Short progression-free survival of ALK inhibitors sensitive to secondary mutations in ALK-positive NSCLC patients

Naoki Haratake1, Takashi Seto1, Shinkichi Takamori1, Ryo Toyozawa1, Kaname Nosaki1, Naoko Miura1, Taro Ohba1, Gouji Toyokawa2, Kenichi Taguchi3, Masafumi Yamaguchi1, Mototsugu Shimokawa4,5 & Mitsuhiro Takenoyama1

1 Department of Thoracic Oncology, National Hospital Organization Kyushu Cancer Center, Fukuoka, Japan
2 Department of Thoracic Surgery, National Hospital Organization Kyushu Medical Center, Fukuoka, Japan
3 Department of Pathology, National Hospital Organization Kyushu Cancer Center, Fukuoka, Japan
4 Department of Biostatistics, Yamaguchi University Graduate School of Medicine, Yamaguchi, Japan
5 Clinical Research Institute, National Hospital Organization Kyushu Cancer Center, Fukuoka, Japan

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Correspondence
Takashi Seto, Department of Thoracic Oncology, National Hospital Organization Kyushu Cancer Center, 3-1-1 Notame, Minami-ku, Fukuoka 811-1395, Japan. Tel: +81 92 541 3231 Fax: +81 92 551 4585 Email: setocruise@gmail.com

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Abstract

Background: Most non-small cell lung cancer (NSCLC) patients relapse on anaplastic lymphoma kinase-tyrosine kinase inhibitor (ALK-TKI) therapy because of acquired resistance. Rebiopsy is recommended to provide optimal therapy after relapse for some ALK-TKI therapies; however, little clinical data exists on the clinical efficacy of ALK-TKI tailored to secondary mutation.

Methods: A retrospective study was conducted to analyze the patterns of ALK-TKI treatment and clinical outcomes, including progression free survival (PFS), of ALK-positive NSCLC patients who received rebiopsy. Based on the rebiopsy results, secondary mutations in the ALK gene that were shown to be associated with the efficacy of ALK-TKI therapy in the preclinical or clinical setting were defined as “sensitive mutations (SM)”.

Results: Among 71 patients who received ALK-TKI for NSCLC at our institution, 20 patients received rebiopsy, and secondary SM were found in eight patients. The objective response rate (ORR) of the cases with SM who received ALK-TKI therapy was 88.9%, while the ORR of the patients without SM who received ALK TKI or chemotherapy was 20.0%; however, the PFS of the patients with SM was relatively short (with SM vs. without SM: 5.6 months vs. 5.1 months).

Conclusions: The selection of ALK-TKI based on the rebiopsy result was associated with a high ORR and relatively short PFS. The mechanism responsible for the short PFS of sensitive ALK-TKI to secondary mutation should be clarified.

Introduction

Lung cancer is the leading cause of cancer-related death worldwide. Recent advances in chemotherapy and molecular targeted therapy have led to the remarkable improvement of survival of lung cancer patients, especially those with non-small cell lung cancer (NSCLC). From 3%–5% of NSCLC patients have anaplastic lymphoma kinase (ALK) rearrangement. The superiority of crizotinib, a first generation ALK-tyrosine kinase inhibitor (ALK-TKI), to standard chemotherapy has been demonstrated in patients with ALK-positive advanced NSCLC previously treated with platinum-based chemotherapy. Sequentially, next generation ALK-TKIs, such as alectinib, ceritinib, brigatinib and lorlatinib, have shown remarkable clinical efficacy in ALK-positive NSCLC. However, even if patients initially respond well to ALK-TKI, the majority eventually experience disease progression due to various mechanisms, including secondary mutations within the ALK tyrosine kinase domain and activation of alternative signaling pathways.

Next-generation ALK-TKIs have been reported to overcome some secondary mutations mediating resistance to crizotinib. Importantly, each ALK-TKI has different
sensitivity to secondary mutations, and the clinical significance of rebiopsy to clarify the secondary mutation has been reported in various studies. For instance, L1196M, which is a common gatekeeper mutation in ALK-positive NSCLC, is resistant to crizotinib and sensitive to next generation ALK-TKIs. On the other hand, it was reported that the solvent front ALK G1202R and compound ALK C1156Y/L1198F mutations are only sensitive to lorlatinib and crizotinib, respectively.

Thus, rebiopsy in ALK-TKI-refractory patients has attracted increasing attention as it assists in determining optimal treatment strategy; however, it is unclear whether or not rebiopsy should be performed to improve the prognosis of the ALK-positive NSCLC patients.

The objective of this study was to analyze the significance of sequential therapy with ALK-TKI based on the results of rebiopsy in ALK-positive NSCLC patients.

**Methods**

Patients with advanced or recurrent, histologically-confirmed ALK-positive NSCLC, who received ALK inhibitors in clinical practice at National Kyushu Cancer Center from March 2007 to April 2018 were included in this retrospective analysis. The ALK status of the patients was confirmed by immunohistochemistry, fluorescence in situ hybridization, or a reverse-transcriptase polymerase chain reaction (RT-PCR). Positivity of any of these tests indicated the rearrangement of the ALK gene. In addition, ALK fusion variants and secondary mutations were analyzed by a RT-PCR and direct-sequencing.

We retrospectively evaluated the patient characteristics, treatment regimens, and clinical outcomes. Data on the treatment patterns and outcomes were collected from medical records.

Tumor responses were assessed with computed tomography (CT), magnetic resonance imaging (when clinically indicated), and positron emission tomography-CT (when clinically indicated), before and during treatment, and were repeated approximately every two months. Responses were classified as progressive disease, stable disease, partial response, complete response or nonevaluable, on the basis of response evaluation criteria in solid tumors (RECIST) 1.1.

A rebiopsy was performed in the lesions that had shown progression. Based on the rebiopsy results, secondary mutations in the ALK gene that were shown to be associated with the efficacy of ALK-TKI therapy in the preclinical or clinical setting were defined as “sensitive mutations (SM)” (i.e., alectinib to L1196M). The sensitivity of the secondary mutations in the 20 cases to each ALK-TKI is shown in Table 1. In most cases, lorlatinib had sensitivity to the secondary mutations, but lorlatinib was not available at the time in some cases. Suitable and available ALK-TKIs were selected based on the rebiopsy findings. A resistance mutation analysis was performed using direct sequencing with capillary electrophoresis of biopsy samples obtained after ALK-TKI treatment.

This retrospective study was approved by the Ethics Committee of National Kyushu Cancer Center (2013–77), and conducted in accordance with the Declaration of Helsinki.

**Results**

**Clinicopathological characteristics of patients**

Seventy-one patients who had ALK-positive NSCLC were treated with ALK-TKIs. Among these, 20 ALK-positive patients underwent rebiopsy after disease progression on an ALK-TKI. The median follow-up period was 37.1 months. The characteristics of the patients are shown in Table 2.

The median age of the patients was 45 years (range: 28–68 years). There were eight male patients (40.0%) and 12 female patients (60.0%). Four patients (20.0%) had a history of smoking. Eighteen patients were diagnosed with clinical stage IV (90.0%), one with clinical stage III (5.0%), and one with postoperative recurrence (5.0%). The histological diagnosis was adenocarcinoma in all 20 patients. In addition, variants 1, 2, 3, and others were detected by a RT-PCR before first line treatment in seven (35.0%), six (30.0%), four (20.0%), and three (5.0%) patients, respectively. Four (20.0%) patients underwent three or more biopsies after disease progression.

**Treatment**

In first line treatment, 16 (80.0%), and four (20.0%) patients received cytotoxic chemotherapy and ALK-TKI therapy, respectively. With regard to the ALK-TKIs that were administered, nine (45.0%), nine (45.0%), and two (10.0%) patients received crizotinib, alectinib, and ceritinib as the first ALK-TKI treatment.

Regarding the number of ALK-TKIs administered, eight (40.0%) and 12 (60.0%) patients received two and three

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**Table 1** The sensitivity of anaplastic lymphoma kinase-tyrosine kinase inhibitors to secondary mutations in the 20 cases

|                          | Crizotinib | Ceritinib | Alectinib | Lorlatinib |
|--------------------------|-----------|-----------|-----------|------------|
| L1196M                   | Resistant | Sensitive | Resistant | Sensitive  |
| I1171T                   | Resistant | Sensitive | Resistant | Sensitive  |
| I1171N                   | Resistant | Sensitive | Resistant | Sensitive  |
| G1269A                   | Resistant | Sensitive | Sensitive | Sensitive  |
| G1202R                   | Resistant | Resistant | Resistant | Resistant  |
| G1123S + C1156Y          | Resistant | Resistant | Resistant | Resistant  |
| C1156Y + G1123S          | Resistant | Resistant | Resistant | Resistant  |
ALK-TKIs, respectively. Fifteen (75.0%) received crizotinib, 15 (75.0%) received alectinib, and 15 (75.0%) received ceritinib, six (30.0%) received lorlatinib. Detailed information regarding the treatment patterns is shown in Figure 1 with individual swimmer plots (time to treatment failure) for all patients.

Molecular characteristics of patients undergoing rebiopsy

The results of rebiopsy were used for the evaluation of the mechanisms of ALK-TKI resistance. In the rebiopsied specimen, the original ALK rearrangement existed in all 20 patients. Secondary mutations were identified in 10 of all 24 biopsy specimens (41.7%). Secondary mutations included I1171N (n = 2), I1171T (n = 1), G1296A (n = 1), L1196M (n = 5), G1202deletion (n = 1), G1123S + C1156Y and C1156Y + G1202R (n = 1 [in the same one case at the second and third biopsy, respectively]). The individual responses to the next ALK-TKI of each patient who received a repeat biopsy are listed in Tables 3 and 4 and Figure 1.

We first compared the objective responses to ALK-TKI therapy of 20 rebiopsied patients with and without secondary SM. The objective response rate (ORR) of the cases with secondary SM who received ALK-TKI therapy was 88.9%, while the ORR of the patients without secondary SM who received ALK TKI was 20.0% (Table 5).

We next compared the treatment outcomes to sequential therapy among patients with and without secondary SM. The median progression free survival (PFS) achieved by the eight patients with nine secondary SM cases who received ALK-TKI therapy was 5.6 months, while the median PFS of the 12 patients with 16 cases with nonsecondary SM who received next line treatment (nontailored ALK-TKI or chemotherapy) was 5.1 months (Table 5). With regard to overall survival (OS), among the eight patients with at least one secondary SM on rebiopsy, the median OS was 37.0 months, while the median OS among the patients without any secondary sensitive ALK mutations was 49.0 months (Table 5).

Out of the 20 cases, five cases showed progression in the central nervous system (CNS) during the next line therapy. Of those, just one case (Patient ID Number 10 in Table 3, in Fig 1) showed progression in the CNS only, and the other four cases (Patient ID Numbers 5, 7, 15, 16 in Table 3, in Fig 1) showed progression in both the CNS as well as extracranial lesions.

Detailed individual data on the secondary mutations are provided in Tables 3 and 4 and Figure 1.

### Discussion

Previous studies have reported that rebiopsy could provide further information, including histological or genetic changes that might be helpful in optimizing the next treatment24,25; however, little clinical data exists regarding the prognostic impact of rebiopsy on ALK-positive NSCLC patients. In this retrospective analysis, we evaluated the treatment course and clinical efficacy of ALK-TKI in ALK-positive NSCLC patients who received rebiopsy after

### Table 2 The clinical characteristics of the patients treated with ALK-TKIs

|                      | Total (n = 20) | Secondary sensitive mutation* (+) (n = 8) | Secondary sensitive mutation* (−) (n = 12) |
|----------------------|---------------|------------------------------------------|------------------------------------------|
| Median age (years)   | Median 45     | 42.5                                     | 50                                       |
| Range                | 27–68         | 27–53                                    | 40–67                                    |
| Sex                  |               |                                          |                                          |
| Male                 | 8 (40.0%)     | 2 (25.0%)                                | 6 (50.0%)                                |
| Female               | 12 (60.0%)    | 6 (75.0%)                                | 6 (50.0%)                                |
| ECOG PS              |               |                                          |                                          |
| 0                    | 12 (60.0%)    | 4 (50.0%)                                | 8 (66.7%)                                |
| 1                    | 7 (35.0%)     | 4 (50.0%)                                | 3 (25.0%)                                |
| 2                    | 1 (5.0%)      | 0 (0.0%)                                 | 1 (8.3%)                                 |
| Smoking history      |               |                                          |                                          |
| Never                | 16 (80.0%)    | 8 (100.0%)                               | 8 (66.7%)                                |
| Former/current       | 4 (20.0%)     | 0 (0.0%)                                 | 4 (33.3%)                                |
| Histology            |               |                                          |                                          |
| Adenocarcinoma       | 20 (100.0%)   | 8 (100.0%)                               | 12 (100.0%)                              |
| Clinical stage       |               |                                          |                                          |
| III                  | 1 (5.0%)      | 1 (12.5%)                                | 0 (0.0%)                                 |
| IV                   | 18 (90.0%)    | 6 (75.0%)                                | 12 (100.0%)                              |
| Postoperative recurrence | 1 (5.0%)  | 1 (12.5%)                                | 0 (0.0%)                                 |
| ALK variants         |               |                                          |                                          |
| Variant 1            | 7 (35.0%)     | 2 (25.0%)                                | 5 (41.7%)                                |
| Variant 2            | 6 (30.0%)     | 4 (50.0%)                                | 2 (16.7%)                                |
| Variant 3            | 4 (20.0%)     | 1 (12.5%)                                | 3 (25.0%)                                |
| Variant 5            | 2 (10.0%)     | 0 (0.0%)                                 | 2 (8.3%)                                 |
| Variant 1 + 6        | 1 (5.0%)      | 1 (12.5%)                                | 0 (0.0%)                                 |

ALK, anaplastic lymphoma kinase; ECOG PS, Eastern Cooperative Oncology Group performance status; TKI, tyrosine kinase inhibitor.
relapse on ALK-TKI, and the administration of ALK-TKIs based on the secondary sensitive mutations was associated with a high ORR and relatively short PFS (87.5% and 5.4 months, respectively).

Some clinical trials have demonstrated that there is good efficacy of second generation ALK-TKI in comparison to chemotherapy for crizotinib-pretreated ALK-positive NSCLC patients. In addition, some studies showed the remarkable efficacy of next generation ALK-TKI tailored to the secondary mutation. In the current study, each patient’s in vitro ALK-TKI-sensitivity profile and ALK resistance mutations were used to select the next ALK-TKI for the treatment of ALK-TKI therapy refractory patients. For example, L1196M (shown in cases 1–4, in Fig 1) and I1171N (shown in cases 6 and 7 in Fig 1) are reported to be associated with sensitivity to ceritinib, brigatinib, and lorlatinib, and resistance to crizotinib and alectinib. Similarly, I1171T (shown in Fig 1 [case 5]) and G1296A (shown in Fig 1 [case 8]) are reported to be associated with sensitivity to crizotinib and lorlatinib, and resistance to crizotinib and alectinib. We respectively selected the suitable ALK-TKI based on these data, and good responses were observed in those cases (ORR: 88.9%; Fig 1 [cases 1–8] and Table 3). On the other hand, in the cases without secondary SM (Fig 1 [cases 9–20]), chemotherapy or remaining ALK-TKI was selected, and the ORR was relatively low (20.0%); however, the PFS of the patients with SMs was relatively short (with SM vs. without SM: 5.6 months vs. 5.1 months). These results suggest that once resistance to an ALK-TKI emerges, even when it is a secondary SM, some ALK-independent resistance mechanisms, such as bypass signaling, might emerge in a combinatory manner or over a short period after ALK-TKI failure, which should be clarified by comprehensive testing modalities, such as next generation sequencing. If a new treatment strategy (i.e., combination treatment with a next generation ALK-TKI and agents targeting the bypass track) that overcomes this resistance mechanism is established, then the prognosis of ALK-TKI refractory patients will be remarkably improved in comparison to the existing treatment. Recently, the phase II study of lorlatinib has been reported. The cohort with at least one secondary mutation in the baseline tumor biopsy had a PFS of 11.0 months, while the cohort with no secondary mutation in the baseline biopsy had a PFS of 5.4 months. This result suggests that the PFS in the current study was relatively short; however, the previous cohorts included patients treated with not only second-generation ALK-TKIs, but also crizotinib as pretreatment of lorlatinib. Whether or not only crizotinib or noncrizotinib ALK-TKIs were administered as pretreatment might affect the efficacy of lorlatinib. Among the patients treated only with crizotinib as pretreatment of
### Table 3: Detailed Information on Each Patient Who Underwent a Rebiopsy (Patients with a Sensitive Mutation at the First Rebiopsy)

| Patient No. | ALK-TKI Prior to Rebiopsy | Pattern of Progression on the Immediate Preceding ALK-TKI | Rebiopsy Result (biopsy site) SM/not SM | Immediate next-line ALK-TKI Therapy | Treatment-related adverse events (Grade 3 or 4) | Response by RECIST 1.1 | PFS (months) | Pattern of Progression (including the rebiopsy site or not) |
|-------------|---------------------------|----------------------------------------------------------|-----------------------------------------|-------------------------------------|-----------------------------------------------|---------------------------|--------------|----------------------------------------------------------|
| 1–1         | Alectinib                 | Extracranial                                             | L1196M (LN) SM                          | Ceritinib (750 → 300 mg/day)        | Diarrhea (Grade 3)                           | PR                        | 21.0         | Extracranial (not including)                             |
| 1–2         | Ceritinib                 | Extracranial                                             | G1202del (effusion) not SM              | Alectinib (600 mg/day)              | -                                             | PR                        | 7.5          | Extracranial (including)                                 |
| 2           | Crizotinib                | Extracranial                                             | L1196M (PM) SM                          | Alectinib (600 mg/day)              | -                                             | PR                        | 19.4         | Extracranial (including)                                 |
| 3           | Crizotinib                | Extracranial                                             | L1196M (LN) SM                          | Ceritinib (300 mg/day)              | -                                             | PR                        | 8.0          | Extracranial (including)                                 |
| 4–1         | Crizotinib                | Extracranial                                             | 1196M (liver) SM                        | Alectinib (600 mg/day)              | -                                             | SD                        | 4.0          | Extracranial (including)                                 |
| 4–2         | Alectinib                 | Extracranial                                             | 1196M (liver) SM                        | Lorlatinib (100 mg/day)             | -                                             | PR                        | 9.3          | Extracranial (including)                                 |
| 4–3         | Lorlatinib                | Extracranial                                             | 1196M (liver) not SM                    | Chemotherapy                       | -                                             | PD                        | 0.9          | Extracranial (including)                                 |
| 5           | Crizotinib                | Extracranial                                             | I1171T (effusion) SM                    | Ceritinib (450 mg/day)              | Platelet count decreased (Grade 3)            | PR                        | 4.1          | CNS + extracranial (including)                           |
| 6           | Alectinib                 | Extracranial                                             | I1171N (liver) SM                       | Ceritinib (300 mg/day)              | -                                             | PR                        | 5.6          | Extracranial (not including)                             |
| 7           | Alectinib                 | CNS + extracranial                                        | I1171N (lung) SM                        | Ceritinib (750 → 600 mg/day)        | -                                             | PR                        | 2.2          | CNS + extracranial (including)                           |
| 8           | Alectinib, Crizotinib     | Extracranial                                             | G1269A (lung) SM                        | Ceritinib (750 mg)                 | -                                             | PR                        | 1.0          | Non-cancer-related death                                 |

ALK, anaplastic lymphoma kinase; CNS, central nervous system; LN, lymph node; PD, progressive disease; PFS, progression-free survival; PM, pulmonary metastasis; PR, partial response; SD, stable disease; SM, sensitive mutation; RECIST 1.1, response evaluation criteria in solid tumors 1.1; TKI, tyrosine kinase inhibitor. Ceritinib was administered between meals.
## Table 4
Detailed information on each patient who underwent rebiopsy (patients without a sensitive mutation at the first rebiopsy)

| Patient No. | ALK-TKI (s) prior to rebiopsy | Pattern of progression on the immediate preceding ALK-TKI | Rebiopsy result (biopsy site) | SM/not SM to next-line ALK-TKI | Immediate next-line therapy | Treatment-related adverse events (Grade 3 or 4) | Response by RECIST 1.1 | PFS (months) | Pattern of progression (including the rebiopsy site or not) |
|-------------|-------------------------------|---------------------------------------------------------|-------------------------------|--------------------------------|-------------------------------|------------------------------------|----------------------|-------------|-------------------------------------------------------------|
| 9–1         | Crizotinib Extracranial       | C1156Y + G1202R (liver) non-SM                          | Ceritinib (750 → 600 mg/day)  | ALT increased (Grade 3)       | SD                            | 25.2                               | Extracranial (including)             |
| 9–2         | Ceritinib Extracranial        | C1156Y + G1123S (liver) non-SM                          | Alectinib (600 mg/day)        | -                              | PR                            | 5.8                                | Extracranial (including)             |
| 10          | Crizotinib CNS + extracranial | No mutation (lung) non-SM                               | Alectinib (600 mg/day)        | -                              | SD                            | 16.2                               | CNS (not including)                 |
| 11          | Alectinib Extracranial        | G1202R (pleural dissemination) non-SM                   | Ceritinib (750 → 500 mg/day)  | AST increased (Grade 3)        | SD                            | 13.1                               | Extracranial (including)             |
|             |                               |                                                        |                               |                                | Fatigue (Grade 3)             |                                    |                                    |
| 12          | Ceritinib Extracranial        | No mutation (bone) non-SM                               | Alectinib (600 mg/day)        | -                              | SD                            | 3.1                                | Extracranial (including)             |
| 13          | Crizotinib Extracranial       | No mutation (LN) non-SM                                 | Ceritinib (600 mg/day)        | -                              | PR                            | 35.0                               | Extracranial (including)             |
| 14          | Alectinib CNS + extracranial  | No mutation (renal) non-SM                              | Ceritinib (600 mg/day)        | -                              | SD                            | 8.5                                | Extracranial (including)             |
| 15          | Alectinib Extracranial        | No mutation (lung) non-SM                               | Chemotherapy                  | -                              | SD                            | 1.7                                | CNS + Extracranial (including)       |
| 16          | Crizotinib CNS + extracranial | No mutation (LN) non-SM                                 | Chemotherapy                  | -                              | SD                            | 2.7                                | CNS + Extracranial (including)       |
| 17          | Alectinib Extracranial        | No mutation (lung) non-SM                               | Crizotinib (500 mg/day)       | -                              | PD                            | 1.4                                | Extracranial (including)             |
| 18          | Alectinib CNS + extracranial  | No mutation (lung) non-SM                               | Crizotinib (500 mg/day)       | -                              | PD                            | 5.1                                | Extracranial (including)             |
| 19          | Crizotinib CNS + extracranial | No mutation (effusion) non-SM                            | Ceritinib (450 mg/day)        | -                              | SD                            | 2.1                                | Extracranial (including)             |
| 20          | Ceritinib Extracranial        | No mutation (effusion) non-SM                            | Alectinib (600 mg/day)        | -                              | SD                            | 3.0                                | Extracranial (including)             |

†Stereotactic radiotherapy was performed for new brain metastases, and crizotinib or alectinib was continued beyond PD. ALK, anaplastic lymphoma kinase; TKI, tyrosine kinase inhibitor; SM, sensitive mutation; RECIST 1.1, response evaluation criteria in solid tumors 1.1; PR, partial response; SD, stable disease; PD, progressive disease; PFS, progression free survival; CNS, central nervous system; LN, lymph node. Ceritinib was administered between meals.
lorlatinib, the median PFS of lorlatinib was not reached (95% confidence interval [CI]: 12.5 months–not reached), while among those treated with noncrizotinib ALK-TKIs as pretreatment of lorlatinib, the median PFS of lorlatinib was 5.5 months (95% C.I., 2.7–9.0 months). 11 In the current study, nine cases were treated only with crizotinib before rebiopsy, while 11 cases were treated with ALK-TKIs including noncrizotinib ALK-TKIs before the rebiopsy. This might be the one reason for the short PFS in the current study. In addition, in the phase II study, lorlatinib was administered in all cases, whereas in the current study, the most frequently administered tailored ALK-TKI was ceritinib, and most cases with pretreatment of ALK-TKIs received alectinib. This might also explain the relatively short PFS in the current study.

Regarding CNS disease, five of the 20 cases after rebiopsy showed progression in the CNS during next-line therapy. Of those, one case showed progression in the CNS only, and the other four showed progression in both the CNS as well as extracranial regions. Therefore, progression in the CNS might be a source of bias that affected the PFS in the current study. We propose to perform detailed analyses to clarify the efficacy of tailored ALK-TKIs against CNS lesions that progress on alectinib in a larger population.

Next generation ALK TKIs, such as alectinib and brigatinib, showed superior efficacy in the primary treatment of ALK-positive NSCLC compared with crizotinib. 10,11 However, as shown in Figure 1, the PFS of patients who received next-generation ALK-TKIs tailored to secondary mutations is relatively short, and the PFS of patients receiving next-generation ALK-TKIs in first-line treatment is expected to be long. Patients with alectinib-refractory ALK-positive NSCLC who received brigatinib also showed relatively short PFS, even though the cases had an SM to brigatinib. 12 Using better and new-generation ALK-TKIs on a priority basis might improve the prognosis of patients with ALK-positive NSCLC. Yoda et al. also mentioned that the emergence of secondary resistance mutations could be prevented with upfront treatment using the third-generation inhibitor lorlatinib. 25 The results of an ongoing randomized phase 3 trial comparing lorlatinib to crizotinib as first-line therapy for advanced ALK-positive lung cancer (NCT03052608) might partly support this hypothesis.

The present study had several noteworthy limitations. It was a retrospective analysis performed in a single institution. The retrospective nature of the study might have induced a selection bias, and the duration of follow-up was also limited. Furthermore, the number of patients who received repeated biopsy was small. Thus, additional prospective analyses of ALK-positive patients are needed to confirm these findings. In addition, resistance mechanisms are sometimes heterogeneous between lesions in a single patient, and the resistance mechanisms were analyzed in the rebiopsy lesions only; however, it is not realistic to perform a rebiopsy for all lesions. With regard to the difference in the OS between patients with and without SMs, the year of the diagnosis may have influenced survival because of differences in the ALK-TKIs that were available at that time; however, ALK-TKIs sensitive to the secondary mutation based on the rebiopsy results were selected, and the efficacy of ALK-TKIs tailored to secondary mutations targeted for analysis in the current study.

## Conclusion

The selection of ALK-TKI based on secondary SM was associated with a high ORR and relatively short PFS. Using better and new generation ALK-TKIs on a priority basis could improve the prognosis of ALK-positive NSCLC patients, and the mechanism responsible for the short PFS of sensitive ALK-TKI to secondary mutation should be clarified. Larger-scale and well-controlled prospective studies should be performed to confirm these observations.

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## Disclosure

All authors had full access to the data in the study and take responsibility for its integrity and the accuracy of the data analysis.

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