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A NEW ANALYTICAL RP-HPLC METHOD DEVELOPMENT AND VALIDATION FOR THE QUANTITATIVE ESTIMATION OF DACOMITINIB IN API FORM AND MARKETED PHARMACEUTICAL DOSAGE FORM

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ABSTRACT

A new, simple, rapid, precise, accurate and reproducible RP-HPLC method for estimation of Dacomitinib in bulk form and marketed formulation. Separation of Dacomitinib was successfully achieved on a Symmetry ODS C18 (4.6 x 250mm, 5µm) column in an isocratic mode of separation utilizing Acetonitrile: Methanol in the ratio of 80:20% v/v at a flow rate of 1.0 mL/min and the detection was carried out at 272nm. The method was validated according to ICH guidelines for linearity, sensitivity, accuracy, precision, specificity and robustness. The response was found to be linear in the drug concentration range of 10-50mcg/mL for Dacomitinib. The correlation coefficient was found to be 0.999 for Dacomitinib. The LOD and LOQ for Dacomitinib were found to be 1.1µg/mL and 3.2µg/mL respectively. The proposed method was found to be good percentage recovery for Dacomitinib, which indicates that the proposed method is highly accurate. The specificity of the method shows good correlation between retention times of standard solution with the sample solution. Therefore, the proposed method specifically determines the analyte in the sample without interference from excipients of pharmaceutical dosage forms.

KEYWORDS: Dacomitinib, RP-HPLC, Accuracy, Robustness, Method, Validation, ICH Guidelines.

INTRODUCTION

Dacomitinib, designed as (2E)-N-16-4-(piperidin-1-yl) but-2-enamide, is an oral highly selective Quinazalone part of the second-generation tyrosine kinase inhibitors which are characterized by the irreversible binding at the ATP domain of the epidermal growth factor receptor family kinase domains.[1] Dacomitinib was developed by Pfizer Inc and approved by the FDA on September 27, 2018. Some evidence in the literature suggests the therapeutic potential of Dacomitinib in the epithelial ovarian cancer model, although further investigations are needed. Preclinical data suggested that Dacomitinib increases the inhibition of the epidermal growth factor receptor kinase domain as well as the activity in cell lines harboring resistance mutations such as T790M. This activity further produced a significant reduction of EGFR phosphorylation and cell viability. In these studies, non-small cell lymphoma cancer cell lines with L858R/T790M mutations where used and an IC50 of about 280 nmol/L was observed.[2] Dacomitinib is an irreversible small molecule inhibitor of the activity of the human epidermal growth factor receptor (EGFR) family (EGFR/HER1, HER2, and HER4) tyrosine kinases. It achieves irreversible inhibition via covalent bonding to the cysteine residues in the catalytic domains of the HER receptors.[3] The affinity of Dacomitinib has been shown to have an IC50 of 6nmol/L. The ErbB or epidermal growth factor (EGF) family plays a role in tumor growth, metastasis, and treatment resistance by activating downstream signal transduction pathways such as such as Ras-Raf-MAPK, PLCgamma-PKC-NFkB and PI3K/AKT through the tyrosine kinase-driven phosphorylation at the carboxy-terminus. Around 40% of cases show amplification of EGFR gene and 50% of the cases present the EGFRvIII mutation which represents a deletion that produces a continuous activation of the tyrosine kinase domain of the receptor. The IUPAC name of Dacomitinib is (E)-N-[4-(3-chloro-4-

fluoroanilino)-7-methoxyquinazolin-6-yl]-4-piperidin-1-ylbut-2-enamide. The Chemical Structure of Dacomitinib is shown in follows

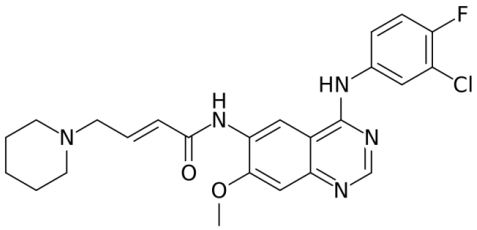

Fig-: Chemical Structure of Dacomitinib.

MATERIALS AND METHODS

Table 7: Instruments Used.

S.No.	Instruments and Glass wares	Model
1	HPLC	WATERS Alliance 2695 separation module, Software: Empower 2, 996 PDA Detector.
2	pH meter	Labindia
3	Weighing machine	Sartorius
4	Volumetric flasks	Borosil
5	Pipettes and Burettes	Borosil
6	Beakers	Borosil
7	Digital ultra sonicator	Labman

Table 8: Chemicals Used.

S.No.	Chemical	Brand Names
1	Dacomitinib (Pure)	Local Market
2	Water and Methanol for HPLC	LICHROSOLV (MERCK)
3	Acetonitrile for HPLC	Merck

HPLC METHOD DEVELOPMENT

Preparation of Standard Solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks[4] add about 7ml of Methanol and sonicate to dissolve and removal of air completely and make volume up to the mark with the same Methanol.

Further pipette 0.3ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Preparation of Sample Solution

Take average weight of the Powder and weight 10 mg equivalent weight of Dacomitinib sample into a 10mL clean dry volumetric flask and add about 7mL of Diluent and sonicate to dissolve it completely and make volume up to the mark with the same solvent.[5]

Further pipette 0.3ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Procedure

Inject the samples by changing the chromatographic conditions and record the chromatograms, note the conditions of proper peak elution for performing validation parameters as per ICH guidelines.[32,33]

Mobile Phase Optimization

Initially the mobile phase tried was methanol: Water and ACN: Water with varying proportions. Finally, the mobile phase[6] was optimized to ACN: Methanol 80:20% v/v) respectively.

Optimization of Column

The method was performed with various C18 columns like Symmetry, Zodiac and Xterra. Symmetry ODS C18 (4.6 x 250mm, 5µm) Column was found to be ideal as it gave good peak shape and resolution at 1ml/min flow.

Preparation of Mobile Phase

Accurately measured 800 ml (80%) of HPLC Acetonitrile and 200 ml of Methanol (20%) were mixed and degassed in a digital ultra sonicater for 15 minutes and then filtered through 0.45 µ filter under vacuum filtration.[7]

Diluent Preparation

The Mobile phase was used as the diluent.

Method Validation Parameters

System Suitability

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Further pipette 0.3ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Procedure

The standard solution was injected for five times and measured the area for all five injections in HPLC. The %RSD for the area of five replicate injections[8] was found to be within the specified limits.

Specificity

Preparation of Standard Solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Further pipette 0.3ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Preparation of Sample Solution

Take average weight of the Powder and weight 10 mg equivalent weight of Dacomitinib sample into a 10mL clean dry volumetric flask and add about 7mL of Diluent and sonicate to dissolve it completely and make volume up to the mark with the same solvent.

Further pipette 0.3ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Procedure

Inject the five replicate injections of standard and inject the three replicate injections sample solutions and calculate the assay[9-12] by using formula:

$$\%ASSAY = \frac{\text{Sample area}}{\text{Standard area}} \times \frac{\text{Weight of standard}}{\text{Dilution of standard}} \times \frac{\text{Dilution of sample}}{\text{Weight of sample}} \times \frac{\text{Purity}}{100} \times \frac{\text{Weight of tablet}}{\text{Label claim}} \times 100$$

Linearity

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Preparation of Level – I (10ppm of Dacomitinib)

Take 0.1ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – II (20ppm of Dacomitinib)

Take 0.2ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – III (30ppm of Dacomitinib)

Take 0.3ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – IV (40ppm of Dacomitinib)

Take 0.4ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Preparation of Level – V (50ppm of Dacomitinib)

Take 0.5ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Procedure

Inject each level into the chromatographic system[13] and measure the peak area.

Plot a graph of peak area versus concentration (on X-axis concentration and on Y-axis Peak area) and calculate the correlation coefficient.

Precision

Repeatability

Preparation of Dacomitinib Product Solution for Precision

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7ml of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.3ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

The standard solution was injected for six times and measured the area for all six injections in HPLC. The %RSD for the area of six replicate injections was found to be within the specified limits.[14]

Intermediate Precision

To evaluate the intermediate precision[15] (also known as Ruggedness) of the method, Precision was performed on different days by maintaining same conditions.

Procedure

Analyst 1

The standard solution was injected for six times and measured the area for all six injections in HPLC. The %RSD for the area of six replicate injections was found to be within the specified limits.

Analyst 2

The standard solution was injected for six times and measured the area for all six injections in HPLC. The

%RSD for the area of six replicate injections was found to be within the specified limits.

Accuracy

For preparation of 50% Standard stock solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.15ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

For preparation of 100% Standard stock solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents[16] and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.3ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

For preparation of 150% Standard stock solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.45ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.

Procedure

Inject the Three replicate injections of individual concentrations (50%, 100%, 150%) were made under the optimized conditions. Recorded the chromatograms and measured the peak responses. Calculate the Amount found and Amount added for Dacomitinib and calculate the individual recovery and mean recovery values.[17]

Limit of Detection (LOD) and Limit of Quantification (LOQ)

Preparation of 0.597µg/ml solution (LOD)

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7ml of Methanol and sonicate to dissolve and removal of air completely and make volume up to the mark with the same Methanol.

Further pipette 0.00597ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Preparation of 1.811µg/ml solution (LOQ)

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7ml of Methanol and sonicate to dissolve and removal of air completely and make volume up to the mark with the same Methanol.

Further pipette 0.01811ml of the above Dacomitinib stock solutions into a 10ml volumetric flask and dilute up to the mark with Methanol.

Robustness

The analysis was performed in different conditions to find the variability of test results.[18] The following conditions are checked for variation of results.

For preparation of Standard solution

Accurately weigh and transfer 10 mg of Dacomitinib working standard into a 10ml of clean dry volumetric flasks add about 7mL of Diluents and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution)

Take 0.3ml of stock solution in to 10ml of volumetric flask and make up the volume up to mark with diluent.[19]

Effect of Variation of Flow Conditions

The sample was analyzed at 0.9ml/min and 1.1ml/min instead of 1ml/min, remaining conditions are same. 20µl of the above sample was injected and chromatograms were recorded.

Effect of Variation of Mobile Phase Organic Composition

The sample was analyzed by variation of mobile phase i.e. ACN: Methanol was taken in the ratio and 75:25, 85:15 instead of 80:20, remaining conditions are same. 20µl of the above sample was injected and chromatograms were recorded.

RESULTS AND DISCUSSION

Analytical Method Development

Optimization of Analytical Method

Optimized Chromatographic Conditions

Column	:	Symmetry ODS C18 (4.6 x 250mm, 5µm)
Column temperature	:	Ambient
Wavelength	:	272 nm
Mobile phase ratio	:	ACN: Methanol (80:20% v/v)
Flow rate	:	1.0mL/min
Injection volume	:	20 µl
Run time	:	8 minutes

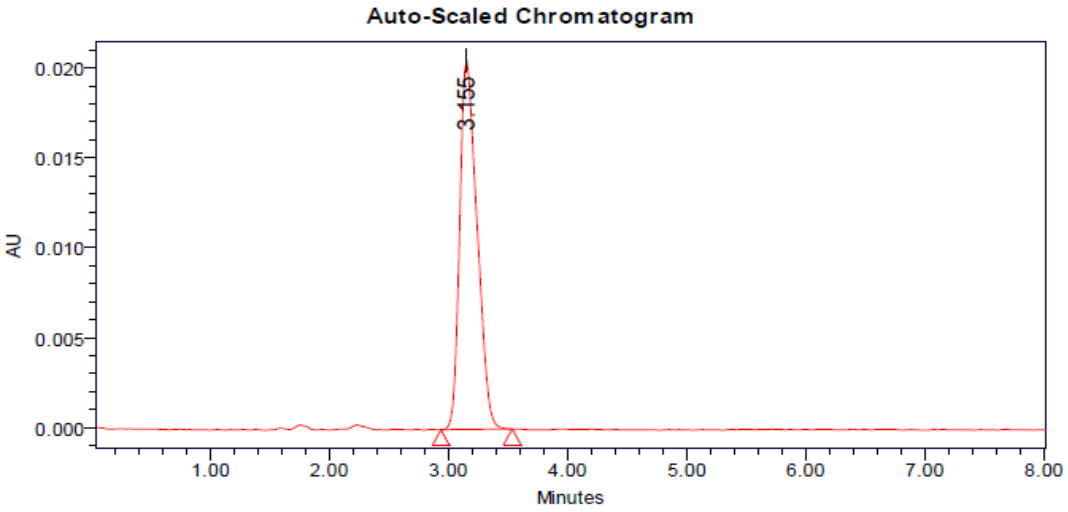

Fig-9: Optimized Chromatographic Condition.

Validation of Method

The developed method was validated for linearity and range, accuracy, precision, Limit of detection, Limit of quantitation and robustness as per ICH guidelines.[32,33]

System Suitability Parameters: To evaluate system suitability parameters[20-22] such as theoretical plates, tailing factor and retention time of five replicate injections of standard Dacomitinib of concentration 30µg/ml was used and the % RSD values were calculated.

Table 12: Results of system suitability for Dacomitinib.

S.No.	Peak Name	RT	Area (µV*sec)	Height (µV)	USP Plate Count	USP Tailing
1	Dacomitinib	3.192	225645	20584	6286	1.38
2	Dacomitinib	3.146	225847	20965	6358	1.39
3	Dacomitinib	3.123	228656	20758	6285	1.41
4	Dacomitinib	3.167	228547	20859	6278	1.40
5	Dacomitinib	3.158	229658	20968	6395	1.42
Mean			227670.6			
Std. Dev.			1810.899			
% RSD			0.795403			

Specificity

The ICH documents define specificity as the ability to assess unequivocally the analyte in the presence of components that may be expected to be present, such as impurities, degradation products, and matrix components.

Analytical method was tested for specificity[23] to measure accurately quantitates Dacomitinib in drug product.

Table 17: Results of Assay (Standard) for Dacomitinib.

S.No.	Peak Name	RT	Area (µV*sec)	Height (µV)	USP Plate Count	USP Tailing
1	Dacomitinib	3.146	220136	20568	6125	1.36
2	Dacomitinib	3.123	220187	20653	6132	1.38
3	Dacomitinib	3.192	220175	20548	6129	1.34
4	Dacomitinib	3.164	220196	20698	6187	1.35
5	Dacomitinib	3.181	220134	20548	6159	1.35
Mean			220165.6			
Std. Dev.			28.91885			
% RSD			0.013135			

$$\%ASSAY = \frac{\text{Sample area}}{\text{Standard area}} \times \frac{\text{Weight of standard}}{\text{Dilution of standard}} \times \frac{\text{Dilution of sample}}{\text{Weight of sample}} \times \frac{\text{Purity}}{100} \times \frac{\text{Weight of tablet}}{\text{Label claim}} \times 100$$

= 99.24%

The % purity of Dacomitinib in pharmaceutical dosage form was found to be 99.24%.

Linearity

The linearity was analyzed through the standard curves ranging from 10.0µg/ml to 50.0µg/ml. The linearity[24] was evaluated by linear regression analysis, which was calculated by the least-square regression analysis.

Table 19: Data for Linearity.

Concentration µg/ml	Average Peak Area
10	78683
20	146545
30	213584
40	279895
50	346568

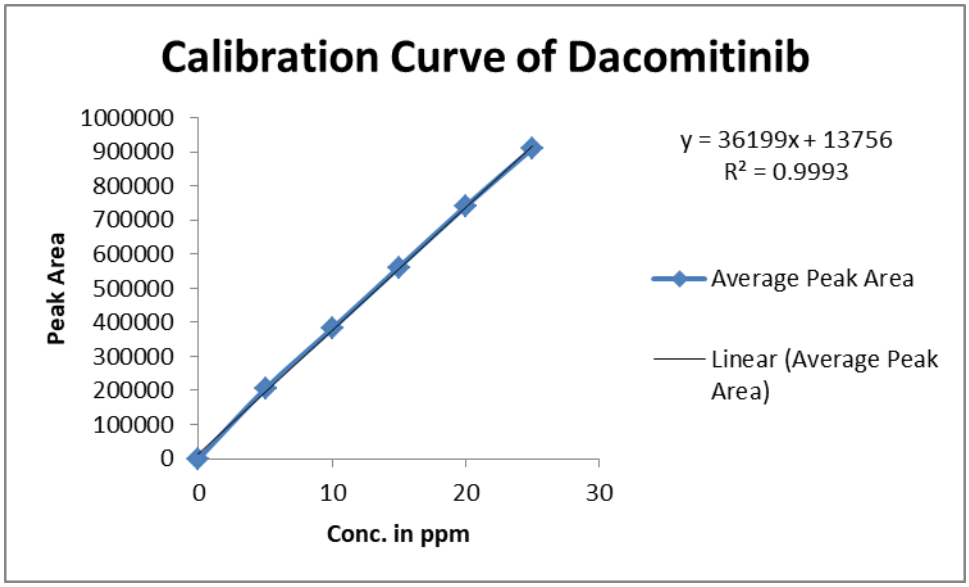

Fig-25: Calibration Curve of Dacomitinib.

Linearity Plot

The plot of Concentration (x) versus the Average Peak Area (y) data of Dacomitinib is a straight line.

Y = mx + c

Slope (m) = 6867.2

Intercept (c) = 5866.2

Correlation Coefficient (r) = 0.99

Validation Criteria: The response linearity is verified if the Correlation Coefficient is 0.99 or greater.

Conclusion: Correlation Coefficient (r) is 0.99, and the intercept is 5866. These values meet the validation criteria.

Precision

The precision[25] of an analytical procedure expresses the closeness of agreement (degree of scatter) between a series of measurements obtained from multiple sampling of the same homogeneous sample under the prescribed conditions.

Repeatability

Obtained Six (6) replicates of 100% accuracy solution as per experimental conditions. Recorded the peak areas and calculated % RSD.

Table 20: Results of Method Precision for Dacomitinib.

S. No.	Peak name	Retention time	Area (µV*sec)	Height (µV)	USP Plate Count	USP Tailing
1	Dacomitinib	3.165	225645	20562	6125	1.36
2	Dacomitinib	3.163	225847	20645	6129	1.36
3	Dacomitinib	3.158	226542	20534	6135	1.35
4	Dacomitinib	3.167	226598	20564	6189	1.36

5	Dacomitinib	3.171	226584	20549	6138	1.35
6	Dacomitinib	3.181	226859	20685	6179	1.37
Mean			226345.8			
Std. Dev			482.1068			
%RSD			0.212996			

Intermediate Precision

Analyst 1

Table 21: Results of Ruggedness for Dacomitinib.

S.No	Peak Name	RT	Area (µV*sec)	Height (µV)	USP Plate Count	USP Tailing
1	Dacomitinib	3.165	226534	20653	6235	1.35
2	Dacomitinib	3.163	226542	20598	6198	1.36
3	Dacomitinib	30158	225989	20653	6254	1.36
4	Dacomitinib	3.167	226512	20548	6281	1.35
5	Dacomitinib	3.171	226531	20653	6199	1.36
6	Dacomitinib	3.171	225898	20658	6253	1.35
Mean			226334.3			
Std. Dev.			304.2622			
% RSD			0.13443			

Analyst 2

Table 22: Results of Intermediate Precision Analyst 2 for Dacomitinib.

S.No.	Peak Name	RT	Area (µV*sec)	Height (µV)	USP Plate count	USP Tailing
1	Dacomitinib	3.173	225487	20542	6253	1.35
2	Dacomitinib	3.134	225484	20532	6098	1.36
3	Dacomitinib	3.161	225364	20541	6254	1.35
4	Dacomitinib	3.174	226513	20534	6235	1.36
5	Dacomitinib	3.199	225487	20549	6199	1.36
6	Dacomitinib	3.199	226532	20451	6235	1.35
Mean			225811.2			
Std. Dev.			553.0524			
% RSD			0.244918			

Accuracy

Accuracy[26] at different concentrations (50%, 100%, and 150%) was prepared and the % recovery was calculated.

Table 26: The Accuracy Results for Dacomitinib.

%Concentration (at specification Level)	Area	Amount Added (ppm)	Amount Found (ppm)	% Recovery	Mean Recovery
50%	109283.3	15	15.060	100.40%	100.42%
100%	212732	30	30.124	100.413%	
150%	316263.3	45	45.201	100.446%	

Limit of Detection for Dacomitinib

The detection limit[27] of an individual analytical procedure is the lowest amount of analyte in a sample which can be detected but not necessarily quantitated as an exact value.

LOD= 3.3 × σ / s

Where

σ = Standard deviation of the response

S = Slope of the calibration curve

Result

= 0.597µg/ml

Limit of Quantitation for Dacomitinib

The quantitation limit[28] of an individual analytical procedure is the lowest amount of analyte in a sample which can be quantitatively determined.

LOQ=10×σ/S

Where

σ = Standard deviation of the response

S = Slope of the calibration curve

Result

= 1.811µg/ml

Table-: Results of LOD & LOQ.

SE of Intercept	556.1832432
SD of Intercept	1243.66354
LOD	0.597636545
LOQ	1.811019833

SE of Intercept = Excel Function (Data Analysis → Regression)
SD of Intercept = SE of Intercept * √N
LOD = 3.3 * (SD of Intercept/Slope)
LOQ = 10 * (SD of Intercept/Slope)

Robustness

The robustness[29] was performed for the flow rate variations from 0.9 ml/min to 1.1ml/min and mobile phase ratio variation from more organic phase to less organic phase ratio for Dacomitinib. The method is robust only in less flow condition and the method is robust even by change in the Mobile phase ±5%. The standard and samples of Dacomitinib were injected by changing the conditions of chromatography. There was no significant change in the parameters like resolution, tailing factor, asymmetric factor, and plate count.

Table 27: Results for Robustness of Dacomitinib.

Parameter used for sample analysis	Peak Area	Retention Time	Theoretical plates	Tailing factor
Actual Flow rate of 1.0 mL/min	225645	3.155	6125	1.36
Less Flow rate of 0.9 mL/min	236586	3.488	6452	1.38
More Flow rate of 1.1 mL/min	219865	2.877	6098	1.42
Less organic phase	235848	4.705	6126	1.43
More organic phase	241245	2.090	6324	1.39

Forced Degradation Studies

The specificity of the method can be demonstrated by applying stress conditions[30-31] using acid, alkaline, peroxide, thermal, UV, water degradations. The sample was exposed to these conditions the main peak of the drug was studied for peak purity that indicating the method effectively separated the degradation products from the pure active ingredient.

Table 34: Results of Forced Degradation Studies for Dacomitinib.

S.No.	Stress Condition	Peak Area	% of Degraded Amount	% of Active Amount	Total % of Amount
1	Standard	225645	0	100%	100%
2	Acidic	190015.65	15.79	84.21	100%
3	Basic	187353.04	16.97	83.03	100%
4	Oxidative	190985.92	15.36	84.64	100%
5	Thermal	183020.65	18.89	81.11	100%
6	Photolytic	181034.98	19.77	80.23	100%
7	Water	210549.34	6.69	93.31	100%

SUMMARY AND CONCLUSION

The analytical method was developed by studying different parameters. First of all, maximum absorbance was found to be at 272nm and the peak purity was excellent. Injection volume was selected to be 20µl which gave a good peak area. The column used for study was Symmetry ODS C18 (4.6 x 250mm, 5µm) because it was giving good peak. Ambient temperature was found to be suitable for the nature of drug solution. The flow rate was fixed at 1.0ml/min because of good peak area and satisfactory retention time. Mobile phase is Acetonitrile: Methanol (80:20% v/v) was fixed due to good symmetrical peak. So this mobile phase was used for the proposed study. Run time was selected to be 8.0min because analyze gave peak around 3.155 and also to reduce the total run time. The percent recovery was found to be 98.0-102 was linear and precise over the same range. Both system and method precision were found to be accurate and well within range. The analytical method was found linearity over the range of 10-50µg/ml of the Dacomitinib target concentration. The analytical passed both robustness and ruggedness tests. On both cases, relative standard deviation was well satisfactory.

REFERENCES

1. https://go.drugbank.com/drugs/DB11963
2. https://pubchem.ncbi.nlm.nih.gov/compound/Dacomitinib
3. https://en.wikipedia.org/wiki/Dacomitinib
4. Sharma BK. Instrumental methods of chemical analysis, Introduction to analytical chemistry, 23th Ed. Goel publishing house Meerut, 2004, P12-23.
5. H.H. Willard, L.L. Merritt, J.A. Dean, F.A. Settle. Instrumental methods of analysis, 7th edition, CBS publishers and distributors, New Delhi, 1986; P.518-521, 580-610.
6. John Adamovies, Chromatographic analysis of pharmaceutical, Marcel Dekker Inc. New York, 2nd Ed, P.74, 5-15.
7. Gurdeep Chatwal, Sahm K. Anand. Instrumental methods of chemical analysis, 5th edition, Himalaya publishing house, New Delhi, 2002; P.1.1-1.8, 2.566-2.570.

8. D. A. Skoog. J. Holler, T.A. Nieman. Principle of instrumental analysis, 5th edition, Saunders college publishing, 1998; P.778-787.
9. Skoog, Holler, Nieman. Principals of instrumental a nalysis 5th Ed, Harcourt Publisher's international company, 2001; P.543-554.
10. William Kemp. Organic spectroscopy, Palgrave, New York, 2005; P.7-10, 328-330.
11. P.D. Sethi. HPLC: Quantitative analysis pharmaceutical formulations, CBS publishers and distributors, New Delhi (India), 2001; P.3-137.
12. Michael E, Schartz IS, Krull. Analytical method development and validation, 2004; P. 25-46.
13. R. Snyder, J. Kirkland, L. Glajch. Practical HPLC method development, 2nd Ed, a Wiley international publication, 1997, P.235, 266-268, 351-353.653-600.686-695.
14. Basic education in analytical chemistry. Analytical science, 2001: 17(1).
15. Method validation guidelines international conference on harmonization; GENEVA, 1996.
16. Berry RI, Nash AR. Pharmaceutical process validation, Analytical method validation, Marcel Dekker Inc. New work, 1993; 57: 411-28.
17. Anthony C Moffat, M David Osselton, Brian Widdop. Clarke's analysis of drugs and poisons, Pharmaceutical press, London, 2004; P.1109-1110, 1601-1602.
18. Klaus Florey, Analysis profile of drugs substances, Academic press, New York, 2005; P.406-435.
19. P.N. Arora, P.K. Malhan. Biostatistics, Himalaya Publishers house, India, P.113, 139-140,154.
20. L. R. Snyder, J.J. Kirkland, and J. W. Dolan, Introduction to Modern Liquid Chromatography, John Wiley & Sons, New York, 2009.
21. M.W. Dong, Modern HPLC for practicing scientists. Wiley, 2006.
22. L. R. Snyder, J.J. Kirkland, and J. L. Glajch, Practical HPLC Method Development, John Wiley & Sons, New York, 1997.
23. S. Ahuja and H. T. Rasmussen (Ed), HPLC Method Development for Pharmaceuticals, Academic Press, 2007.
24. S. Ahuja and M.W. Dong (Ed), Handbook of Pharmaceutical Analysis by HPLC, Elsevier/Academic Press, 2005.
25. Y. V. Kazakevich and R. LoBrutto (ed.), HPLC for Pharmaceutical Scientists, Wiley, 2007.
26. U. D. Neue, HPLC Columns: Theory, Technology, and Practice, Wiley-VCH, New York, 1997.
27. M. C. McMaster, HPLC, a practical user's guide, Wiley, 2007.
28. Doserge, Wilson and Gisvold's text book of organic medicinal and pharmaceutical chemistry, 8th Ed, Lippincott Company, 1982, P.183-197.
29. Snyder, Lloyd R.; Dolan, John W. (2006). High-Performance Gradient Elution: The Practical Application of the Linear-Solvent-Strength Model. Wiley Interscience.
30. Giddings, J. Calvin (1965) Dynamics of Chromatography, Part I. Principles and Theory. Marcel Dekker, Inc., New York. p. 281.
31. K., Robards (1994). Principles and practice of modern chromatographic methods. Haddad, P. R., Jackson, P. E. Amsterdam: Elsevier/Academic Press.
32. ICH Guidance. Validation of analytical methods - definition and terminology. Q2A. Geneva: International Conference on Harmonization. Nov 2005.
33. ICH Guidance, Validation of analytical procedures - methodology. Q2B. Geneva: International Conference on Harmonization. Nov 2005.
34. Kirti Kumari*, Pankaj Thakur, Sayali Warde, Vijay Munipalli, Raman Mohan Singh, HPLC Method Development and Validation for Quantitative Estimation of Dacomitinib in Pharmaceuticals Dosage Form, International Journal of Pharmacy and Pharmaceutical Research, (Ijppr. Human), 2021; 22(3): 606-620.
35. Venu Kamani, M. Sujatha, G. S. N. Koteswara Rao *, N. Vinod Kumar, Stability Indicating RP-HPLC Method for the Analysis of Dacomitinib and its Related Impurities in Bulk and Oral Solid Dosage Formulations, Journal of Pharmaceutical Research International, Issue: 2021 - Volume 33 [Issue 40B], Pages: 187-199, DOI: 10.9734/jpri/2021/v33i40B32278.
36. Mahesh.M1*, N.Devanna2, An Advanced Biological Matrix Extraction method development and Validation for Dacomitinib in Human Plasma (K2 EDTA plasma) by using LC-MS/MS, Eur. Chem. Bull, 2023; 12(5): 2406-2416.