LNG-451 (BLU-451) is a potent, CNS-penetrant, wild-type EGFR sparing inhibitor of EGFR exon 20 insertion mutations

Poster Number 3332

Brion W. Murray,[1] Anjali Pandey,[1] Bruce Roth,[1] Tracy Saxton,[1] Daniel J. Estes,[1] Himanshu Agrawal,[2] Santosh Vishwakarma,[2] Gurulingappa Hallur,[2] Ishtiyaque Ahmad,[2] Ravi Trivedi,[2] Helen Jenkins,[1] Paul G. Pearson[1]

[1]Lengo Therapeutics, Cambridge, MA, USA[a]; [2]Jubilant Biosys Limited, Bengaluru, Karnataka, India.
[a]A wholly owned subsidiary of Blueprint Medicines Corporation, Cambridge, MA, USA.

Background

- Epitheilal growth factor receptor (*EGFR*) exon 20 insertions (ex20ins) are oncogenic driver mutations that constitutively upregulate kinase activity, are the third most common type of activating *EGFR* mutation in patients with lung cancer, and are not potently targeted by many inhibitors of common activation mutations such as L858R and exon 19 mutations[1]
- *EGFR* ex20ins are in-frame insertions of one to seven amino acids in the αC helix or following the αC helix,[2] with the three most prevalent insertions V769_D770insASV, D770_N771insSVD, and H773_V774insNPH accounting for half of the cases[1]
- *EGFR* ex20ins alter the EGFR active site in a manner which renders the first three generations of EGFR inhibitors generally ineffective[3]
- While there are approved therapies such as mobocertinib and amivantamab, and others in clinical development, none have demonstrated meaningful central nervous system (CNS) activity, and can be associated with treatment-limiting adverse events, including wild-type (WT) EGFR-mediated toxicities[4,5]
- BLU-451 (formerly known as LNG-451) was designed as a covalent inhibitor to potently inhibit EGFR ex20ins mutations, spare WT EGFR, and be CNS penetrant

Methods

- BLU-451 cellular activity was tested by cell viability assays in both tumor and Ba/F3 engineered cell lines expressing *EGFR* mutations as well as cell lines dependent on WT *EGFR*
- Kinome engagement was assessed using broad panels of ligand binding assays as well as radiometric enzymatic assays
- In an *EGFR* ex20ins-driven cell line-derived xenograft (CDX) tumor model, pharmacokinetics/pharmacodynamics (PK/PD) analysis of BLU-451 and comparators were performed in skin, and large intestine tissue
- The *in vivo* antitumor activity of BLU-451 was assessed in a CDX tumor model using Ba/F3 cells expressing *EGFR* ex20ins, and an *EGFR* ex20ins patient-derived xenograft (PDX) tumor model

Results

- In cell viability assays, BLU-451 had similar potency to mobocertinib against *EGFR* ex20ins and greater potency than osimertinib (**Table 1**)
- In WT EGFR dependent cell viability assays, BLU-451 was less potent than mobocertinib and similar in potency to osimertinib (**Table 1**)
- BLU-451 was potent against uncommon oncogenic point mutations (e.g., G719S, L861Q; **Table 1**)

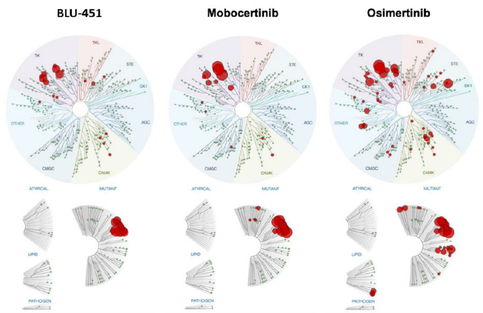
Table 1: BLU-451 spared WT EGFR and inhibited EGFR ex20ins and other oncogenic mutations

Construct	Cell line	BLU-451	Osimertinib	Mobocertinib
IC_{50} values (nM) for EGFR exon 20				
SVD (D770_N771insSVD)	Ba/F3	**53**	268	39
SVD-vendor2 (D770_N771insSVD)	Ba/F3	**26**	150	13
ASV (V769_D770insASV)	Ba/F3	**78**	271	39
NPH (H773_V774insNPH)	Ba/F3	**75**	210	105
FQEA (A763_Y764insFQEA)	Ba/F3	**61**	122	32
NPG (H770_N771insNPG)	Ba/F3	**7**	26	3
IC_{50} values (nM) for WT EGFR				
A431	A431	**1660**	1003	1,124
EGFR Ba/F3	Ba/F3	**1960**	982	363
H2073	H2073	**921**	505	25
IC_{50} values (nM) for common and uncommon EGFR mutants				
EGFR L861Q	Ba/F3	**6**	–	19
EGFR G719S	Ba/F3	**8**	–	7
EGFR G719S/T263P	Ba/F3	**11**	–	24
EGFR L858R	Ba/F3	**3**	–	4
Exon 19 Deletion (E746-A750)	PC9	**13**	14	2

IC_{50} values for BLU-451 in a LoVo cell line harboring WT EGFR/KRAS, and in a Ba/F3 vector control line were 6980 nM and >10000 nM respectively. IC_{50}, half-maximal inhibitory concentration; WT, wildtype. All data from n≥3 measurements.

- In ligand binding assays (**Figure 1**), BLU-451 (1 μM) inhibited 1.7% of kinases tested by >90% (7/409). The off-target kinases (e.g., TEC family kinases) had a similarly positioned cysteine to EGFR, with the exception of PAK3. BLU-451 was highly selective compared to other EGFR inhibitors (e.g., osimertinib)
- In radiometric enzymatic assays run in the presence of 10 μM ATP, BLU-451 (1 μM) inhibited 2.2% of kinases by >90% (8/370)

Figure 1: BLU-451 is a highly selective kinase inhibitor



- BLU-451 (10 and 50 mg/kg QD) treatment of HuPrime® LU0387 in a PDX model harboring *EGFR* ex20ins (H773-V774insNPH) resulted in marked tumor regression with minimal change in body weight (**Figure 2A**)
- BLU-451 (10 and 50 mg/kg QD) treatment of a non-small cell lung cancer (NSCLC) LXFE 2478 PDX model harboring *EGFR* ex20ins (V769_D770insASV) resulted in regression with minimal change in body weight (**Figure 2B**)
- In a Ba/F3 CDX model harboring the *EGFR* ex20ins (V769_D770insASV), BLU-451 was equally effective as mobocertinib. Treatment with mobocertinib led to 17.5% body weight loss (**Figure 2C**)

Figure 2: Oral daily administration of BLU-451 resulted in tumor regression in NSCLC LU0387 (A) and LXFE 2478 (B) PDX and Ba/F3 subcutaneous CDX tumor models (C) harboring *EGFR* ex20ins mutations

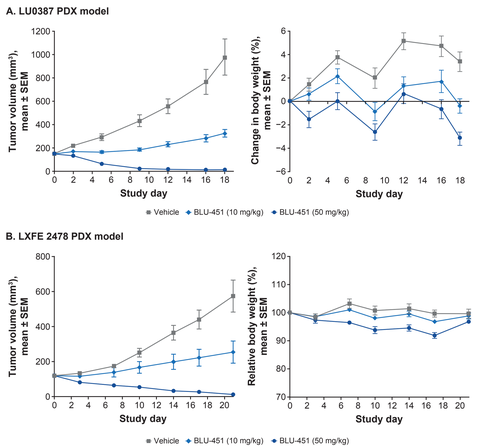
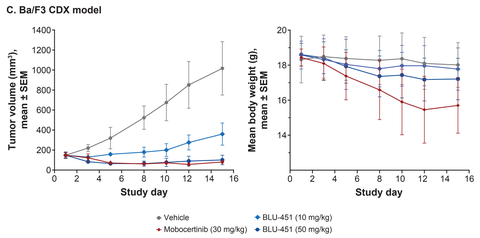


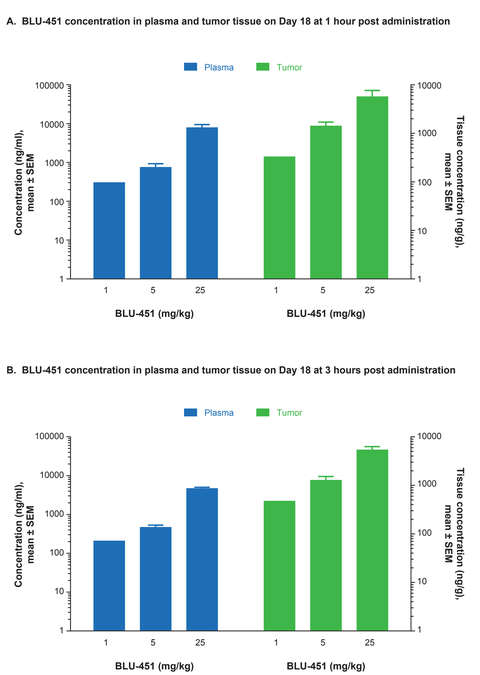
Figure 2: (continued)



SEM, standard error of the mean.

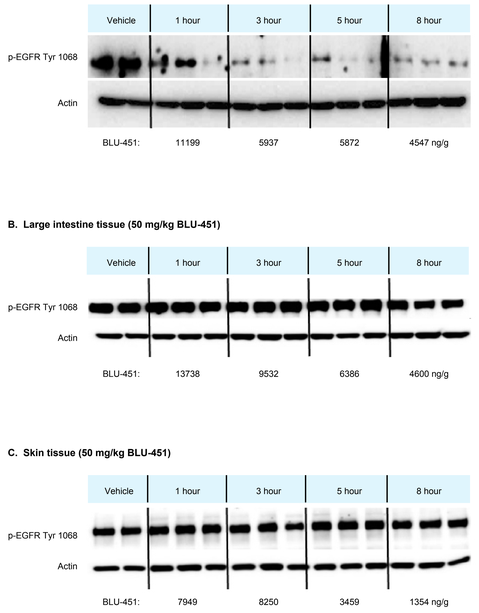
- Concentration of BLU-451 in plasma and tumor tissue on Day 18, 1 hour (h) (**Figure 3A**) and 3 h (**Figure 3B**) post administration, in the Ba/F3 cell-derived subcutaneous xenograft tumor model harboring the *EGFR* V769_D770insASV mutation
- BLU-451 exposure in plasma and tumor exceeded the corresponding cellular potency (IC_{50}=38.4 ng/mL) consistent with the observed activity

Figure 3: BLU-451 achieved high concentrations in both plasma and tumor tissues



- Single-dose PK/PD time course analysis of BLU-451 (50 mg/kg) was performed in a Ba/F3 *EGFR* ex20ins V769_D770insASV xenograft tumor model using phosphorylation of EGFR on tyrosine residue 1068 as a surrogate marker for EGFR activation (vehicle tissue taken 3 h post dose) (**Figure 4**)
- Similar exposures to BLU-451 were observed in tumor, large intestine, and skin tissues
- BLU-451 potently inhibited EGFR ex20ins in tumor tissue (**Figure 4A**) but had weak inhibition of WT EGFR in both large intestine and skin tissue samples (**Figure 4B-C**)

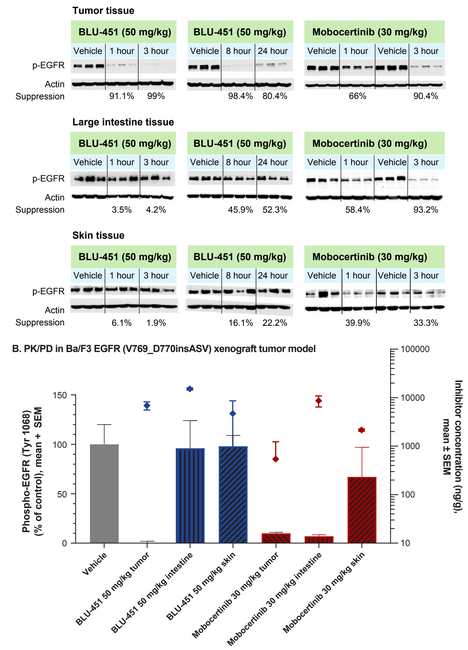
Figure 4: BLU-451 potently suppressed EGFR phosphorylation in *EGFR* ex20ins-dependent tumor tissue, but not in skin and large intestine tissue despite high exposures



pEGFR, phosphorylated EGFR; tyr 1068, tyrosine residue 1068.

- A single-dose PK/PD study of BLU-451 and mobocertinib was performed in a Ba/F3 EGFR (V769_D770insASV) xenograft tumor model using phosphorylation of EGFR on tyrosine residue 1068 as a surrogate marker for EGFR activation. Vehicle tissue for BLU-451 treatment comparisons was taken at either 3 h or 24 h post dose (**Figure 5**)
- BLU-451 potently inhibited EGFR ex20ins in the tumor tissue but poorly inhibited WT EGFR in the skin and large intestine tissue samples. Mobocertinib potently inhibited EGFR ex20ins in tumor tissue but also suppressed WT EGFR in skin and large intestine tissues (**Figure 5A**)
- PK/PD analysis showed comparable exposures for BLU-451 and mobocertinib in tumor, large intestine, and skin tissues 3 h post dosing. Both inhibitors suppressed EGFR phosphorylation in tumors, but only BLU-451 did not potently suppress EGFR phosphorylation in the large intestine and skin tissue samples (**Figure 5B**)

Figure 5: BLU-451 and mobocertinib potently inhibited *EGFR* ex20ins phosphorylation in tumor tissue, but only BLU-451 did not suppress WT EGFR phosphorylation in large intestine or skin tissue



Conclusions

- BLU-451 is a WT EGFR sparing, CNS-penetrant investigational EGFR ex20ins inhibitor
  - BLU-451 is a potent inhibitor of EGFR ex20ins as well as other uncommon EGFR point mutations
  - BLU-451 is selective for EGFR ex20ins relative to the human kinome and WT EGFR sparing in both cellular proliferation and *in vivo* preclinical tumor models
  - BLU-451 treatment resulted in marked tumor regression in both CDX and PDX *EGFR* ex20ins tumor models
  - Treatment with BLU-451, at doses which led to marked antitumor activity, had minimal impact on body weight in CDX and PDX *EGFR* ex20ins models
  - In *EGFR* ex20ins tumor models, BLU-451 had high plasma and tumor exposures that exceeded cell-based potency of BLU-451
  - PK/PD studies showed that BLU-451 potently suppressed EGFR ex20ins in tumors but spared WT EGFR in large intestine and skin tissue samples
- These *in vitro* and *in vivo* results strongly support a first-in-human phase 1/2 clinical trial of BLU-451 in patients with advanced or metastatic solid tumors harboring *EGFR* ex20ins mutations (NCT05241873)[6]

References

1. Riess JW *et al. J Thorac Oncol.* 2018;13:1560–1568; 2. Vyse S *et al. Signal Transduct Target Ther.* 2019;4:5; 3. Robichaux JP *et al. Nature.* 2021;597:732–737; 4. Riely GJ *et al. Cancer Discov.* 2021;11:1688–1699; 5. Park K *et al. J Clin Oncol.* 2021;39:3391–3402; 6. Study of BLU-451 in Advanced Cancers With EGFR Exon 20 Insertion Mutations. NCT05241873. https://clinicaltrials.gov/ct2/show/NCT05241873 [Accessed Mar 14, 2022].

Acknowledgements

Medical writing support was provided by Kyle Wild, MSc, and editorial support was provided by Travis Taylor, BA, all of Paragon, Knutsford, UK, supported by Blueprint Medicines Corporation, Cambridge, MA, according to Good Publication Practice guidelines.

Disclosures

BWM, AP, BR, TS, DJE, HJ, and PGP were employees of Lengo Therapeutics when this study was conducted. HA, SV, GH, IA, and RT are employees of Jubilant Biosys Limited, Bengaluru, India. Data in this poster were generated by Lengo Therapeutics and its collaborators.

Poster available for download at: