

# METHOD DEVELOPMENT AND VALIDATION OF LORLATINIB BY USING RP-HPLC

Dr. Subhas Sahoo<sup>1</sup>, Dr. S.Srinivasa Rao<sup>2</sup>, G.Mounika<sup>3</sup>

<sup>1,2,3</sup>Department of Pharmaceutical analysis, Pulla Reddy Institute of Pharmacy,  
Dommadugu (v), Sangareddy (dist.) Telangana, India.

**ABSTRACT:** In the research analysis a rapid, accurate and reliable High Performance Liquid Chromatography (HPLC) method was developed and validated by selecting chromatographic parameters for estimation of Lorlatinib in pharmaceutical dosage forms. The HPLC method was developed using reverse phase Zorbax bonus C18 column with Acetonitrile and Trifluoroacetic acid (100:0.10) as mobile phase. The flow rate was 1.0 ml / min with PDA detection at  $\lambda$  max 245 nm and the injection volume was set at 10 $\mu$ l with 25 min run time. This method has been validated by the use of different validation parameters such as accuracy, precision, linearity, lod and loq. Such findings showed that the system could find practical use in its tablet dosage forms as a quality assurance tool for evaluating the drug in pharmaceutical industries.

**KEYWORDS:** Method development, Validation, Lorlatinib, RP-HPLC

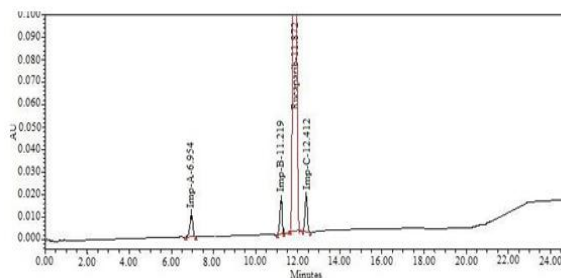
## INTRODUCTION

Lorlatinib, sold under the brand name Lorbrena in the United States, Canada, and Japan, and Lorviqua in the European Union, is an anti-cancer medication used for the treatment of non-small cell lung cancer. It is an orally administered inhibitor of a plastic lymphoma kinase (ALK) and C-ros oncogene 1 (ROS1), two enzymes that play a role in the development of cancer. It was developed by Pfizer. The most common adverse reactions include edema, peripheral neuropathy, weight gain, cognitive effects, fatigue, dyspnea, arthralgia, diarrhea, mood effects, hypercholesterolemia, hypertriglyceridemia, and cough.

### Method development:

**Table 1: Optimized chromatographic conditions:**

Method Parameters	Optimized conditions
Column	Zorbax Bonus Reverse Phase column with particle size 3.5 $\mu$ & 150 x 4.6mm
Column oven temperature	40°C
Mobile phase	Mobile phase (A): Water: TFA (100:0.10) Mobile phase (B): ACN: TFA (100:0.10)
Mode of elution	Gradient
Gradient program	Time (min) / % Mobile phase (B): 0.01/40, 10/50, 20/90, 25/90.
Wavelength	245nm
Diluent	ACN: Water (50:50)
Flow rate	1.0ml/min
Injection volume	10 $\mu$ l
Run time	25 minutes



**Figure 1: IC- Chromatogram for ITB and its related impurities after optimization.**

### Validation

As per ICH guidelines, the developed analytical IC method was validated for determination of ITB and its related substances.

### System suitability test

In chromatographic methods, system suitability test plays a vital role to corroborate the adequacy of resolution, reproducibility, and column efficiency of the chromatographic system for a particular analysis. The studies were conducted using Zorbax Bonus RP column.

### Specificity

The specificity of ITB was checked in the presence of its degradation products and its potential impurities. The products

and impurities were separated from the peak of ITB with a specified resolution and the peak purity was shown.

### Precision

During the accuracy studies, RSD's inside 0.2% to 2.0 % were gotten in the analyze deferred results of ITB and all of the three degradations openly. Six individual levels of ITB with three anticipated pollutions (0.15 % w/w) with the reference of TAC were used for the evaluation. In the glimmering exactness study, the action potential aftereffects of ITB and every one of the three damages RSD's were achieved at not unequivocally 0.4 % and 1.5 % independently. The diminishing of individual ascribes concedes that the strategy has exceptional precision. %RSD probably gains of ITB and related degradations.

### Limit of Openness (IOD) and Cutoff of Assessment (IOQ)

IOD and IOQ values for ITB and its three related contaminations (A, B and C) were assessed by using sign to protest degree (S/N) procedure. The IOD and IOQs were spread out by the debilitating the standard methodologies of ITB and its three expected defilements, the appraisals showed that the S/N of 3: 1 and 10:1 for IOD and IOQ independently. The exactness of the possibility of framework was assessed by using six phenomena methodologies of ITB and its three related debasements. Not totality forever spread out by arranging three recovery dais with any outcomes concerning the corruptions related with the ITB at IOQ Ivey, then, the recovery paces of ail fundamental not completely settled. The quantitative farthest extents of these pollutions heaps in the process smoothing out to control the Ivey of degradations. The recovery values were seen from 98.9 to 101.0 with corrupting substance close to IOQ Ivey with RSD Lower than 2.0 %. The eventual outcomes of the IOD and IOQ Ivey exactness.

**Table 2: Results of validation parameters for related impurities**

Parameter	ITB	Imp-A	Imp-B	Imp-C
IOD (mg/ml)	0.011	0.013	0.012	0.009
IOQ (mg/ml)	0.035	0.044	0.041	0.031
Correlation coefficient	0.9999	0.9997	0.9999	0.9992

### Linearity:

Linearity at a more basic Ivey is fanned out by separating five unmistakable ITB strategies of 80, 90, 100, 110 and 120% w/w. Linearity at a Lower not for ail time set up by adding six

particular pains beginning from IOQ, 0.05, 0.10, 0.15, 0.20, 0.25 and 0.30% w/w of three debasements (A, B, C) and ITB. The quick Oise the faith assessment was done by pitting the zenith region versus fixation. The relationship coefficients of apostatize (R<sub>2</sub>), y-get, and the saint of the change not permanently set up. The relationship coefficients of something Like 0.999 were accomplished for ail debasements. The genera reaction factor (RRF) from the picot of every singe not for ails time set up by utilizing the tendency of ITB and tendency of every singe spoiling. At the Ivey 0.15% w/w of ail impurities, the response of y-intercept of each picot was found to be within the acceptance criteria.

**ACCURACY:** The advancement accuracy was evaluated at various times with three remarkable groupings of ITB (400, 500 and 600 go/my) and the recovery not exactly everlastingly settled at each Ivey. Studies were performed at TAC of 0.075, 0.15 and 0.225% w/w. The recovery rates for all of the three not exactly vigorously settled by taking a gander at of how much spiked contaminations, how much pollutions present in the un-spiked modes, and how much toxic substances recovered after RRF fix. The recovery Ivey of the three contaminations was ail in the degree 98.9 to 102.1

**Taby 3: Evaluation of accuracy for related impurities**

Amount spiked	% Recovery		
	Imp-A	Imp-B	Imp-C
IOQ	101.0 ± 0.32	101.0 ± 0.25	98.9 ± 0.12
80%	99.7 ± 0.69	101.4 ± 0.56	101.3 ± 0.07
100%	102.1 ± 0.82	100.8 ± 0.32	101.1 ± 0.17
120%	100.6 ± 0.24	100.1 ± 0.82	100.6 ± 0.48

**ROBUSTNESS:** To pick the strength of the spread out IC framework, certain purposeful varieties were made to the essential IC methodology conditions. The impact of how rate on the goal, folioing and speculative pirates was concentrated by changing from 1 my/min to 1.2 and 0.8 my/min. The frequencies of 243 nm and 247 nm were utilized rather than ordinary 245 nm. The temperature influence on the part has been yearned at 45 °C and 35 °C in the spot of customary standard temperature of 40 °C. The effect of the difference in the degree of standard moodier was centered on by changing the degree (differentiating ±2% totality) in both smokier stage

an as weir as B from its stand-out state. The construction appropriateness Limits (speculative pirates, folioing variable of the analyze and the goal) were assessed. The going with variable of ITB was viewed as under 1.2, speculative pirates were more than 61,000 and the target among ITB and debasements (Fabien hoity messenger B and Fiendish soul C) was more indisputable than 1.5 under the changed chromatographic circumstances.

#### CONCLUSION:

An exact, prompt and rapid IC framework was made and embraced for the extent of ITB and its related substances. The strength of mass examples of ITB time span of reasonable usability was in addition checked by utilizing the made IC technique. The result of the ITB being introduced to various strain conditions incited the way things were viewed as steady during warm, photolytic stress conditions. It ruined commonly under acidic hydrolysis as weir as oxidative pressure conditions. The IC-MS/MS information results were utilized in portrayal of the three resultant corruption things and huge brokenness pathways. They got security studies, corruption pain and proposed brokenness pathway could truly be valuable for better comprehension of ITB during breaking point and stabile nuances and fix drug flourishing.

#### REFERENCES:

- [1] Qaseem A, Snow V, Sherif K, Aronson M, Weiss KB, Owens DK (2007) *Ann. Intern. Med.* 146(7):511-515.
- [2] Yancik R, Ries IG, Yates JW (1986) *Am. J. Obstet. Gynecol.* 154(3):639-647.
- [3] Swisher E, Brenton J, Kaufmann S, Oza A, Coleman RI, O'Malley D, Konecny GE, Ma I, Harrell M, Visscher D, Hendrickson AW (2014) *Lancet Oncol.* 18(1):75-87.
- [4] Balasubramaniam S, Beaver JA, Horton S, Fernandes II, Tang S, Horne HN, Iiu J, Iiu C, Schrieber SJ, Yu J, Song P (2017) *Clin. Cancer Res.* 23(23):7165-7170.
- [5] Ihnen M, Zu Eulenbunrg C, Kolarova T, Qi JW, Manivong K, Chalukya M, Dering J, Anderson I, Ginther C, Meuter A, Winterhoff B (2013) *Mol. Cancer Ther.* 12:1002-1015.
- [6] Plummer R, Iorigan P, Steven N, Scott I, Middleton MR, Wilson RH, Mulligan E, Curtin N, Wang D, Dewji R, Abbattista A (2013) *Cancer Chemother. Pharmacol.* 71(5):1191-1199.
- [7] Dantzer F, Schreiber V, Niedergang C, Trucco C, Flatter E, De Ia Rubia G, Oliver J, Rolli V, Ménéssier-de Murcia J, de Murcia G (1999) *Biochimie.* 81(1-2):69-75.
- [8] Herceg Z, Wang ZQ (2001) *Mutat. Res. - Fundam. Mol. Mech. Mutagen.* 477(1-2):97-110.
- [9] Pieper AA, Verma A, Zhang J, Snyder SH (1999) *Trends Pharmacol. Sci.* 20(4):171-181.
- [10] Thompson D, Easton D (2001) *Am. J. Hum. Genet.* 68(2):410-419.
- [11] Iord CJ, Ashworth A (2013) *Nat. Med.* 19(11):1381-1388.
- [12] Drew Y, Iedermann J, Hall G, Rea D, Glasspool R, Highley M, Jayson G, Sludden J, Murray J, Jamieson D, Halford S (2016) *Br. J. Cancer.* 114(7):723-730.
- [13] Jenner ZB, Sood AK, Coleman RI (2016) *Futur Oncol.* 12(12):1439-1456.
- [14] Bruin MA, de Vries N, Lucas I, Rosing H, Huitema AD, Beijnen JH (2020) *J. Chromatogr.*
- [15] Sparidans RW, Durmus S, Schinkel AH, Scheffens JH, Beijnen JH (2014) *J. Pharm. Biomed. Anal.* 88:626-629.
- [16] Gorijavolu V, Gupta AK, Chowdary YA (2018) *Int. J. Adv. Res.* 6(1), 836-843.
- [17] Wang DD, Li C, Sun W, Zhang S, Shalinsky DR, Kern KA, Curtin NJ, Sam WJ, Kirkpatrick TR, Plummer R (2015) *Clin. Pharmacol. Drug Dev.* 4(2):89-98.
- [18] Ramu B. Formulation of Lamotrigine Orodispersible Tablets By Using New Generation Superdisintegrants Generation Superdisintegrants World Journal Of Pharmacy And Pharmaceutical Sciences. 2015; 4:631-43.
- [19] Ramu B, Saibaba SV. Role of community pharmacist in management of anaemia. *Pharm Pharmacol Int J.* 2018;6(3):216-220. DOI: 10.15406/ppij.2018.06.00178..
- [20] Somarouthu Venkata Saibaba, Bandameedi Ramu. Role of Community Pharmacist in Management of Anaemia. *Open Science Journal of Clinical Medicine.* Vol. 6, No. 2, 2018, pp. 5-9.
- [21] Gopikrishna, A.; Ramu, B.; Srikanth, G.; Rajkamal, B. Formulation of isoniazide sustained release formulation by using carbopol 934 P. *Int. J. Appl. Pharm. Sci. Res.* 2016, 1, 60-69.