The anaplastic lymphoma kinase (ALK) gene encodes a receptor tyrosine kinase, and many kinds of ALK fusion genes have been found in a variety of carcinomas. There is almost no detectable expression of ALK in adults. However, through ALK gene rearrangement, the resultant ALK fusion protein is aberrantly overexpressed and dimerized through the oligomerization domains, such as the coiled-coil domain, in the fusion partner that induces abnormal constitutive activation of ALK tyrosine kinase. This results in dysregulated cell proliferation. ALK gene rearrangement has been observed in 3%-5% of non-small-cell lung cancers, and multiple ALK inhibitors have been developed for the treatment of ALK-positive lung cancer. Among those inhibitors, in Japan, 3 (4 in the USA) ALK tyrosine kinase inhibitors (TKIs) have been approved and are currently used in clinics. All of the currently approved ALK-TKIs have been shown to induce marked tumor regression in ALK-rearranged non-small-cell lung cancer; however, tumors inevitably relapse because of acquired resistance within a few years. This review focuses on ALK-TKIs, their resistance mechanisms, and the potential therapeutic strategies to overcome resistance.

1 INTRODUCTION

The ALK gene encodes a single transmembrane tyrosine kinase, which is widely conserved from Caenorhabditis elegans to Homo sapiens. ALK was first identified as the fusion gene nucleophosmin (NPM) 1-ALK in ALCL in 1994. After the discovery of ALK, its function was investigated, but its detailed characteristics were largely unknown until recently. In 2007, a novel ALK fusion gene, EML4-ALK, was discovered in lung cancers as a strong driver oncogene. Subsequently, research on ALK in the context of cancer and the development of ALK inhibitors has received increased attention. In addition to the ALK fusion gene, ALK point mutation-mediated constitutive activation of ALK has been discovered in neuroblastoma and thyroid cancers. To date, a number of ALK fusion oncogenes and ALK point mutations have been discovered in various types of cancer. Because aberrant constitutive activation of ALK tyrosine kinase induces dysregulated cell growth, which results in tumor development, many ALK-TKIs have been developed and tested in clinical trials, mainly for the treatment of ALK-rearranged NSCLC. Subsequently, three ALK-TKIs have been approved in Japan (4 in the USA) and used clinically, and several ALK inhibitors are under development or clinical evaluation. The ALK-TKIs have often shown marked tumor shrinkage in ALK-rearranged cancer patients and induce prolonged clinical responses; however, the tumors inevitably relapse because of acquired resistance mediated by multiple mechanisms. To overcome this resistance, it is important to identify the
molecular mechanisms underlying resistance and develop effective therapeutic strategies corresponding to each resistance mechanism. This review focuses on the molecular mechanisms of acquired resistance to ALK-TKIs in ALK-rearranged NSCLC and discusses the potential therapeutic strategies for overcoming resistance.

2 | CHARACTERISTICS OF ALK

2.1 | Function of ALK in normal cells and cancer cells

ALK was first discovered as a fusion oncogene, NPM1-ALK in ALCL, which is an aggressive CD30+ T-cell lymphoma.1 Mathew et al7 mapped the mouse homolog of ALK to chromosome 17 and confirmed that human ALK was located on the short arm of chromosome 2. Anaplastic lymphoma kinase shows greatest sequence similarity to the insulin receptor subfamily of kinases and encodes a single-pass transmembrane receptor tyrosine kinase. Pleiotrophin, midkine, and heparin have been reported as potential ligands of ALK for inducing its activation.8-10 However, as different results have been reported by other groups, the actual ligand remains to be identified. Finally, FAM150A and FAM150B were discovered by 2 different groups in 2015 and reported as genuine ligands of ALK.11,12 Expression of ALK protein in human adults has been shown only in a few cell types in the CNS and a few organs. Additionally, ALK has been shown to be expressed in the thalamus, hypothalamus, midbrain, and dorsal root ganglion in a mouse model during development.13,14 However, ALK knockout mice develop without obvious defects and have shown no differences in life spans and only mild behavioral phenotypes.15,16

Several types of ALK gene rearrangements (ALK fusion genes) have been found in many types of cancers. More than 10 years after the first identification of the ALK fusion gene, NPM-ALK, in ALCL, the EML4-ALK fusion oncogene was found in NSCLC. Until then, it had been believed that fusion oncogene-driven cancer was relatively rare in solid tumors. However, 3%-5% of NSCLC tumors have the ALK fusion gene. ALK rearrangement has been found in many types of cancer, and various fusion partners have been identified. The common characteristics of ALK fusion genes are that: (i) the ALK fusion protein is expressed constitutively through the active promoter of the fusion partner; and (ii) the fusion partner protein harbors the conserved oligomerization domain, such as the coiled-coil domain, which enables constitutive activation of ALK through dimerization or oligomerization. ALK-rearranged cancers are highly susceptible to aberrant growth signaling exerted from the constitutively activated ALK tyrosine kinase. Thus, inhibition exerted by the ALK-TKI markedly inhibits the tumor growth of ALK-rearranged cancer cells. In addition, transgenic mice expressing EML4-ALK in the lung under a surfactant protein C promoter, which is exclusively expressed in type II alveolar epithelial cells, developed hundreds of adenocarcinoma nodules in both lungs within a few weeks of birth.17 An EML4-ALK lung cancer mouse model has also been easily generated by inhalation of adenoviruses of the Crispr-Cas9 system targeting the intron of EML4 and ALK.18

In NB, which is a secondary frequent childhood cancer, various point mutations in the ALK kinase domain have been identified. Most of those point mutations have been shown to cause constitutive activation of ALK tyrosine kinase. Bresler et al19 examined the genetic abnormalities of >1500 NB patients by NGS and found that 8% of NB patients had point mutations in the ALK kinase domain, such as F1174L (the phenylalanine residue at 1174 changes to leucine) or R1275Q.

2.2 | Detection of ALK-positive cancer

To detect ALK-positive ALCL, IHC analysis using anti-ALK antibodies (such as clone SA4), which recognize the tyrosine kinase domain of ALK, has been used. The expression of the ALK fusion protein is much lower in ALK-rearranged lung cancer than in ALK-positive ALCL. Initially, ALK-rearranged lung cancer was mainly screened for by using FISH, which can detect ALK gene split by using 2 different fluorescent probes set on both sides of the break point of the ALK gene. In addition, multiplex RT-PCR-based screening has also been used to detect known ALK fusion genes. Major ALK fusion genes include EML4-ALK variant 1 (E13:A20; fusion at exon 13 of EML4 and exon 20 of ALK), variant 3 (E6:A20), and variant 2 (E20:A20), but a number of fusion partners and patterns have been found, which makes it difficult to set the multiplex PCR primer to not miss rare ALK fusion genes. After the establishment of highly sensitive IHC staining methods of ALK,20 IHC and FISH are now widely used to screen ALK-rearranged NSCLC. Currently, NGS can be used to detect ALK fusion genes by sequencing the intron between exons 19 and 20 because most ALK gene rearrangements occur in this intron of ALK, with rare exceptions.21,22

2.3 | Drugs for targeting ALK

A number of ALK-TKIs have been developed and evaluated in clinical trials; three ALK-TKIs, crizotinib, alectinib, and ceritinib, have been approved in many countries, including the USA, the EU, and Japan, and brigatinib has been approved in the USA. The following ALK inhibitors are clinically available or currently under clinical evaluation.

2.3.1 | Crizotinib

Initially, crizotinib was developed as a cMET receptor TKI. However, soon after the discovery of ALK fusion gene-positive NSCLC, a phase I clinical trial of crizotinib for treatment of ALK-rearranged NSCLC was started, in August 2008, because crizotinib can potently inhibit ALK in addition to cMET.23 In the phase I clinical trial, 149 ALK-rearranged NSCLC patients were screened by ALK split FISH and treated with crizotinib. The ORR and PFS were reported to be 60.8% and 9.7 months, respectively.24 Similar ORR and PFS have been observed in the subsequent phase II clinical trial.25 Based on the results of phase I and II clinical trials, the US FDA granted accelerated approval to crizotinib for the treatment of ALK-rearranged
NSCLC in 2011, and crizotinib was also approved in Japan in 2012. In the phase III clinical trials, the PFS was longer and ORR was higher for crizotinib than for chemotherapy. Crizotinib has been recommended as a first-line treatment of ALK-rearranged NSCLC. Although crizotinib often shows marked tumor shrinkage in ALK-rearranged NSCLC patients, crizotinib reportedly has limited potential against tumors metastasized to the brain because of poor penetration into the CSF. Indeed, disease progression has often been seen in the brain. One report showed that the crizotinib concentration in the CSF was 0.26% of that in the plasma. A study using ABC transporter ABCB1 (abcb1a and abcb1b) knockout mice reported that the crizotinib concentration in the CSF was low, mainly because crizotinib was pumped out with ABCB1. In addition, crizotinib is highly active against ROS1, which has a high degree of homology with ALK at the ATP binding site. The ALK gene has been observed in approximately 1% of NSCLC cancer patients.

### 2.3.2 Alectinib

Alectinib was developed as a selective ALK inhibitor, and the IC$_{50}$ of ALK has been shown to be 2 nmol/L, which is lower than that of crizotinib. In addition, alectinib has been shown to be active against several crizotinib-resistant mutations, such as L1196M or G1269A mutations, in ALK in vitro and in vivo xenograft models. A phase I/II clinical trial of alectinib against ALK-TKI treatment-naive ALK-rearranged NSCLC patients was carried out in Japan. The recommended dose of alectinib was determined to be 300 mg twice daily. The ORR was quite high (93.5%), and the PFS was reported to be over 3 years. Another phase I/II clinical trial of alectinib for ALK-rearranged NSCLC patients who showed progression after crizotinib treatment was undertaken mainly in the USA. The results showed that the recommended dose was 600 mg twice a day, which is double that decided on after the trial in Japan. Even in patients with crizotinib treatment history, an ORR of 52% and a PFS of 8.2 months were observed. As alectinib is not a substrate of ABCB1 and undergoes relatively little pumping out through the blood brain barrier, high CSF penetration was reported. Accordingly, alectinib showed activity in brain metastases. Two phase III clinical trials of alectinib compared with crizotinib in treatment-naive ALK-rearranged NSCLC patients were carried out in Japan (J-ALEX) and globally (ALEX). In these trials, the recommended dose of alectinib was 300 mg twice daily in J-ALEX, and 600 mg twice daily in ALEX. The PFS was longer for alectinib than for crizotinib (25.7 months for alectinib vs 10.4 months for crizotinib in J-ALEX, and 25.7 months for alectinib vs 11.1 months for crizotinib in ALEX).

### 2.3.3 Ceritinib

Ceritinib has a very low IC$_{50}$ of 150 pM to ALK in vitro; that is, 20- to 30-fold lower than that of crizotinib. Similar to alectinib, ceritinib is also active against several crizotinib-resistant mutations, such as L1196M and G1269A in ALK. Phase I clinical trials for ALK-rearranged NSCLC patients with and without prior crizotinib treatment showed an ORR of 58% and a PFS of 7.0 months. It was reported that the response was seen even in CNS metastasis cases. A phase III clinical trial comparing ceritinib with chemotherapy in advanced ALK-rearranged lung cancer with (ASCEND-5) or without prior crizotinib and chemotherapy (ASCEND-4) showed the superiority of ceritinib over chemotherapy. The median PFS in the ASCEND-4 trial was 16.6 months for ceritinib vs 8.1 months for chemotherapy, and even in patients previously treated with both chemotherapy and crizotinib, the median PFS was 5.4 months for ceritinib and 1.6 months for chemotherapy. In addition, ceritinib treatment has been shown to be affected by low-fat meals: it was shown that a lower dose of 450 mg once a day with a low-fat meal gave similar exposure and a more favorable gastrointestinal safety profile than 750 mg once a day in the fasting state in ALK-rearranged NSCLC (ASCEND-8). Moreover, ceritinib has been shown to be active against ROS1-rearranged NSCLC patients.

### 2.3.4 Brigatinib

Brigatinib was first reported as a selective ALK-TKI with a sub-nanomolar IC$_{50}$ against ALK in vitro and was also shown to be active against crizotinib-resistant L1196M gatekeeper mutation. In the phase I/II clinical trial of brigatinib, 71 crizotinib refractory ALK-rearranged lung cancer patients were treated with brigatinib, and the ORR was 62% and the PFS was 13 months. Serious treatment-emergent adverse events were observed in some patients, and 6% of the patients died during treatment or within 31 days of the last dose of brigatinib. Thus, severe adverse events occurred, so the phase II recommended dose was set at 90 mg once daily and 180 mg once daily with a 7-day lead-in at 90 mg. In the follow-up phase II study, the ORR was 45% in a group treated with 90 mg brigatinib once daily and 54% in patients treated with 180 mg once daily with a 7-day lead-in at 90 mg, and the PFS periods for the treatment groups were 9.2 and 12.9 months, respectively. Severe (grade $\geq 3$) treatment-emergent adverse events were seen in 3% of the patients, and those pulmonary toxicities tended to be associated with older patients within shorter intervals (<7 days) between the last crizotinib and the first brigatinib. No such severe pulmonary adverse events were seen after dose escalation to 180 mg after a 7-day lead-in at 90 mg.

Brigatinib has been shown to be active against almost all of the crizotinib-, alectinib-, or ceritinib-resistant mutations, including the G1202R mutation in ALK, which is highly resistant to crizotinib, alectinib, or ceritinib. However, from an analysis of a few brigatinib-resistant cases, G1202R point mutations were also seen. In 2017, brigatinib was approved in the USA for the treatment of ALK-rearranged NSCLC patients with crizotinib-refractory or -intolerable disease. Of note, it was reported that brigatinib has some activity against ROS1-rearranged cancer, and EGFR-C797S/T790M activating mutation mediated osimertinib resistance by combining with anti-EGFR antibody.
2.3.5 Lorlatinib

Lorlatinib is the first macrocyclic ALK/ROS1-TKI designed from crizotinib. Lorlatinib has been shown to be active against almost all ALK mutants, including the G1202R mutant, and resistant to first- and second-generation ALK-TKIs in vitro and in vivo. In addition, lorlatinib effectively penetrates into the CSF because it is not a substrate of p-glycoprotein. In the experimental model, lorlatinib induced marked tumor shrinkage in an intracranially injected brain metastasized model. The results of a phase I study showed that the objective response rate was 46% for 41 patients, and the recommended dose for the phase II study was 100 mg once daily. The analysis of the CSF concentration in the clinical evaluation showed a ratio of lorlatinib in CSF to serum of >0.6, and a response was observed in intracranial metastasized tumor. Currently, a phase I/II study is ongoing, and a phase III study comparing lorlatinib with crizotinib has been started.

2.3.6 Other ALK inhibitors

To date, several ALK-TKIs have been developed and are currently being evaluated in multiple clinical trials. Entrectinib is a potent inhibitor of neurotrophic receptor tyrosine kinases (NTRKs), ROS1, and ALK and is being evaluated in a clinical trial. From the results of a phase I/II study, entrectinib gave a response rate of 57% for ALK-rearranged tumors and 85% for ROS1-rearranged cancers. Central nervous system metastasized tumors also showed responses to entrectinib in the trial. Ensatinib (X-396) is an orally available small molecule ALK-TKI that potently inhibits multiple crizotinib-resistant mutants. In a phase I/II clinical trial of ensartinib, the response rate was 55% in ALK-positive lung cancer patients. Additionally, intracranial activity was observed in all of the patients with brain metastasis. Phase III clinical trials are currently ongoing. In addition, other ALK inhibitors, ASP-3026, TSR-011, CEP-37440, and PLB1003 have been developed or are currently under clinical evaluation.

3 MECHANISMS OF ALK-TKI RESISTANCE

3.1 Mutation or gene amplification of ALK-mediated resistance

In patients with ALK-rearranged lung cancer, when crizotinib was used as the initial treatment of ALK-TKI, >60% of the patients experienced partial response or complete response, and the PFS of crizotinib was approximately 10 months, which suggests that recurrence due to acquired resistance occurs within 1 year in >50% of cases. In 2010, ALK-L1196M (the ALK-1196th amino acid leucine is converted to methionine) and C1156Y mutations were found in a patient refractory to crizotinib by analyzing the cells in pleural effusion; those ALK mutants conferred resistance to crizotinib. Subsequently, various mutations, such as G1269A, I1151T-ins, G1202R, S1206Y, and I1171T, have been reported as crizotinib-resistant mutations in addition to the above two, and the L1196M gatekeeper mutation, which is in the equivalent position of the EGFR-TKI-resistant gatekeeper mutation EGFR-T790M, has been most frequently observed in crizotinib-resistant patients. However, the resistant mutation patterns in Japanese patients and their population frequency have not been reported.

Similarly, mutations resistant to ceritinib and alectinib, second-generation ALK-TKIs, and the next-generation ALK inhibitors brigatinib and lorlatinib have also been reported during the analysis of clinical samples resistant to each TKI. Alectinib and Ceritinib have previously been approved for crizotinib-refractory or -intolerability patients, and they have been recently approved for treatment-naive ALK-positive lung cancer patients. G1202R, F1174C/L/V, and G1213S mutations have been found to be ceritinib-resistant in an analysis of patients treated with ceritinib after crizotinib. G1202R, V1180L, and I1171T/N/S mutations have been reported as alectinib-resistant mechanisms. Among these secondary mutations, the G1202R mutation is resistant to all three of these ALK inhibitors, but the F1174C/L/V ceritinib-resistant mutations are sensitive to alectinib; additionally, the I1171T/N/S and V1180L mutations, which confer alectinib resistance, are sensitive to ceritinib. In contrast, brigatinib and lorlatinib have also been shown to be effective for various ALK-TKI-resistant mutations, including L1196M (Figure 1A). However, it has been shown that at least 2 mutations occur within the ALK kinase region, which makes them resistant. Regarding brigatinib, E1210K + S1206C, and E1210K + D1203N mutation has been found in patients with brigatinib resistance. For lorlatinib, the C1156Y + L1198F mutation in ALK has been found to confer resistance in lorlatinib-resistant patients. Interestingly, an L1198F double mutation has also been shown to be resensitized to crizotinib as the phenylalanine substitution at L1198 does not clash with crizotinib and in fact moves slightly closer to the inhibitor (Figures 1B and 2).

3.2 Bypass pathway or other mechanism-mediated resistance

In the crizotinib-resistant cases in which the resistance mechanisms were examined, secondary resistance mutation or ALK fusion gene amplification was observed in only one-third of the cases. From the analysis of the remaining cases and the cell line models, it has been reported that amplification of the cKIT gene with increased expression of stem cell factor (SCF) as its ligand, activation of EGFR was found to be the mechanism of resistance to crizotinib (Figure 3). The concomitant mutation of KRAS or EGFR has also been reported, but with low frequency. Activation of IGF1R has been found in crizotinib-resistant cases. Furthermore, in studies using cultured cell lines and cell lines derived from resistant patients, it has been reported that resistance due to activation of Src is observed relatively frequently, but the detailed molecular mechanism underlying activation of Src is not yet clear. As crizotinib is a potent inhibitor of cMET with an even lower IC50 than that for ALK, and was originally developed as a cMET inhibitor, cMET overactivation, such as MET gene amplification-mediated resistance, has not been observed.
A ceritinib-resistant mechanism other than secondary mutation is involved in an activating mutation of MEK (MAP2K1-K57N), and an MEK inhibitor with ALK-TKI has been found to overcome the resistance. Analysis of cell lines derived from ceritinib-resistant patients has shown that overexpression of p-glycoprotein ABCB1, which is one of the ABC transporters that transport hydrophobic substrates from cells or across the blood brain barrier to protect the brain, conferred resistance to ceritinib and crizotinib but not to alectinib or lorlatinib. Thus, the resistance can be overcome by alectinib or lorlatinib, which have not been shown to be substrates of p-glycoprotein, or by the combination of a p-glycoprotein inhibitor, such as MS20969 or verapamil. Of note, ceritinib has been shown to actively inhibit IGF1R, so IGF1R activation-mediated resistance, found in crizotinib-refractory cases, might not be observed.

An alectinib-resistant mechanism other than secondary mutation has been shown in cMET activation through MET gene amplification or elevation of its ligand hepatocyte growth factor (HGF) from an analysis of patients’ samples and cell line models. cMET activation-mediated resistance would be able to be overcome by crizotinib. Basically, cMET overactivation can potentially confer resistance to any ALK-TKIs except for crizotinib (Figure 3).

To overcome resistance-mediated bypass pathway activation, combination therapy to inhibit both ALK and the bypass pathways are considered to be necessary. However, no combination therapy has been clinically approved that overcomes bypass pathway-mediated resistance. Thus, further preclinical studies and clinical evaluations are needed to test combination therapies based on resistance mechanisms. In addition, feedback re-activation through the MAPK pathway by decrease of dual specificity phosphatase 6 (DUSP6), a phosphatase that negatively regulates the MAPK pathway, or other mechanisms contribute to the resistance to ALK inhibitors. Indeed, an up-front ALK inhibitor with MEK inhibitor treatment enhanced tumor shrinkage and prolonged initial response in an H3122 tumor-bearing mouse model. Currently, up-front inhibitors of both ALK and MAPK signaling pathways are being evaluated in several clinical trials. In addition, other combination therapies that inhibit distinct pathways, such as ceritinib + CDK4/6 inhibitor, ceritinib + mTOR inhibitor, and alectinib + anti-angiogenesis inhibitor, have been found to be activated or related to ALK-TKI resistance in cancer in vivo or in vitro experiments and have been evaluated in clinical trials.
In addition, EMT-mediated resistance has also been found in patients’ specimens and in cell line models. Gainor et al examined a series of ALK-rearranged lung cancer patients and examined cellular EMT status by IHC of vimentin and E-cadherin. They found that 42% of patients’ specimens harbored EMT characteristics. Interestingly, approximately half of the EMT-positive specimens also had secondary mutations in the ALK kinase domain; thus, EMT might not be the sole driver of the resistance. In contrast, histological transformation from adenocarcinoma to SCLC has been reported in several ALK-TKI-refractory cases, such as alectinib-resistant cases. Similar to the SCLC transformation, which is well-known in EGFR-TKI-resistant cases, classic cytotoxic chemotherapy is thought to be effective for overcoming resistance.73,74

3.3 | Challenges for the future

Currently, multiple ALK-TKIs (three in Japan and 4 in the USA) have been approved and are used clinically. In Japan, all three ALK-TKIs have been approved as first-line therapies for ALK-rearranged NSCLC. From the direct comparison between alectinib and crizotinib as first-line therapies, alectinib has been shown to have significantly longer PFS than crizotinib. There are no published data available for the direct comparison of the PFS periods of ceritinib and crizotinib or alectinib, but there are data comparing them with that of chemotherapy. In that study, compared with the chemotherapy-treated group’s PFS of 8.1 months, the PFS of ceritinib was 16.6 months, which is longer than that of first-line crizotinib but shorter than that of first-line alectinib. Both ceritinib and alectinib showed significant tumor shrinkage after crizotinib failure; however, it is believed that crizotinib has limited efficacy after alectinib or ceritinib because most crizotinib-resistant mutations are sensitive to alectinib or ceritinib, but not vice versa. In addition, brigatinib or next-generation ALK-TKIs have been shown to be effective for almost all single crizotinib, ceritinib, or alectinib-resistant mutations. Therefore, in the near future, it would be important to select the best sequencing therapy on the basis of resistance mechanisms. As the number of ALK-rearranged lung cancer patients is relatively small, each drug has a relatively long PFS period, and a number of ALK-TKIs have been developed recently. Currently, the median overall survival of ALK-TKI-treated patients is >4 years.75 Therefore, it is almost impossible to decide the order of ALK-TKIs based on overall survival. Thus, it remains very difficult to decide which drug should be used first.

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Approximately one-third of ALK-TKI-treated patients acquire resistance to TKIs through bypass pathway activation. Presently, however, there is no clear clinical evidence of the effectiveness of ALK-TKI combination therapy with an inhibitor that targets bypass pathways, such as cMET or cKIT. Thus, further studies are needed to identify bypass pathways and new effective therapeutic strategies to overcome the resistance. In addition, tumor heterogeneity is a difficult hurdle that must be overcome. Multiple resistance mechanisms, such as multiple mutations, are often observed in 1 patient. Moreover, recently collected data from NGS analysis of circulating tumor DNA samples has shown dynamic changes in therapy-persistent/resistant cancers. Thus, further studies are needed in the following areas:

- Elucidation of unidentified resistance mechanisms in approximately 25% of resistant cases.

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**FIGURE 3** Bypass pathway activation-mediated anaplastic lymphoma kinase-tyrosine kinase inhibitor (ALK-TKI) resistance (stromal cell-mediated ligand secretion from tumor- or stromal cell-mediated receptor tyrosine kinase activation). Left panel, cKIT gene amplification with stem cell factor (SCF) (a ligand of cKIT) secretion from stromal cell-mediated crizotinib resistance. Middle panel, hepatocyte growth factor (HGF) upregulation led to cMET activation-mediated resistance. HGF was secreted from tumor or stromal cells. Otherwise, cMET gene amplification solely induced the activation of cMET, which resulted in ALK-TKI resistance. Right panel, ERBB receptor, including epidermal growth factor receptor (EGFR) activation-mediated resistance. Various ligands for the ERBB family can induce activation of ERBB receptors and induce resistance. CAF, cancer-associated fibroblast; EML4, echinoderm microtubule-associated protein-like 4; NRG, neuregulin; TGF-A, transforming growth factor-α.
Resistance mechanisms and fusion variants (are G1202R mutations only seen in EML4-ALK-variant 3 patients?).

Sensitive detection methods to identify the resistance mechanisms from blood (and tissues).

Heterogeneity and dominance of the populations with resistant tumors.

Drugs to overcome compound-resistant mutations to lorlatinib.

Up-front combination therapy to effectively eradicate persistent cancer cells.

Understanding the immune checkpoint in ALK-rearranged lung cancer.

In addition, it is also important to fully understand the mechanisms and functions of ALK tyrosine kinase itself and the downstream signaling of ALK, including the substrates of ALK fusion proteins. A recent study showed that some drug-resistant cells have drug-dependent growth characteristics, called "drug addiction," instead of oncogene addiction. In the drug-addicted-resistant cells, TKI removal induced hyperactivation of oncogene signaling, which resulted in induction of apoptosis. Moreover, intermittent kinase inhibitor treatment has been shown to prolong survival in vivo. Indeed, intermittent kinase inhibitor treatment strategies have been shown to prolong survival in vivo. Therefore, further studies are needed to develop better therapies.

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CONFLICTS OF INTEREST

The author has no conflict of interest.

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