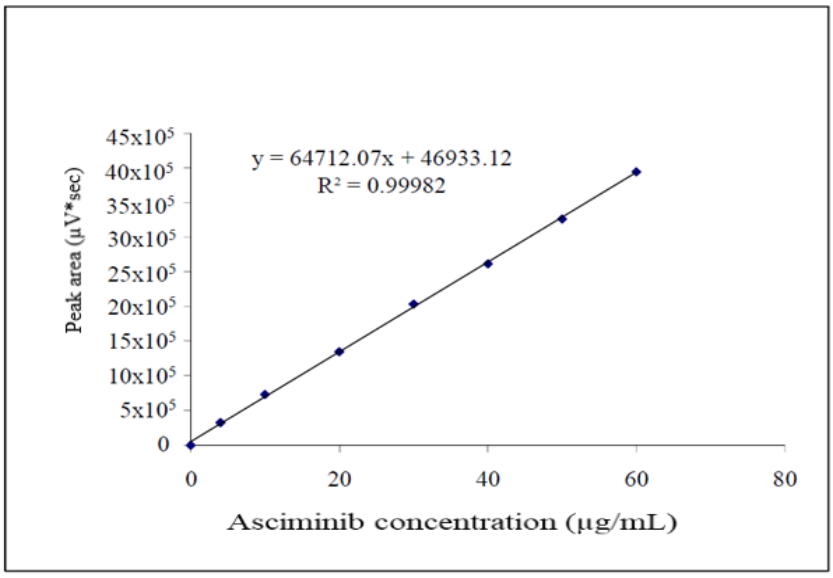

Figure 3: Calibration plot of asciminib.

3.2.3. Method precision

The degree of agreement between a set of measurements made using repeated samples of the same homogeneous material under the specified conditions was considered the precision of the method, and it was typically stated as relative standard deviation (13). The percent relative standard deviation value for method precision results of asciminib was found to be 0.38%. The percent relative standard deviation value for intermediate

precision results of asciminib was found to be 0.74%. The results were well under the usually accepted 2 percent limit. As a result, the new method's precision has been confirmed. The results were showed in Tables 4-5.

Table 4: Method precision studies of asciminib.

S. No	Area of asciminib
1.	2622418
2.	2611582
3.	2614570
4.	2628269
5.	2615321
6.	2637651
Mean	2621635
S.D	9920
RSD%	0.38

Table 5: Intermediate precision studies of asciminib.

S. No	Area of asciminib
1.	2653214
2.	2613206
3.	2615427
4.	2626539
5.	2600362
6.	2641488
Mean	2625039
S. D	19535
RSD%	0.74

3.2.4. Sensitivity

The limit of quantification (LOQ) was defined as the lowest quantity of analyte in a sample that was quantitatively identified with appropriate accuracy. The limit of detection (LOD) was defined as the lowest amount of analyte in a sample that was detected but not necessarily quantitated (13). The LOD and LOQ for asciminib were found to be 0.12 and 0.4 µg/mL respectively.

3.2.5. System suitability parameters

Analytical processes included testing for system compatibility. According to the ICH, a system suitability test was frequently used to assess a chromatographic system's resolution, column efficiency, and repeatability to ensure it was suitable for specific analysis. (13). The new approach was tuned to produce a symmetrical peak and high theoretical plates (N). The total number of theoretical plates was above 2000, which was deemed sufficient for the system suitability test. According to the standards, the tailing factor was within the specified limitations. These findings demonstrate that the proposed strategy can produce data of acceptable quality. The results were tabulated in Table 6.

Table 6: System suitability parameters for asciminib.

S. No	RT (min)	USP Plate Count	Tailing
1.	0.754	5634	1.08
2.	0.757	5623	1.12
3.	0.750	5580	1.10
4.	0.752	5600	1.11
5.	0.755	5558	1.09
6.	0.759	5567	1.09
Mean	0.755	5594	1.10

3.2.6. Robustness

The influence of slight alterations in chromatographic settings was used to determine the robustness of the analytical process. The percent RSD of assay of asciminib was less than 2.0 in all of the deliberately changed chromatographic settings. The system suitability parameters were not changed while varying the conditions, hence the method was robust. The results were showed in the Table 7.

3.2.7. Specificity and selectivity

The method's specificity and selectivity were tested by looking for interference peaks in the chromatograms of blank and placebo samples. In the retention time ranges, the UPLC chromatograms for the drug matrix (combination of the medicine and placebos) revealed nearly no interference peaks. As a result, the proposed UPLC approach in this study was selective. Figures 4 and 5 show the chromatograms of blank and working placebo solutions, respectively.

Table 7: Robustness studies of asciminib.

Condition	Retention time (min)	Peak tailing	Plate count	%RSD of assay of asciminib
Flow rate (-) 0.45 mL/min	0.925	1.12	6510	0.72
Flow rate (+) 0.55 mL/min	0.631	1.11	4361	0.21
Mobile phase (-) 18O:82B	0.903	1.14	5702	0.21
Mobile phase (+) 22O:78B	0.625	1.10	5588	0.21

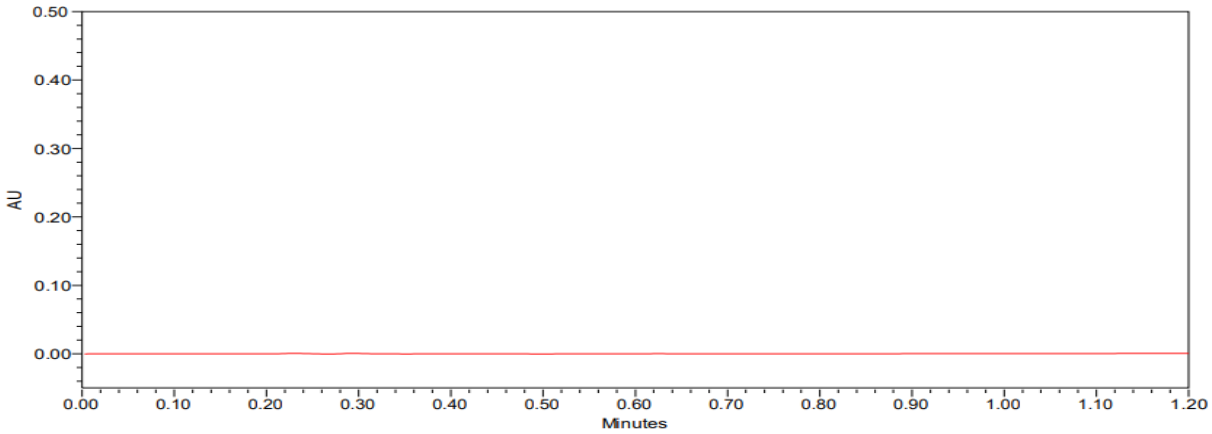

Figure 4: Chromatogram of blank.

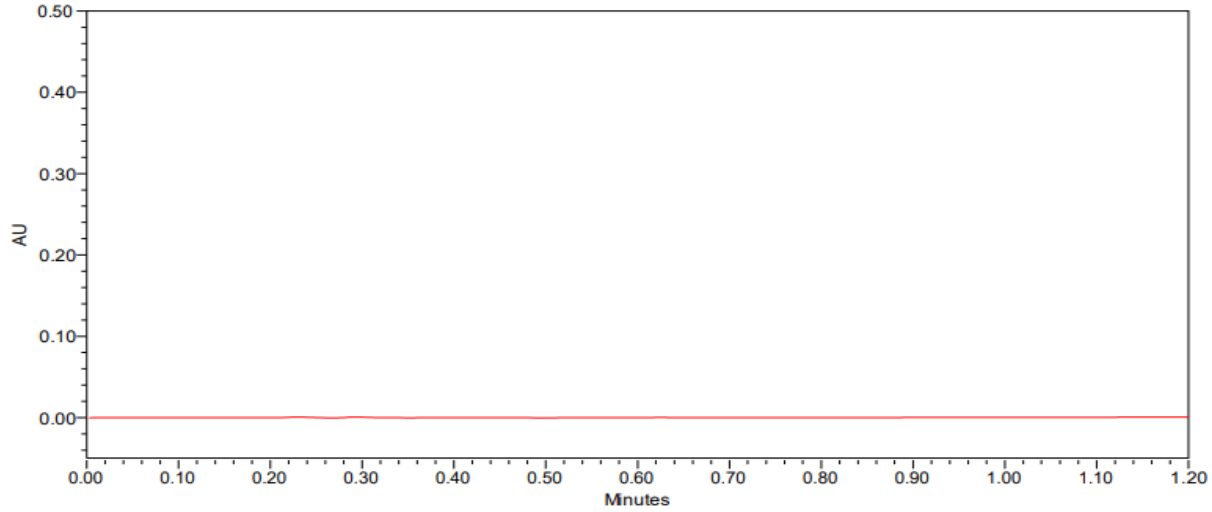

Figure 5: Chromatogram of placebo.

3.2.8. Forced degradation studies

Asciminib was subjected to a variety of stress conditions, which include hydrolysis, base, oxidative, acid, photo stability, and thermal degradation, as per ICH guidelines. The proposed UPLC approach was used to monitor degradation behaviour on a regular basis. The TUV detector results from the forced deterioration results revealed that the asciminib peaks were pure and homogenous in all of the stressful conditions studied. Also, the drug was more degraded in peroxide degradation and less degraded in hydrolysis degradation conditions. Degradant peaks were eluted in acid, thermal, peroxide and base conditions. All the results of stability studies were displayed in Tables 8-14. The chromatogram peaks of degradation studies were showed in Figures 6-11.

Table 8: Acid degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2524786	96.4	3.6	18.137	72.185
6 hrs	2356301	90.0	10.0	18.133	72.184
12 hrs	2236589	85.4	14.6	18.139	72.185
18 hrs	2108547	80.5	19.5	18.901	72.554
24 hrs	1998549	76.3	23.7	18.905	72.505

Table 9: Base degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2538564	96.9	3.1	18.134	72.186
6 hrs	2384154	91.0	9.0	18.142	72.187
12 hrs	2263052	86.4	13.6	18.131	72.179
18 hrs	2138547	81.7	18.3	18.133	72.175
24 hrs	2066953	78.9	21.1	18.656	72.763

Table 10: Peroxide degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2510547	95.9	4.1	18.366	72.528
6 hrs	2330256	89.0	11.0	18.354	72.517
12 hrs	2228795	85.1	14.9	18.359	72.514
18 hrs	2058961	78.6	21.4	18.363	72.527
24 hrs	1975896	75.4	24.6	18.368	72.521

Table 11: Reduction degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2548594	97.3	2.7	18.138	72.186
6 hrs	2406845	91.9	8.1	18.145	72.183
12 hrs	2279658	87.0	13.0	18.614	72.185
18 hrs	2175478	83.1	16.9	18.137	72.172
24 hrs	2120563	81.0	19.0	18.737	72.625

Table 12: Thermal degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2566854	98.0	2.0	18.136	72.181
6 hrs	2500210	95.5	4.5	18.144	72.188
12 hrs	2360325	90.1	9.9	18.143	72.188
18 hrs	2285968	87.3	12.7	18.603	72.146
24 hrs	2245173	85.7	14.3	18.603	72.151

Table 13: Photolysis degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2571578	98.2	1.8	18.133	72.152
6 hrs	2564587	97.9	2.1	18.158	72.167
12 hrs	2535214	96.8	3.2	18.172	72.149
18 hrs	2495896	95.3	4.7	18.129	72.151
24 hrs	2445716	93.4	6.6	18.734	72.667

Table 14: Hydrolysis degradation studies of asciminib.

Time	Peak area	% Label claim	% Degraded	Purity angle	Purity threshold
Initial	2585646	98.7	1.3	18.157	72.182
6 hrs	2554756	97.6	2.4	18.162	72.154
12 hrs	2543021	97.1	2.9	18.155	72.136
18 hrs	2521053	96.3	3.7	18.117	72.147
24 hrs	2505231	95.7	4.3	18.739	72.526

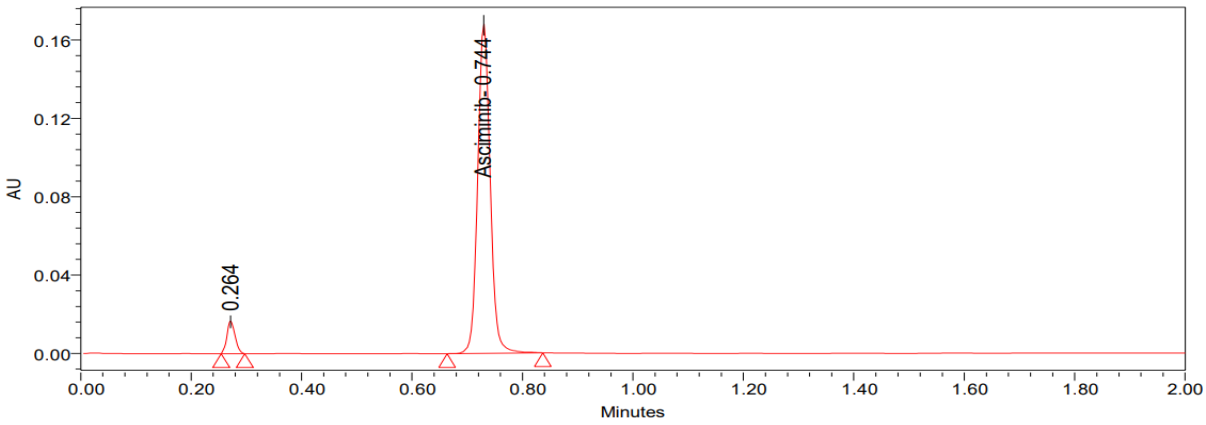

Figure 6: Acid degradation chromatogram.

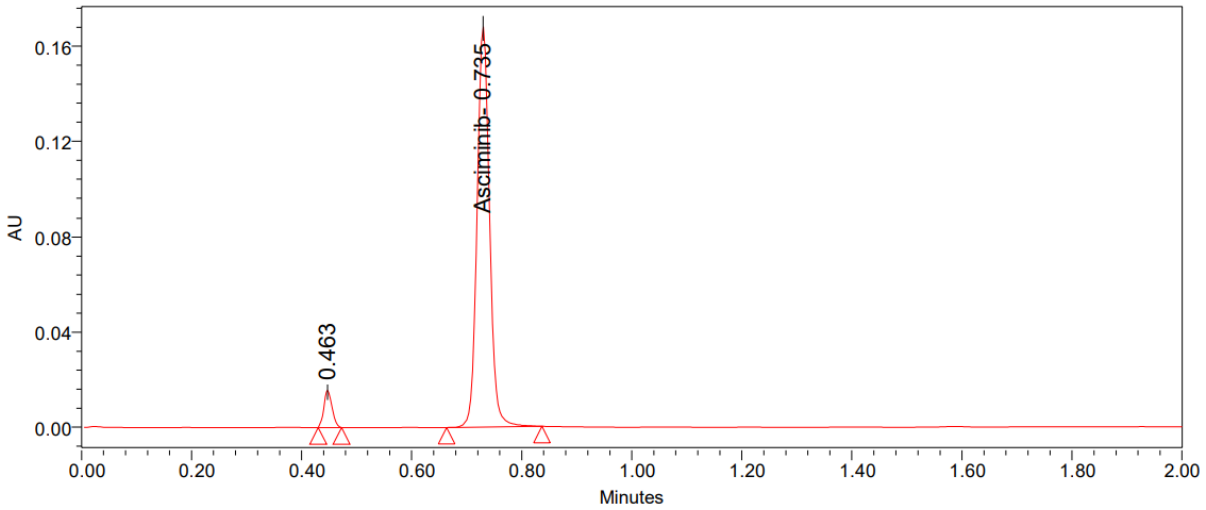

Figure 7: Base degradation chromatogram.

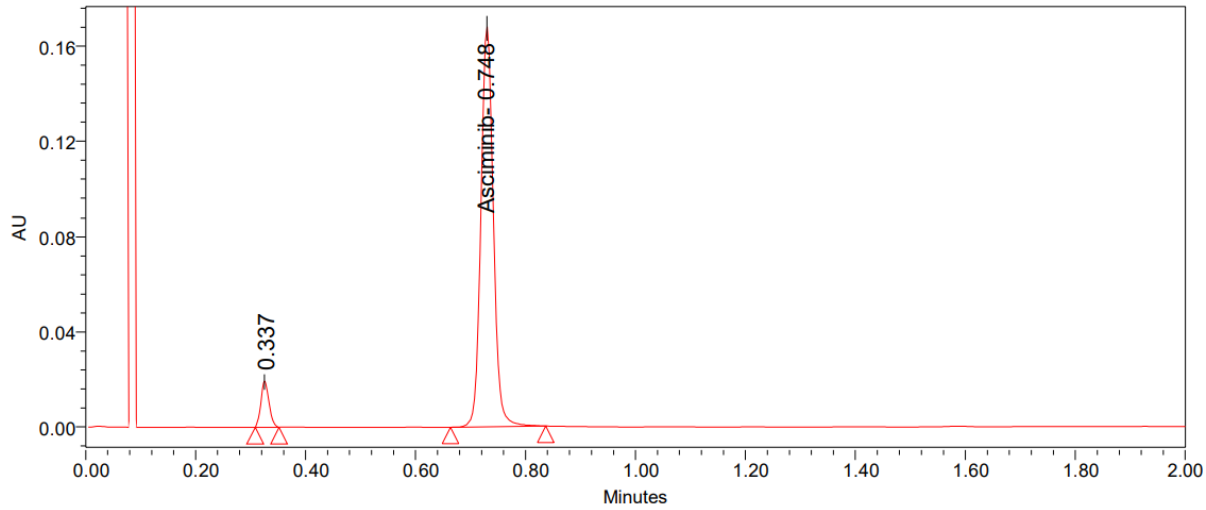

Figure 8: Peroxide degradation chromatogram.

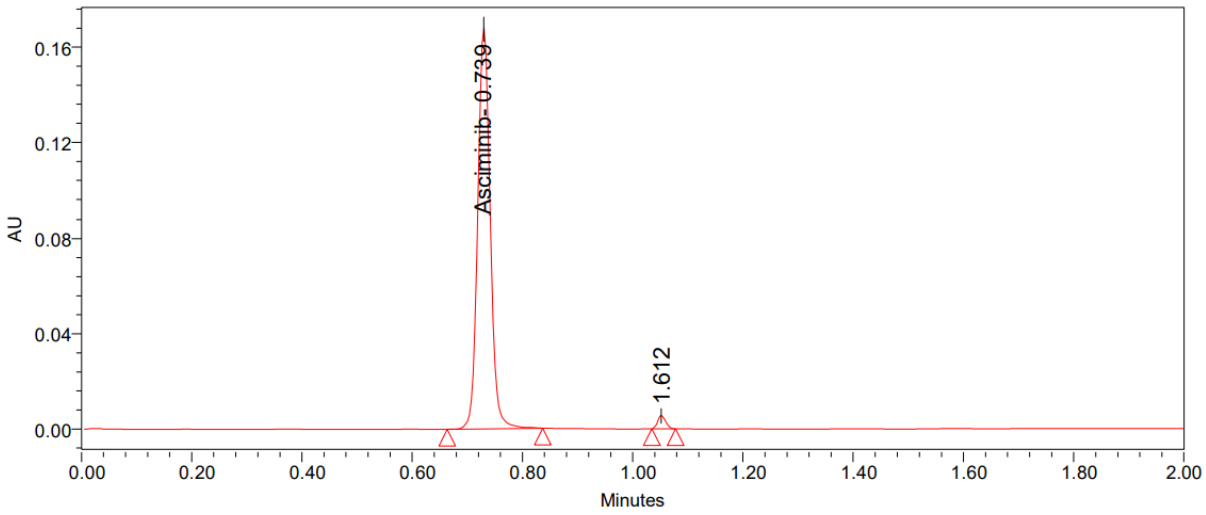

Figure 9: Thermal degradation chromatogram.

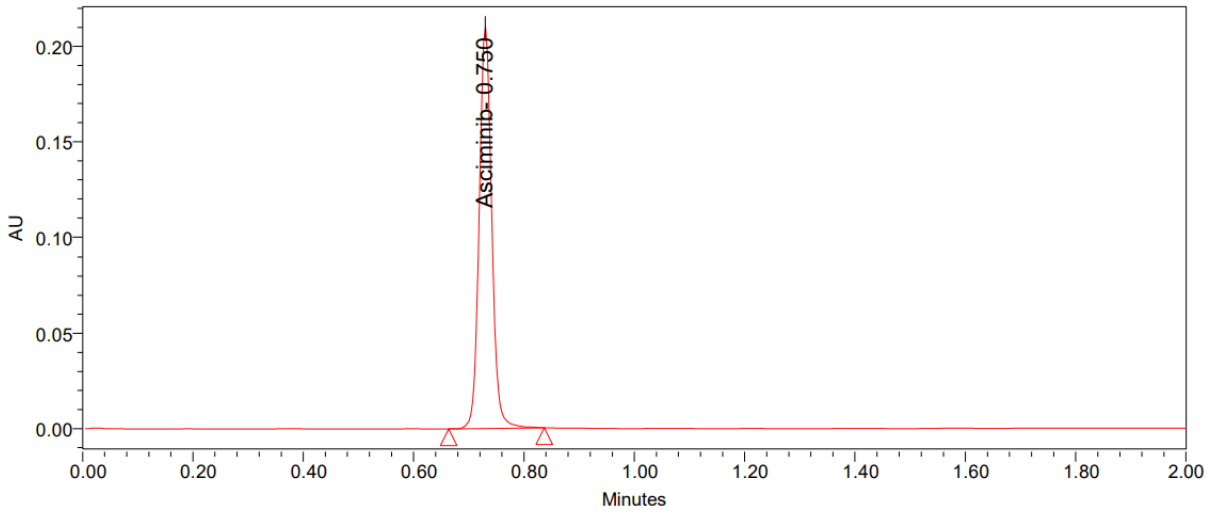

Figure 10: Photo degradation chromatogram.

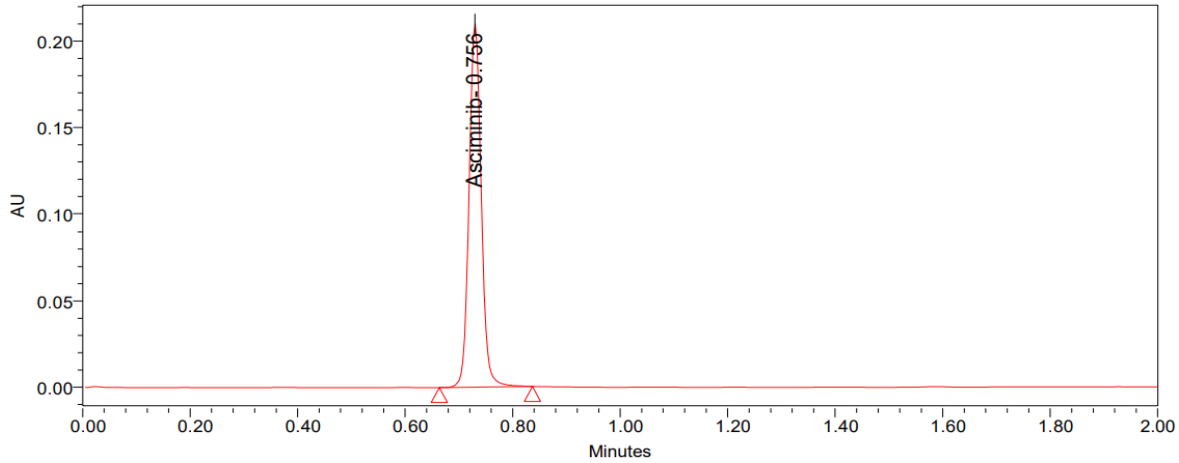

Figure 11: Hydrolytic degradation chromatogram.

3.3. Assay

The suggested method's application to the analysis of formulations is its key feature. Hence the market sample of asciminb was collected and analysed by employing the proposed method. According to the label claim, the drug content obtained from the values of sample solutions was found to be in the permissible range of 90–110 percent. The % assay of asciminib was found to be 40.08 mg/tablet. The study confirmed that the created UPLC method was accurate and easy enough to be used on a daily basis. The suggested assay method's high content results indicate that this technique can be engaged for quantitative regular quality control study of pharmaceutical dosage forms. The results were displayed in Table 15.

Table 15: Results of marketed formulation analysis.

Compound name	Brand name	Label claim (mg)	Assay (mg/tablet)
Asciminib	Scemblix	40	40.08

4. CONCLUSION

For the estimate of asciminib in tablet dose form, a simple, accurate, and specific approach was established. Asciminib had retention time of 0.754 minutes. The percent RSD of method precision and intermediate precision were found to be 0.38 % and 0.74 %. For asciminib, recovery was 100.5 %. The LOD and LOQ values for asciminib calculated from regression equations were 0.12 µg/mL and 0.4 µg/mL consecutively. Regression equation of asciminib was y = 64712.07x + 46933.12. There were some degradation peaks in acid, base, thermal and peroxide stressed conditions, according to the results of the forced degradation test. This drug was recently approved by FDA and there was no analytical method reported. This was the analytical method which has reduced retention time and run time. Hence, the method created was fast, short, simple, and economic, and it might be utilized in frequent quality control tests in industries.

5. CONFLICT OF INTEREST

The authors declare that there is no conflict of competing financial interests.

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7. REFERENCES

1. Patel M. Review Article: Chromatography Principle and Applications Mimansha. Int J Pharmacy&Pharmaceutical Res [Internet]. 2018;13(4):288–93. Available from: <URL>.

2. Priyadarshini R, Raj GM, Gopal Shewade D. Chromatography – The Essence of Bioanalysis. Eur J Biomed Pharm Sci [Internet]. 2016;3(1):366–77. Available from: <URL>.

3. Khan H, Ali J. UHPLC: Applications in Pharmaceutical Analysis. Asian J Pharm Anal [Internet]. 2017;7(2):124–31. Available from: <URL>.

4. Sri RS, Sri KB, Mounika C. A Review on Comparative study of HPLC and UPLC. Res J Pharm Technol [Internet]. 2020;13(3):1570–4. Available from: <URL>.

5. Srivastava B, Sharma BK, Baghel US, Yashwant, Sethi NK. Ultra Performance Liquid Chromatography (UPLC) : A Chromatography Technique. Int J Pharm Qual Assur [Internet]. 2010;2:19–25. Available from: <URL>.

6. Chandarana C, Manani L, Patel V, Prajapati P. UPLC: A Prominent Analytical Technique For Pharmaceuticals. Int J Pharm Sci Rev Res [Internet]. 2016;37(2):192–201. Available from: <URL>.

7. Roge AB, Firke SN, Dhane RM, Gunjkar VJ, Vadvalkar SM. Novel Achievement of HPLC: UPLC. Int J PharmTech Res [Internet]. 2011;3(3):1423–9. Available from: <URL>.

8. Gumustas M, Kurbanoglu S, Uslu B, Ozkan SA. UPLC versus HPLC on Drug Analysis: Advantageous, Applications and Their Validation Parameters. Chromatographia [Internet]. 2013 Nov 27;76(21–22):1365–427. Available from: <URL>.

9. Swartz ME, Murphy B. New Frontiers in Chromatography. Am Lab [Internet]. 2005 [cited 2023 May 13];22–7. Available from: <URL>.

10. Breccia M, Colafigli G, Scalzulli E, Martelli M. Asciminib: An investigational agent for the treatment of chronic myeloid leukemia. Expert Opin Investig Drugs [Internet]. 2021 Aug 3;30(8):803–11. Available from: <URL>.

11. Luna A, Pérez-Lamas L, Boque C, Giraldo P, Xicoy B, Ruiz Nuño C, et al. Real-life analysis on safety and efficacy of asciminib for ponatinib pretreated patients with chronic myeloid leukemia. Ann Hematol [Internet]. 2022 Oct 23;101(10):2263–70. Available from: <URL>.

12. Cortes JE, Hochhaus A, Takahashi N, Larson RA, Issa GC, Bombaci F, et al. Asciminib monotherapy for newly diagnosed chronic myeloid leukemia in chronic phase:Tthe ASC4FIRST phase III trial. Futur Oncol [Internet]. 2022 Dec;18(38):4161–70. Available from: <URL>.

13. Hubbard WK. International Conference on Harmonisation; Guideline on the Validation of Analytical Procedures: Methodology; Availability. Fed Regist [Internet]. 1997;62(96):27464–7. Available from: <URL>.