CASE REPORT

Crizotinib-Resistant *ROS1* G2101A Mutation Associated With Sensitivity to Lorlatinib in *ROS1*-Rearranged NSCLC: Case Report

Parvin Begum, PhD,[a] Wanyuan Cui, MBBS,[a] Sanjay Popat, PhD[a,b,*]

[a]*Lung Unit, Royal Marsden NHS Foundation Trust, London, United Kingdom*
[b]*Institute of Cancer Research, London, United Kingdom*

Received 14 March 2022; revised 31 May 2022; accepted 28 June 2022
Available online - 5 July 2022

ABSTRACT

ROS1 gene rearrangements occur in 1% to 2% of NSCLC. Acquired "on-target" mutations within the ROS1 kinase domain are a known resistance mechanism to the first-line ROS1 inhibitor crizotinib. Here, we report the first case of a patient with an acquired *ROS1* G2101A resistance mutation after first-line crizotinib, who responded to lorlatinib. The response was dramatic but short in duration.

Keywords: Non–small cell lung cancer; *ROS1* fusion; ctDNA; Lorlatinib; Case report

Introduction

ROS1 gene fusions are known oncogenic drivers in NSCLC, accounting for 1% to 2% of cases. Crizotinib and entrectinib are approved by the Food and Drug Administration and European Medicines Agency for first-line treatment of *ROS1*-rearranged NSCLC. Nevertheless, secondary resistance to these remains a challenge. Lorlatinib is a brain-penetrant, adenosine triphosphate-competitive, small molecule inhibitor of *ALK* and *ROS1*, which has efficacy in patients with both *ALK* and *ROS1* kinase resistance mutations, such as the solvent-front G1202R mutation and its *ROS1* analog G2032R.[1] Genomic predictors of lorlatinib durable response post crizotinib or post entrectinib are important in drug decision-making.

Case Presentation

A 75-year-old female never smoker presented with dyspnea. Results of initial computed tomography (CT) pulmonary angiogram with subsequent 18F-fluorodeoxyglucose positron emission tomography (PET)-CT and brain magnetic resonance imaging (MRI) revealed a right lower lobe lung primary with widespread metastases involving supraclavicular and bilateral mediastinal nodes, pleura (with effusion), lungs, liver, adrenal gland, multiple bone, and multiple brain sites. Result of pleural aspirate confirmed metastatic ROS1-positive, TTF1-positive adenocarcinoma by immunohistochemistry. Circulating tumor DNA (ctDNA) next-generation sequencing (NGS) (Guardant360 CDx, Guardant Health, Redwood City, CA) confirmed a dominant *CD74-ROS1* fusion at 2.0% variant allele frequency (VAF), alongside additional driver variants, including *CDKN2A* E120*, *TP53* H193Y, *TP53* A159D, and *KRAS* K117N all at lower VAFs.

The patient commenced crizotinib 250 mg twice daily with denosumab because entrectinib was unavailable. Result of response imaging with PET-CT and MRI brain

*Corresponding author.

Disclosure: Dr. Cui declares receiving grant funding from the Breast Cancer Trials and honoraria from Pfizer, United States, Merck, United States, Janssen, and AstraZeneca, United Kingdom outside of the submitted work. Prof. Popat is a consultant to Amgen, AstraZeneca, Bayer, Beigene, Blueprint, Bristol-Myers Squibb, Boehringer Ingelheim, Daiichi Sankyo, Guardant Health, Janssen, Lilly, Merck KGaA, Novartis, Roche, and Takeda. Dr. Begum declares no conflict of interest.

Address for correspondence: Sanjay Popat, PhD, Lung Unit, Royal Marsden Hospital, Fulham Road, London, SW3 6JJ, United Kingdom. E-mail: sanjay.popat@rmh.nhs.uk

Cite this article as: Begum P, Cui W, Popat S. Crizotinib-resistant *ROS1* G2101A mutation associated with sensitivity to lorlatinib in *ROS1*-rearranged NSCLC: case report. *JTO Clin Res Rep*. XXXX;X:XXXXXX.

ISSN: 2666-3643

https://doi.org/10.1016/j.jtocrr.2022.100376

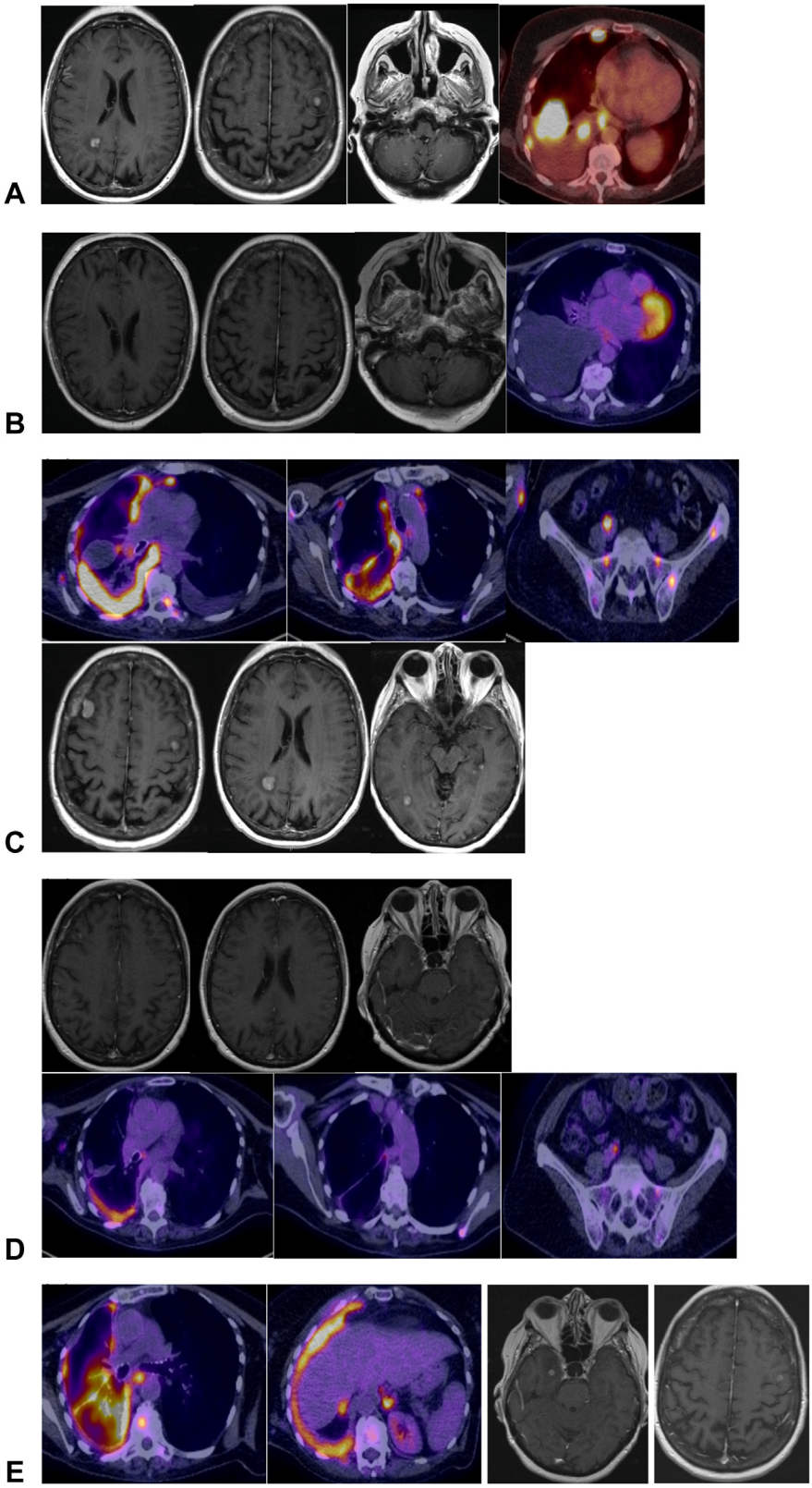

Figure 1. Radiology images on therapy. (*A*) CT, FDG PET-CT, and MRI brain images at diagnosis. (*B*) Response imaging after two months on crizotinib, revealing a good response. (*C*) Imaging at progression on crizotinib, coinciding with emergence of a *ROS1* G2101A variant. (*D*) Imaging revealing a good extracranial and intracranial response after 1 month of lorlatinib treatment. (*E*) Imaging revealing extracranial progression and intracranial oligoprogression at two sites, after 2 months of lorlatinib treatment. NGS result at this point revealed loss of G2101A and emergence of *ROS1* G2032R and L2086F. CT, computed tomography; FDG PET-CT, 18F-fluorodeoxyglucose positron emission tomography-computed tomography; MRI, magnetic resonance imaging.

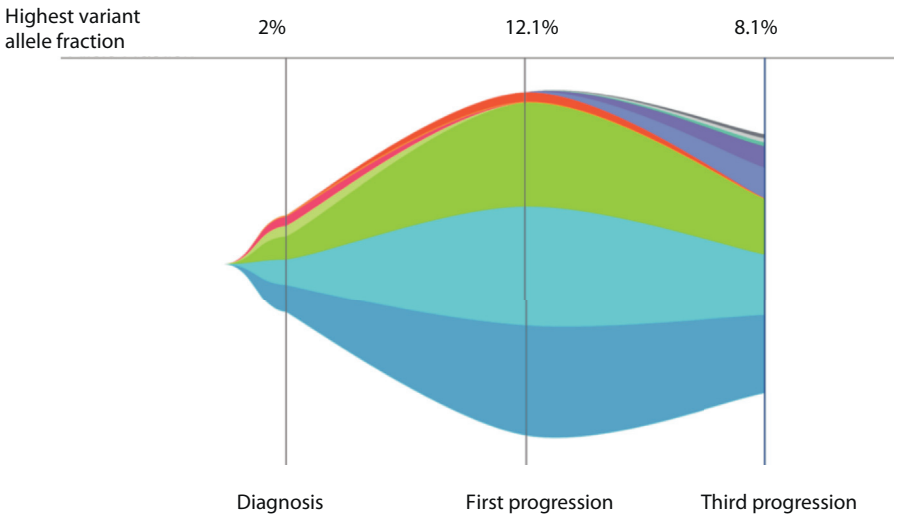

Figure 2. Guardant360 tumor response map. Tumor response map illustrating the dynamic changes in variant allele fractions of (% cfDNA) observed somatic variants at each sample submission. Amplifications are not plotted. cfDNA, cell-free DNA.

after 2 months revealed a rapid intracranial and extracranial partial response (Fig. 1).

Nevertheless, imaging after 3 additional months identified extracranial and intracranial progression (at pleural, nodal, adrenal, skeletal, and brain sites, Fig. 1). At this point, repeat ctDNA NGS for therapy selection identified progressive dominance of the truncal *CD74-ROS1* driver (VAF10.8%) alongside the *CDKN2A* E120* and *TP53* H193Y variants. In addition, an acquired *ROS1* G2101A mutation was identified (VAF 0.5%; Fig. 2). After discussion with the patient and family, she commenced lorlatinib (100 mg once daily). Her response evaluation PET-CT and brain MRI scan 4 weeks after commencement revealed an excellent intracranial and extracranial partial response (Fig. 1) underpinning excellent clinical benefit.

After 2 months, dyspnea deteriorated and PET-CT with brain MRI confirmed widespread extracranial progression with predominantly maintained intracranial response. Result of repeat ctDNA NGS revealed reduction in the *CD74-ROS1* fusion (VAF 7.2%), *CDKN2A* E120*, and *TP53* H193Y drivers, loss of the *ROS1* G2101A target, and emergence of the lorlatinib compound *ROS1* G2032R (VAF 1.8%) and L2086F (VAF 0.6%) resistance mutations (Fig. 3). The patient commenced third-line carboplatin-pemetrexed-bevacizumab combination therapy

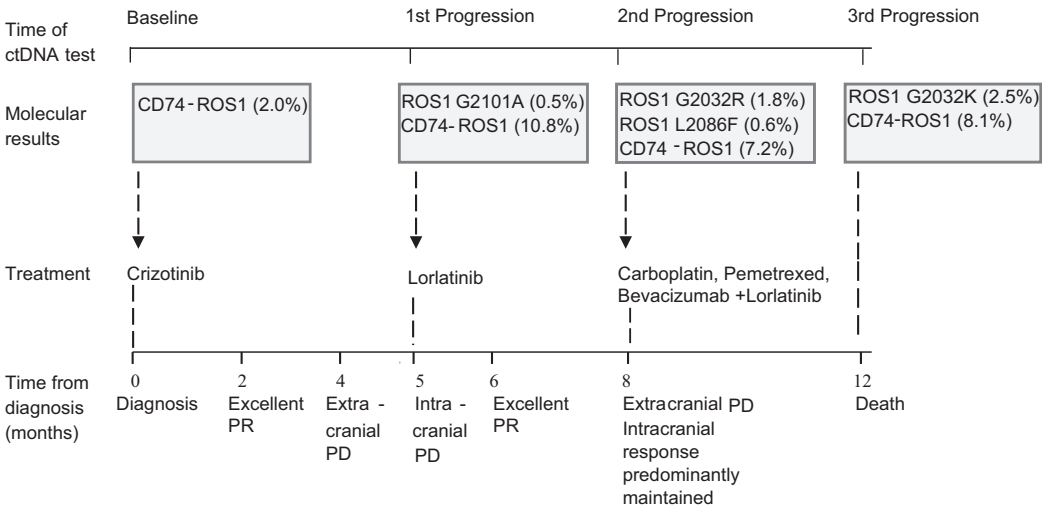

Figure 3. A timeline of the patient's clinical, treatment, and molecular profile. At the first progression on crizotinib treatment, there is emergence of a *ROS1* G2101A variant. Lorlatinib is started at this point with excellent response. At the second progression, there is loss of G2101A and emergence of *ROS1* G2032R and L2086F. At the final progression, while still on lorlatinib, there is emergence of a *ROS1* G2032K variant. Variant allele frequencies are provided in parentheses. PD, progressive disease; PR, partial response.

with continued lorlatinib for ongoing intracranial control, and in parallel, she received stereotactic radiosurgery to two intracranial sites owing to oligoprogressive metastases with edema. Despite an initial response after two cycles of systemic therapy associated with clinical benefit, result of PET-CT following cycle 4 revealed disease progression. Result of ctDNA NGS testing revealed progression of the *CD74-ROS1* driver (VAF 8.1%) with minor reduction of the *CDKN2A* E120* (VAF 5.3%) and *TP53* H193Y (VAF 6.0%) drivers, loss of *ROS1* G2032R, gain of *ROS1* G2032K, and gain in other variants, including *NRAS* G12D. She unfortunately continued to deteriorate and was admitted with dyspnea, acute kidney injury, a raised troponin level, and multiple watershed embolic infarcts in both cerebral hemispheres on MRI imaging results. Progressive clinical and neurological deterioration occurred thereafter, shortly followed by the patient's death.

Discussion

Crizotinib and entrectinib are first-line treatments for *ROS1*-rearranged NSCLC. Nevertheless, acquired resistance remains a major challenge in its management, especially the development of solvent-front and gatekeeper mutations with no other ROS1 kinase inhibitors currently licensed. ctDNA provides a useful minimally invasive tool for the temporal detection of variants that can identify on- or off-target resistance mechanisms and potentially guide treatment. Here, we reveal that the acquisition of the on-target G2101A *ROS1* crizotinib-resistance mutation is associated with lorlatinib response: a novel clinical finding. G2101A has previously been identified as crizotinib resistant in preclinical assays,[2] an analog of *ALK* G1269A, itself previously found to result in responses of short duration to next-generation ALK inhibitors[3] and retains preclinical sensitivity to foretinib,[2] although no preclinical or clinical data have previously been reported for its sensitivity to lorlatinib. G2101A sits away from the *ROS1* L2026 gatekeeper residue[1] and outside the G2302 solvent-front region, and its precise mechanism of crizotinib resistance and lorlatinib sensitivity remains uncertain. It remains unclear whether G2101A results in lorlatinib response of brief duration as observed in this case or whether the brief lorlatinib sensitivity observed here was underpinned by the volume of her disease and diversity of other drivers (*CDKN2A* and *TP53*). Moreover, this case highlights that G2101A resistance may be mediated by on-target compound G2032R-L2086F *ROS1* mutations, previously associated with lorlatinib resistance,[4] as well as with conferring resistance to taletrectinib (DS6051b) and sensitivity to cabozantinib.[5] L2086F, being analogous to *ALK* L1256F, which confers crizotinib and lorlatinib resistance in an *ALK*-positive NSCLC preclinical model through steric interference.[4] Finally, this case demonstrates the utility of ctDNA NGS at diagnosis and each progression point allowing identification of extracranial acquired ROS1-kinase inhibitor resistance mechanisms and optimal drug decision-making.

Conclusion

This is the first report of a *ROS1* G2101A mutation associated with acquired crizotinib resistance and lorlatinib sensitivity with immediate response. We further identify lorlatinib resistance through G2101A loss and compound G2032R-L2086F gain, adding additional evidence for lorlatinib sensitivity to *ROS1* NSCLCs with specific acquired crizotinib-resistance mutations.

CRediT Authorship Contribution Statement

Sanjay Popat: Conceptualization ideas, Writing manuscript, review and editing.

Coordinated patient care.

Parvin Begum: Writing manuscript, review and editing, Creating figures.

Wanyuan Cui: Writing manuscript, review and editing.

Acknowledgments

The authors acknowledge the patient for providing informed consent for her case to be reported. Drs. Popat and Cui acknowledge the National Health Service funding to the National Institute for Health and Care Research Biomedical Research Centre at the Royal Marsden Hospital/Institute of Cancer Research.

References

1. Zou HY, Li O, Engstrom LD, et al. PF-06463922 is a potent and selective next-generation ROS1/ALK inhibitor capable of blocking crizotinib-resistant ROS1 mutations. *Proc Natl Acad Sci*. 2015;112:3493–3498.
2. Song A, Kim TM, Kim D, et al. Molecular changes associated with acquired resistance to crizotinib in *ROS1*-rearranged non-small cell lung cancer. *Clin Cancer Res*. 2015;21:2379–2387.
3. Michels S, Scheel AH, Wündisch T, et al. *ALK* ^{G1269A} mutation as a potential mechanism of acquired resistance to crizotinib in an *ALK*-rearranged inflammatory myofibroblastic tumor. *npj Precision Onc*. 2017;1:4.
4. Lin JJ, Choudhury NJ, Yoda S, et al. Spectrum of mechanisms of resistance to crizotinib and lorlatinib in *ROS1* fusion-positive lung cancer. *Clin Cancer Res*. 2021;27:2899–2909.
5. Papadopoulos KP, Borazanci E, Shaw AT, et al. U.S. phase I first-in-human study of taletrectinib (DS-6051b/AB-106), a ROS1/TRK inhibitor, in patients with advanced solid tumors. *Clin Cancer Res*. 2020;26:4785–4794.