R E V I E W

Developments in anaplastic large-cell lymphoma: targeting the anaplastic lymphoma kinase

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Abstract: Anaplastic lymphoma kinase (ALK) is a receptor tyrosine kinase protein implicated in a variety of hematological malignancies and solid tumors. ALK contributes to the development of cancers in different cell lineages through a variety of genetic mechanisms: gene fusions, activating point mutations, and possibly gene amplification. Recent developments led to significant therapeutic advances, including efficient diagnostic tests and ALK-targeting agents. This review addresses some therapeutic considerations with regard to the use of ALK inhibitors in ALK-positive lymphomas where, in spite of the advanced stage of the disease, long-lasting responses could be obtained in a substantial portion of heavily pretreated patients. Data and mechanisms for the development of resistance to ALK inhibitors will also be presented and discussed.

Keywords: ALK, lymphoma, tyrosine kinase, targeted therapy, crizotinib

Introduction

Anaplastic lymphoma kinase (ALK) is a tyrosine kinase (TK) receptor that has been initially identified through its involvement in chromosomal translocations associated with anaplastic large-cell lymphoma (ALCL). De novo systemic ALK-positive ALCLs are clinically aggressive lymphomas and frequently occur within the first 3 decades of life with a male predominance. ALCLs usually present as stage III–IV disease, with B symptoms, high levels of lactate dehydrogenases, and extranodal involvement.[1–4] ALK-positive ALCLs often have a good clinical response to cytotoxic drugs, but relapses can occur, determining a worse prognosis.[2,5–7]

In addition, the use of cytotoxic therapy bears the risk for long-term toxicities, such as second cancers, lung fibrosis, or cardiac failure, which represent important issues, given the young age of most patients, who usually belong to the teenager/young adult groups.

ALK was identified for the first time in 1994 as a result of its fusion to nucleophosmin 1 (NPM1), which represents the predominant partner in ALCL.[8,9] The NPM1-ALK results from the t(2;5)(p23;q25) chromosomal translocation, present in 50%–70% of ALCLs. NPM1-ALK is deregulated and transforms cells in vitro, activating the signal in several transduction pathways.[10–16]

It has been demonstrated, by using small interfering RNA (siRNA) to specifically downregulate the expression of the NPM1-ALK in ALCL cell lines, that the downregulation of NPM1-ALK resulted in decreased cell proliferation and increased cell apoptosis. All these data point toward a causal relationship between the development of ALK-containing fusions and malignant transformation, thus rendering ALK a potential therapeutic target for both pharmacologic and immunologic intervention.

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In fact, ALK expression is physiologically restricted to the immune privileged nervous system.[15,17]

Industrial interest in ALK's role in cancer and the therapeutic potential of its specific inhibition has increased since 2007 with the discovery of echinoderm microtubule associated protein 4 (EML4) ALK in non-small-cell lung cancer (NSCLC). Subsequently, other tumors, such as inflammatory myofibroblastic tumors, neuroblastomas, and some carcinomas, both in adults and children, were shown to be driven by an altered ALK signal.[18] Recently, several ALK inhibitors have been developed, offering new treatment options in tumors driven by abnormal ALK signaling.[19–22]

In 2011, crizotinib, a small-molecule inhibitor of the receptor tyrosine kinases of the hepatocyte growth factor (c-Met) and ALK, was approved by the Food and Drug Administration in ALK-positive NSCLC. Dramatic activity of crizotinib in ALK-positive ALCL was also demonstrated.[23–25] New and more potent ALK inhibitors are likely to follow shortly. These molecules represent another excellent proof of principle for targeted therapy.[26] As has been observed with other tyrosine kinase inhibitors, resistance has also recently emerged in patients treated with ALK inhibitors.[27–31]

ALK biology

Most of the knowledge of ALK expression in cancer comes from its pathological expression in NSCLCs and neuroblastomas that are more common in adults or children than ALCL or ALK-positive diffuse large-B-cell lymphoma (DLBCL). Nevertheless, what we know now is still useful for ALK-positive lymphomas and will help scientists and clinicians in approaching this disease.

ALK is normally expressed only in the nervous system (thalamus, hypothalamus, midbrain, olfactory bulb, selected cranial, dorsal root, and ganglial cells). It has a role in neural development and differentiation.[32–34] ALK is involved in oncogenesis in both nonhematopoietic and hematopoietic malignancies. The full-length form of ALK is expressed in different types of cancers, including glioblastoma,[32,35] breast cancer, neuroblastoma,[36] Ewing sarcoma,[37] retinoblastoma,[38] DLBCL,[39] and melanoma.[40] Except for neuroblastoma, the pathogenic role of ALK is not clear in these tumors.[41]

A variety of mechanisms leading to aberrant kinase activation and constitutive phosphorylation of downstream pathway components have been identified, including missense mutation, gene amplification, and chromosomal translocation. ALK pathological expression in lymphoma cells is a result of chromosomal translocations that lead to the formation of ALK-containing oncogenic fusion proteins[42] also found in several tumors, as mentioned earlier. The fusions interrupt the chromosome at the level of the *ALK* gene at 2p23. The chromosomal breakpoint involves, in the case of NPM1-ALK fusion, intron 4 of NPM1 and intron 16 of ALK; as result, the N-terminal region of NPM1 is fused to the catalytic domain of ALK, which conserves its protein-kinase domain.[9] The fusion partner at the N terminus is usually widely expressed in normal cells and controls chimeric protein expression and localization.[43] The ALK partner brings an oligomerization domain that mediates constitutive self-association of the ALK fusion, causing constitutive activation of its kinase domain and controlling its expression levels.[41,44] At this time, almost 15 different ALK fusion proteins have been identified (Table 1).[45] As a result, ALK fusion proteins

Table 1 Chromosomal rearrangements involving the *ALK* gene and producing oncogenic fusion proteins in anaplastic large-cell lymphomas and other cancers

Gene	Translocation	Frequency in ALK+ anaplastic large-cell lymphomas	Localization	Reference
NPM	t(2;5)(p23,q35)	78%	Nuclear/cytoplasmic	112–114
ATIC	Inv(2)(p23,q35)	2%	Cytoplasmic	115,116
ALO17	t(2;17)(p23;q25)	1%	Cytoplasmic	117,118
CARS	t(2;11)(p23;p15)	Unknown	Unknown	117,119
CTLC	t(2;17)(p23;q23)	2%	Cytoplasmic	120,121
MYH-9	t(2;22)(p23;q11.2)	1%	Cytoplasmic	118,121
MSN	t(2,X)(p23;q1)	1%	Membrane of the cell	122,123
TFG	t(2;3)(p23;q21)	1%	Cytoplasmic	124,125
TPM3	t(1,2)(q25,p23)	18%	Cytoplasmic	126
TPM4	t(2,19)(p23;p13)	1%	Cytoplasmic	127
RANBP2	t(2,2)(p23;q11)	Unknown	Nuclear membrane	128
SEC31L1	t(2;4)(p23;q21)	Unknown	Cytoplasmic	129
EML4	Inv(2)(p21,p23)	Unknown	Cytoplasmic	130,131
KIF5B	t(2;10)(p23;p11)	Unknown	Cytoplasmic	132
SQSTM1	t(2;5)(p23.1;q35.3)	Unknown	Granules cytoplasmic	133

Abbreviation: ALK, anaplastic lymphoma kinase.

are deregulated, ectopically expressed, and constitutively activated in lymphoid and other types of neoplastic cells. In lymphomas, this leads to growth factor-independent proliferation of lymphocytes,[46–51] the biological target cells of the lymphoma-associated oncogene. ALK fusion plays a key role in neoplastic transformation by altering the phosphorylation of intracellular substrates, which likely contributes to the molecular pathogenesis of ALK-positive ALCL.[15,52] NPM1-ALK is the most common fusion protein in ALK-positive ALCL, whereas clathrin heavy polypeptide (CLTC) ALK is present in ALK-positive DLBCL.[53]

The NPM1 portion mediates the constitutive dimerization and consequent activation of ALK kinase domain and controls its expression levels.[44] NPM1-ALK is able to activate proliferative pathways such as rat sarcoma protein (RAS)/ mitogen activated protein kinase (MAPK),[46] Phospholipase C gamma (PLCγ).[54] Janus kinase (JAK)/ signal transducer and activator of transcription (STAT),[51,55,56] and phosphoinositide 3 kinase (PI3K)/ protein kinase B (Akt).[10,12,57] Data point to the functional link between hematopoietic oncogenic tyrosine kinases and the G(1) cell cycle regulator *Cell division cycle 25 homolog A* (*CDC25A*).[58,59] NPM1-ALK can protect cells from apoptosis mediated by this pathway or by inducing the transcription of antiapoptotic genes, including *Bcl-2* and *Bcl-xL*.[60–62] Animal models showed that the oncogenic role of NPM1-ALK induced a lymphoproliferative disorder in a short period of time.[49,63] The tumorigenicity of NPM1-ALK was first demonstrated in a study by Kuefer et al[50] in which retrovirally transfected bone marrow expressing NPM1-ALK was transplanted into lethally irradiated BALB/cByJ mice. These mice developed B-cell lymphomas within 4–6 months, clearly linking aberrant ALK activation with tumorigenesis. Chiarle et al generated transgenic mice in which NPM1-ALK expression was targeted to T cells, using a cluster of differentiation 4 (CD4) transgene cassette.[49] These mice developed thymic lymphomas and plasma cell neoplasms from 5 weeks of age.

However, the presence of different ALK fusions confers different sensitivity to ALK inhibitors; for example, in vitro NPM1-ALK-expressing cells exhibit a higher response with lower ALK half maximal inhibitory concentration (IC50) values than SEC31 homolog A (SEC31A)-ALK-expressing cells.[64]

ALK is also a frequent object of mutation. Gain-of-function point mutations are critical to activate the kinase domain and may be the cause of secondary drug resistance. This was found first in neuroblastoma,[65] where mutations affect the ALK intracellular segment that is linked to regulatory and catalytic signals. About 10% of sporadic neuroblastoma have somatic nonsynonymous ALK mutations, including K1062M, F1174L/C/I,

F1245C/V/L, and R1275Q amino acid substitutions.[66] In this setting, there is a significant correlation between activating mutations in the ALK TK domain and poor clinical outcome.[67] ALK mutations are also associated with familial neuroblastoma (eg, T1087I, G1128A, and R1275Q).[68–70]

It is not clear how multiple copies of the ALK gene can contribute to tumor pathogenesis, even though rare case reports with amplification of aberrant forms of ALK are documented.[71,72] Frequently, *ALK* amplification co-occurs with amplification of *v-myc avian myelocytomatosis viral oncogene neuroblastoma derived homolog* (*MYCN*).[73] In particular, *ALK* amplifications without any mutations or gene fusions may not have a strong role in tumor pathogenesis, and thus may not represent a good target for ALK inhibitors.

Different diagnostic methods are available to detect ALK rearrangement. Fluorescence in situ hybridization (FISH), reverse transcriptase-polymerase chain reaction (RT-PCR), chromogen in situ hybridization, and immunohistochemistry (ICH) are currently used.[74] FISH is normally used in trials to test ALK fusions, but it does not discriminate between different fusion types. RT-PCR can be used in a rapid and extremely sensitive way, but only when a defined ALK partner is known and novel fusions may be missed. ICH is cheap, but interpretation may be difficult. It can be used as screening in ALK-positive NSCLC, which has a lower incidence than ALK-positive ALCL.[75–77] In a large case series of NSCLCs, a comparison of the sensitivity and specificity among ICH, FISH, and RT-PCR was performed. The study revealed that FISH has the best specificity and ICH the best sensitivity.[78]

ALK testing can be useful not only during diagnosis but also during the monitoring of treatment; in this setting, RT-PCR is the methodology of choice to detect early resistance, which develops with different frequencies in different diseases.

ALK inhibitors and their role in ALCL

ALK-positive ALCLs are currently treated with combined chemotherapy, with an overall response rate (ORR) of 60%–70%.[20] Nevertheless, ALK is a good candidate for the development of targeted treatment because of the lack of wide expression in normal adult tissues. For this and the reasons mentioned earlier, blocking ALK function should not give important toxic effects.[16,28,79] Potential strategies for targeting ALK include immunotherapy, gene silencing, inhibition of downstream signaling pathways, and direct inhibition of its catalytic activity through small-molecule inhibitors. The aim of this target therapy is to obtain a maximum tumor-specific effect with low toxicity, contrary to the conventional citotoxic chemotherapy.[80]

After the identification of different constitutively activated forms of ALK, both as fusion proteins and as mutated ALK forms, the attention focused on the development of small molecules targeting the protein kinase activity to abolish the ALK-dependent cancer cell growth.[45] Kinase inhibitors are directed against the adenosine triphosphate (ATP)-binding site of the catalytic domain, which is highly conserved in ALK TK, with the goal of obtaining successful ALK inhibition. In preclinical studies, several ALK inhibitors have shown activity against NPM1-ALK and EML4-ALK cell lines.[81,82] Initial testing of ALK inhibitors was performed using inhibitors derived from natural products such as staurosporine derivatives, which are not specific inhibitors of ALK.[83] Synergies with heat shock protein 90 inhibitors were also observed.[84] These and other natural-derived compounds inhibit ALK, increasing the proteosome-mediated degradation of ALK protein binding to heat shock protein 90.[85,86] Subsequently, more-potent and more-specific ALK inhibitors have been developed, including at least 18 different classes of small molecule inhibitors of ALK (Table 2). ALK inhibitors have shown activity not only in ALCL but also in ALK-positive DLBCL, inducing tumor growth delay and its regression in murine xenograft.[64,87]

The largest volume of available knowledge relates to crizotinib. Crizotinib (PF-02341066) is the first human ALK inhibitor developed. It is a derivative of aminopyridine and was originally developed as a potent, orally bioavailable, ATP-competitive small-molecule inhibitor of mesenchymal epithelial transition growth factor/hepatocyte growth factor receptor/c-MET.[88] Crizotinib suppresses the proliferation of ALK-positive ALCLs cell lines.[81] This orally available TK inhibitor was being tested in an open-label, multicenter, two-part, dose escalation phase I clinical trial as an MET inhibitor to investigate its safety, tolerability, pharmacokinetics, pharmacodynamics, and activity in 37 patients with advanced cancer (excluding leukemias).[89] The first part of this trial established 250 mg twice daily as the maximum tolerated dose. Three dose-limiting toxicities were observed: a grade 3 increase in alanine transaminase (ALT) (one patient at 200 mg once a day) and grade 3 fatigue (two patients at 300 mg twice a day). The most common adverse events were ocular flashes, nausea, emesis, fatigue, and diarrhea, all of which were manageable and reversible.[90]

The first treatment of an ALK-positive ALCL patient occurred in 2010. A report published in the *New England Journal of Medicine* described two adult patients with recurrent ALK-positive ALCLs who achieved complete response (CR) shortly after receiving crizotinib as a single agent.[23]

In June 2012, the authors updated their experience at the European Hematology Association, reporting on additional patients who achieved complete remission or partial response (PR), demonstrating clearly the activity of crizotinib in ALK-positive ALCLs. ORR was 10/11 (91%) and included 9 CR (82%) and 1 PR, with an ORR of 91% (95% confidence interval, 60%–99%); 7 patients had been in CR for more than 22 months, with a 2 year progression-free survival of 64%. All relapses developed within the initial 3 months of treatment.[24] No parameter could predict the development of a durable response, with the only indication being that patients who were treated with crizotinib after failing an autologous bone marrow transplantation never achieved a durable response, at difference with patients treated after failing an allogeneic bone marrow transplantation. The final results of this study were recently published.[25] These results were recently confirmed in a sponsored trial in which an ORR of 64% among 14 ALK-positive lymphoma patients is reported.[91] A recent phase I study also reported this good safety profile with high response rates in children with relapsed ALK-positive ALCL.[92]

Some experimental evidence suggests that ALK inhibitors could also be efficacious in the treatment of ALK-positive DLBCL.[64,87] The compassionate study previously cited[24,25] reported 2 patients with ALK-positive DLBCL treated with crizotinib with one rapid but transient response.

As has been seen with other targeted therapies, resistance can emerge in some patients who have demonstrated initial response to ALK inhibition, as happened in the studies mentioned earlier, at quite a rapid pace.

Resistance to ALK inhibitors

Acquired resistance to kinase inhibitors is a serious problem in long-term cancer therapy. Data on ALK inhibitors resistance are, for the majority, reported in the context of NSCLCs. The mechanism can be divided into three groups, very similar to what we know about chronic myeloid leukemia (CML). The first one is an ALK mutation (28%), second is resistance resulting from mechanisms that upregulate ALK (such as gene amplification or copy number gain, which occur in 18% of patients), and last is the activation of other transduction pathways; for example, KRAS or mast/stem cell growth factor receptor (KIT) amplification occurs in 30% of cases.[93–95]

The first mechanism was initially reported in 2010 in NSCLCs,[96] and two point mutations in the kinase domain of EML4-ALK were found.

These mutations, C1156Y and L1196M, conferred clinical resistance to crizotinib in NSCLC patients, and

Table 2 Inhibitors of anaplastic lymphoma kinase in development[26,139,147–149]

Name	Therapeutics	Responses	Trials in NHLs	Clinical trials ongoing, references and notes
PF-02341066 (crizotinib; Pfizer, Inc., New York, NY, USA)	Selective ATP competitive oral inhibitor of ALK and c-MET tyrosine kinases and their variants	Overall response rate 91% with 9/11 complete response[25]	Yes	Phase I–II done;[90] ongoing phase III; studies with crizotinib as single agent or in combination
AP-26113 (Ariad, Cambridge, MA, USA)	Dual ALK/epidermal growth factor receptor inhibitor AP-26113 is 10-fold more potent compared with PF-02341066	67% overall response rate with 2/15 complete response in non-small-cell lung cancer[134]	Yes	Well-tolerated; phase I/II[134]
ASP 3026 (Astellas Pharma, Northbrook, IL, USA)	Dual ALK/epidermal growth factor receptor inhibitor		Yes	Phase I: promising safety and tolerability[135]
LDK 378 (Novartis, Basel, Switzerland)	Selective ALK inhibitor	Durable responses in the majority of patients with advanced, ALK+ non-small-cell lung cancer, including crizotinib-resistant patients with and without crizotinib resistance mutations;[107] overall response rate, 70%[136]	Yes	Phase I: safety in ALK positive/genetically abnormal tumors; preliminary results[136,137]
NPV-TAE684 (TAE-684) (Novartis, genomics institut of the Novartis research foundation)	Selective ALK inhibitor; good oral bioavailability in vivo (60%–70%)		No	Phase I discontinued[137]
CEP-28122 (Cephalon, Frazer, PA, USA)	Selective ALK inhibitor			Preclinical[138]
F91873 and F91874 (Istitut de rechercher Pierre Fabre)	ATP noncompetitive inhibitors, pyridoisoquinoline derivatives			Preclinical
PHA-E429 (Nerviano Medical Science)	ATP competitive, crystal structure of the ALK kinase domain in a complex with PHA-E429			Preclinical/phase I[139]
CEP-14083 and CEP-14513 (Cephalon)	Selective ALK inhibitor		No	Phase I
NMS-E628 (Ariad)	ALK inhibitor		No	Phase I[140]
WZ-5-12 (Ambit Biosciences)	ALK small molecules inhibitor			Preclinical[141]
CRL151104 (Chebridge St. Jude)	ATP competitor			Preclinical[41]
CH5424802 (Chugai Pharmaceutical, Tokyo, Japan; Roche, Basel, Switzerland)	Potent and selective orally available ALK inhibitor[142,143]	Overall response rate 93% with 2/46 complete response and durable treatment[144]	No	Mild toxicity; phase I/II trial in non-small-cell lung cancer patients in Japan[144]
X396 (XCovery, West Palm Beach, FL, USA)		Pharmacokinetic properties and toxicity profiles are favorable		Phase I[145]
GSK1838705A (GlaxoSmithKline, Brentford, United Kingdom)	Small-molecule kinase inhibitor of insulin-like growth factor; receptor. Abrogates ALK, and growth of anaplastic large cell lymphoma			Preclinical[146]

Abbreviations: ATP, adenosine triphosphate; ALK, anaplastic lymphoma kinase; NHLs, non Hodgkin lymphomas.

another one, F1174L, causes resistance to the same drug in an inflammatory myofibroblastic tumor patient.[96,97] The acquired resistance to ALK-targeted therapy has been distinguished in "ALK-dominant" and "ALK-nondominant" mechanisms. In the first case, the acquired resistance is a result of the development of novel ALK kinase domain mutation alone or in combination with the increase of rearranged ALK gene copies in cancer cells. These are called ALK-dominant mechanisms because ALK signaling remains dominant in the crizotinib-resistant state. Multiple ALK kinase domain mutations that reduce sensitivity to crizotinib were recently identified.[98] In vitro, a single amino acid substitution became

predominant at higher crizotinib concentrations in two ALCL cell cultures grown at increasing crizotinib concentrations. The authors found that L1196Q substitution conferred resistance to crizotinib, but not to AP26113 and NVP-TAE 694, whereas cells carrying I1171N mutation were resistant to all tested inhibitors.[27] In the clinical report described earlier,[24] the kinase domain of NPM1-ALK could be amplified from peripheral blood samples obtained at the time of relapse in two ALCL patients. Deep sequencing of these products revealed the presence of different mutations: Q1064R at high prevalence (95%) in one patient and I1171N (33%) plus M1328I (14%) in a second patient. All these mutations were not present in samples obtained before crizotinib treatment, which did not contain either these or any other mutation in the ALK kinase domain. I1171N was already discovered in an in vitro screening[17] and commands an intermediate level of resistance to crizotinib (resistance index (RI), 5.8), which, however, is cross-resistant with other anti-ALK TKI such as AP26113 and NVP-TAE684.

In addition, a series of different second oncogenic drivers were also described, coexisting in the same cell with the ALK rearrangement. These cases are ALK-nondominant mechanisms.[99–101] These events should, in theory, bypass the dominance of ALK signaling and replace its oncogenic potential.[27] To overcome resistance, it will be important to differentiate patients who preserve ALK dominance versus those who have diminished ALK dominance.

Future developments

The complexity of mechanisms of acquired resistance recently described suggests that other therapeutic options, including combination of ALK and other targeted approaches, such as immunotherapy,[102,103] will be required in the future.[104] ALK can be a target of antitumor vaccination because ALK-positive cells are dependent on ALK activity for survival and proliferation and ALK is spontaneously immunogenic in ALCL with an antibody and a T-cytotoxic response.[105] In particular, as we learned in preclinical tests in neuroblastoma cell lines, the combination of ALK inhibitors with ALK antibody may work synergistically.[106] Thus, we hypothesize that a combination approach with chemotherapy or vaccination and ALK inhibitors can reduce the burden of the disease obtaining durable responses.[102]

In addition, substantial effort has been focused on optimizing direct kinase inhibition by developing new ALK inhibitors that are structurally different from crizotinib. LDK378 (Novartis Inc., B Basel, Switzerland), CH5424802 (Chugai Pharmaceutical, Tokyo, Japan), AP26113 (Ariad

Pharmaceuticals, Cambridge, MA, USA), and X396 (XCovery, West Palm Beach, FL, USA) are some examples, with many others also in development (Table 2). The experience with these drugs is, for now, limited only to ALK-positive NSCLCs. The first phase I trial in humans of LDK378 (NCT01283516), limited to relapsed/refractory NSCLC patients, demonstrated objective responses in patients with ALK-rearranged NSCLC who were previously treated with crizotinib.[107,108] A phase I/II trial of CH5424802 in crizotinib-naïve patients with ALK-positive NSCLC demonstrated objective responses in 43 of 46 patients enrolled at the maximum tolerated dose, with two CRs and 41 PRs.[109] AP26113, a dual ALK and epidermal growth factor receptor inhibitor, also showed evidence of clinical activity in crizotinib-naïve and crizotinib-resistant NSCLC, with an ORR of 73%.[110]

Conclusion

The initial identification of the genetic lesion at the basis of malignant transformation in ALK-positive ALCLs, originally obtained in 1994,[9] was successfully exploited and brought to the patient bedside in 2010.[23] This compares favorably with the time that elapsed between the discovery of the Philadelphia chromosome in CML and the clinical development of imatinib.[111]

However, it is sad to recognize that ALK-positive ALCL patients could be treated earlier and that ALK inhibitors became available only because ALK alterations were identified in more common cancers such as NSCLC.

The entry of crizotinib in the treatment of ALK-positive ALCLs marked a new era in the therapy of this rare but highly malignant type of tumor. The demonstration of high response rates and durable responses, even in the setting of advanced and resistant disease, should prompt the development of clinical studies in less advanced conditions and in combination with already-active drugs. A particular emphasis should be placed on the need to decrease as much as possible the use of cytotoxic drugs, given their long-term toxic effects and the young age of most of the patients. Drugs of interest could be steroids, vincristine, and brentuximab, given their lack of cross-resistance with crizotinib.

The possibility of obtaining durable responses in a substantial fraction of advanced, heavily pretreated ALK-positive lymphomas illustrates our present inability to forecast the level of heterogeneity present inside a tumor. The type of presentation and clinical history of these patients would suggest they should be placed in the category of highly heterogeneous diseases, such as a metastatic NSCLC, relapsed blast

crisis CML, or relapsed Philadelphia chromosome-positive acute lymphoblastic leukemia, in which monotherapy with TKIs seldom obtains durable responses. Further studies, such as exome sequencing of pre- and posttreatment samples, could hopefully shed light and provide a useful indicator of the level of heterogeneity present inside a tumor at any given time.

The next couple of years will hopefully see the fading of regimens based only on unspecific cytotoxic drugs in favor of more specific and hopefully less toxic approaches.

Disclosure

The authors declare no conflicts of interest in this work.

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