Medical Journal of Bakırköy

Research

Alectinib-induced Non-immune Hemolytic Anemia due to Erythrocyte Membrane Alterations: A Retrospective Evaluation

Alektinib İlişkili Eritrosit Membran Değişikliklerine Bağlı İmmün Olmayan Hemolitik Anemi: Retrospektif Bir Değerlendirme

Emine Gültürk[1], Eyyüp Akkaya[1], Korhan Kapucu[1], Seher Yıldız Tacar[2], Goncagül Akdağ[3], Merve Kahraman Diril[4], Fehmi Hindilerden[1]

[1]University of Health Sciences Türkiye, Bakırköy Dr. Sadi Konuk Training and Research Hospital, Clinic of Hematology, İstanbul, Türkiye
[2]University of Health Sciences Türkiye, Mehmet Akif Ersoy Thoracic and Cardiovascular Surgery Training and Research Hospital, Clinic of Medical Oncology, İstanbul, Türkiye
[3]Kartal Dr. Lütfi Kırdar City Hospital, Clinic of Medical Oncology, İstanbul, Türkiye
[4]İstanbul Esenler Gynecology and Children's Diseases Hospital, Clinic of Internal Medicine, İstanbul, Türkiye

ABSTRACT

Objective: The purpose of this study was to evaluate changes in erythrocyte morphology associated with alectinib and resulting hemolytic anemia.

Methods: This was a retrospective analysis of patients with stage IV non-small-cell lung cancer (NSCLC) treated with alectinib. Erythrocyte morphology (by peripheral-blood film evaluation), hemogram, reticulocyte, direct Coombs tests, serum lactate dehydrogenase (LDH), haptoglobin, and indirect bilirubin levels were evaluated. Demographic characteristics of the patients were also collected.

Results: In total, 13 patients (7 women) with a mean age of 52.0±10.5 years were included. Median pre-alectinib hemoglobin level was 12.1 g/dL [minimum (min): 9.4, maximum (max): 16.2 g/dL]. In total, serum hemoglobin was decreased in 8 patients (61.5%) compared with pre-alectinib levels. The average serum hemoglobin level after alectinib use was determined as 11.6 g/dL (min: 8.5 g/dL, max: 13 g/dL). Serum hemoglobin was <10 g/dL in only 3 patients. *De novo* anemia developed in six patients. Peripheral blood examination revealed numerous microsphero-acanthocytes, some echinocyte, rare fragmented erythrocytes, and generalized anisopoikilocytosis. Serum LDH levels were high in 6 of 13 patients (46.1%) receiving alectinib. Reticulocyte count was high in 10 of 13 patients (76.9%). A decrease in serum haptoglobin was observed in five patients (38.4%). Serum indirect bilirubin was high in two of the patients (15.3%).

Conclusion: Alectinib caused changes in the erythrocyte membrane and non-immune hemolysis in almost all patients using it. Hemolytic anemia was not severe enough to require alectinib dose reduction or discontinuation. Physicians caring for patients receiving alectinib should be alert to hematological changes due to drug use.

Keywords: Alectinib, non-immune hemolytic anemia, erythrocyte membrane changes

ÖZ

Amaç: Çalışmanın amacı, alektinib ile ilişkili eritrosit morfolojisindeki değişiklikleri ve bunun sonucunda ortaya çıkan hemolitik anemiyi değerlendirmekti.

Gereç ve Yöntem: Alektinib ile tedavi edilen evre IV küçük hücreli olmayan akciğer kanseri (KHDAK) hastalarının retrospektif bir analizidir. Eritrosit morfolojisi (periferik kan filmi değerlendirmesi ile), hemogram, retikülosit sayısı, direkt Coombs testleri, serum laktat dehidrogenaz (LDH), haptoglobin, indirekt bilirubin değerlendirildi. Hastaların demografik özellikleri de toplandı.

Bulgular: Toplamda yaş ortalaması 52,0±10,5 yıl olan 13 hasta (7 kadın) dahil edildi. Medyan pre-alektinib hemoglobini 12,1 g/dL [minimum (min): 9,4, maksimum (maks): 16,2 g/dL] idi. Toplamda serum hemoglobini, alektinib öncesi hemoglobin düzeylerine kıyasla 8 hastada (%61,5) azaldı. Alektinib kullanımı sonrası ortalama serum hemoglobin değeri 11,6 g/dL (min: 8,5 g/dL, maks: 13 g/dL) olarak belirlendi. Sadece 3 hastada serum

ÖZ

hemoglobin değeri <10 g/dL idi. Altı hastada de novo anemisi gelişti. Periferik kan incelemesinde tüm hastalarda çok sayıda mikrosfero-akantosit, bir miktar ekinosit, nadir parçalanmış eritrositler ve jeneralize anizopoikilositoz görüldü. Alektinib tedavisi alan 13 hastanın 6'sında (%46,1) LDH yüksekti. On üç hastanın 10'unda (%76,9) retikülosit yüksekti. Beş hastada (%38,4) serum haptoglobin düzeyinde azalma gözlendi. Hastaların 2'sinde (%15,3) serum indirekt bilirubin yüksekti.

Sonuç: Alektinib kullanan hastaların neredeyse tamamında eritrosit membranında değişikliklere ve immün olmayan hemolizlere neden oldu. Hemolitik anemi, alektinib dozunun azaltılmasını veya kesilmesini gerektirecek kadar ciddi değildi. Alektinib tedavisi gören hastaların tedavisiyle ilgilenen hekimler, ilaç kullanımına bağlı hematolojik değişiklikler konusunda dikkatli olmalıdır.

Anahtar Kelimeler: Alektinib, immün olmayan hemolitik anemi, eritrosit zarı değişiklikleri

INTRODUCTION

Lung cancer is responsible for most cancer-related deaths worldwide (1). Among several types of cancer, non-small-cell lung cancer (NSCLC) constitutes more than 80% of all cases. The most commonly encountered subtypes of NSCLC are adenocarcinoma and squamous carcinoma. Compared with 1975, the 5-year survival rate in lung cancer increased by 9% to 20.5% in 2016 (2). Several factors account for this improvement in prognosis, the most important of which is the advent of targeted therapies and immunotherapy. Recent years have witnessed the discovery and elucidation of several genomic aberrations that are crucial in NSCLC pathogenesis. Almost 30% of patients with NSCLC have one or more of these mutations and rearrangements (3). A number of targeted therapies targeting these mutations enable improved survival, particularly in patients with advanced disease.

Anaplastic lymphoma kinase (ALK) rearrangements have been reported in up to 4.5% of patients with NSCLC (4). First discovered in 2007 (5), an inversion in chromosome 2p produces a fusion gene comprising the *echinoderm microtubule-associated protein-like 4 (EML4)* gene and the *ALK* gene. This fusion gene, in turn, activates the tyrosine kinase domain of the *ALK* gene, resulting in the proliferation of lung epithelial cells (6). The first targeted drug developed to address this important tumorigenic pathway was crizotinib, an ALK tyrosine kinase inhibitor (7). Owing to the observed beneficial effects, crizotinib was recommended as a first-line treatment agent in patients with ALK-rearranged NSCLC. However, with the advent of second- and third-generation ALK tyrosine kinase inhibitors with superior survival rates, better central nervous system penetration, and more acceptable adverse effect profiles, alectinib and ceritinib are now considered as first-line treatment in these patients (8,9).

In a network meta-analysis, low-dose alectinib emerged as having the lowest adverse event risk among all ALK inhibitors (10). This does not mean that alectinib is not without adverse events (11). In the ALEX trial, adverse effects related to alectinib included constipation, liver function abnormalities, anemia (in 26% of the drug users), peripheral edema, myalgia, rash, and bradycardia. Most reported anemia cases were mild, and the median time to anemia development was 3.9 months (12). However, there were no data regarding the nature of alectinib-related anemia. After the publication of the ALEX trial (12), several case reports and series (13-15) reported a type of non-immune hemolytic anemia in patients with NSCLC treated with alectinib. These reports pointed to the development of erythrocyte membrane changes as the culprit triggering extravascular hemolysis in the spleen. Compared with the common use of alectinib, studies characterizing alectinib-related anemia are scarce. Thus, we aimed to report our experience with alectinib-induced non-immune hemolytic anemia.

METHODS

Patients and Setting

This is a retrospective analysis of patients with NSCLC who were treated with alectinib and developed nonimmune hemolytic anemia. The oncology departments of the University of Health Sciences Türkiye, Bakırköy Dr. Sadi Konuk Training and Research Hospital and Kartal Dr. Lütfi Kırdar City Hospital participated in the study. The University of Health Sciences Türkiye, Bakırköy Dr. Sadi Konuk Training and Research Hospital Clinical Research Ethics Committee approved the study protocol (decision no: 2022-17-01, date: 05.09.2022). We retrospectively screened NSCLC patients with *ALK* gene rearrangements and treated them with alectinib from the electronic hospital database and patient charts between January 2016 and April 2022. All NSCLC patients treated with alectinib were included in the study. Deceased patients at the time of screening and patients with missing data were excluded from the study.

Data Collection

Clinicodemographic characteristics such as age, sex, presence of comorbid conditions, ECOG performance status, disease stage during inclusion in the study, institution of curative-intent chemoradiotherapy, histologic subtype of NSCLC, percentage of ALK, and presence of organ metastases before alectinib treatment. We also performed

laboratory tests (serum hemoglobin concentration, white blood cell count, platelet count) before alectinib treatment. Hemoglobin levels were measured after alectinib treatment. In addition, we performed hemolysis tests and other tests to better elucidate alectinib-related anemia. These tests involved serum hemoglobin concentration, serum vitamin B12, folic acid, and ferritin levels, serum lactate dehydrogenase (LDH), total and direct bilirubin, haptoglobin levels, direct and indirect Coombs tests, reticulocyte count, peripheral smear, and paroxysmal nocturnal hemoglobinuria panel. Response to alectinib therapy was classified as complete or partial response, stable disease, and progression. We also collected data regarding adverse events (AE) related to alectinib use. Common Terminology Criteria for Adverse Events v3.0 was used to grade observed AE as follows: Grade 1, Mild AE; grade 2, Moderate AE; grade 3, Severe AE; grade 4, life-threatening or disabling AE; and grade 5, death related to AE (16).

Statistical Analysis

Categorical variables are reported as numbers and percentages (%). Continuous variables are given as mean ± standard deviation or median [minimum (min) - maximum (max)] depending on the distribution of the variable. The Wilcoxon signed-rank test was used to evaluate the statistical significance of pre- and post- alectinib serum LDH values, and the paired samples t-test was used for comparing serum hemoglobin values. Statistical analyses were performed by processing the data of the patients with the SPSS v22 (IBM, New York) program.

RESULTS

General Characteristics

We obtained data of 13 patients via screening of the hospital database and records of oncology departments. None of the patients were excluded from the study. Overall, 13 patients (7 females) with a mean age of 52.0±10.5 years were included in the study. All patients had stage 4 NSCLC with an adenocarcinoma subtype. All patients had one or more distant organ metastases. The most common organ metastasis was to the bone (9/13), followed by the brain (5/13). Six of 13 patients were administered cytotoxic chemotherapy before the institution of alectinib. In all cases, the chemotherapy regimen involved a carboplatin + taxane combination. Three patients showed a complete response to alectinib therapy, eight patients had a partial response, and one patient had stable disease under alectinib therapy. The mean duration of alectinib use was 26.3±15.7 months. Eight of 13 patients (57.1%) developed at least one AE other

than anemia related to alectinib use. All observed AEs were grade 1 or 2. No dose reduction or drug discontinuation was not needed because of AEs. Only one patient (patint number 14) required red blood cell (RBC) transfusion because of anemia during her disease. All patients were alive during data collection for this study. Table 1 shows the demographic and clinical characteristics of the study patients.

Anemia Parameters

Before alectinib treatment, 9 of 13 patients already had anemia. The median pre-alectinib hemoglobin level was 12.1 g/dL (min: 9.4, max: 16.2 g/dL). In total, 8 patients (61.5%) had reduced serum hemoglobin levels compared with pre-alectinib hemoglobin levels. The mean serum hemoglobin level after alectinib use was 11.6 g/dL (min: 8.5 g/dL, max: 13 g/dL). Only three patients had serum hemoglobin values below 10 g/dL. *De novo* anemia developed in 6 of 13 patients (patients nos 2, 4, 6, 8, 10, and 13). There was no statistically significant change in the mean hemoglobin values of patients before and after alectinib use (p=0.091, the paired samples t-test). Table 2 depicts the laboratory values of the patients.

In all patients, except patient 8, peripheral blood film demonstrated numerous microsphero-acanthocytes, some echinocytes, rare fragmented erythrocytes, and general anisopoikilocytosis (Figure 1). Patient number 8 showed no erythrocyte membrane changes but had an iron deficiency anemia morphology. However, her serum ferritin level was within the normal range. In patient 10, the peripheral smear showed normochromic normocytic erythrocytes with rare acanthocytes.

Other Laboratory Studies

Serum LDH levels were elevated before alectinib treatment in 3 of 11 patients (27.2%). With alectinib treatment, 6 of 13 patients (46.1%) had elevated LDH levels. Median serum LDH levels before and during alectinib use were 210 U/L and 203 U/L, respectively (p=0.110, the Wilcoxon signed-rank test). Ten of 13 patients (76.9%) had elevated reticulocyte counts. Direct and indirect Coombs tests were negative in all patients. At the time of evaluation, serum haptoglobin levels were reduced in only 5 patients (38.4%). Serum indirect bilirubin levels were elevated in only 2 of the patients (15.3%).

DISCUSSION

The most salient findings of the present study were as follows: (i) Alectinib led to non-immune hemolytic anemia in all patients who used it. (ii) None of the patients required

Table 1. Demographic and some clinical characteristics of the patients

Patient no	Sex	Age (years)	Comorbid condition	Organ metastasis	Pre-alectinib chemotherapy	Response to alectinib	Adverse events other than anemia	Specific adverse events	Duration of alectinib use (months)
1	Female	47	-	Bone	Yes	Partial	Yes	Increased liver transaminases	29.3
2	Female	61	-	Brain	Yes	Partial	No	-	19.5
3	Male	65	-	Bone	No	Complete	Yes	Nausea	21.8
4	Male	71	Hypertension	Pleura	Yes	Stable disease	Yes	Anemia and increased creatinine	51.7
5	Female	46	Breast cancer	Brain, bone, and pleura	No	Partial	Yes	Leg edema and anemia	40.0
6	Male	52	-	Brain, bone	Yes	Partial	Yes	Anemia, hyperbilirubinemia	28.0
7	Male	52	Hypertension Diabetes mellitus	Bone, pleura	No	-	No	-	0.8
8	Female	61	-	Bone, pleura	No	Partial	Yes	Leg edema, myalgia, and anemia	33.5
9	Male	59	-	Bone, adrenal gland, pleura	No	Partial	Yes	Nausea, Diarrheae	33.5
10	Male	44	-	Bone	Yes	Partial	No	-	39.0
11	Male	52	-	Pleura	No	Partial	No	-	5.2
12	Female	31	-	Brain, adrenal gland, pleura	Yes	Complete	No	-	2.3
13	Female	48	-	Liver	No	Complete	Yes	Nausea, increased liver transaminases, myalgia, and anemia	37.2

Table 2. Laboratory values of patients before and during alectinib use

Patient no	Serum hemoglobin (g/dL)		LDH (U/L)		Reticulocyte count (%)	Haptoglobin (g/L)	Indirect bilirubin (mg/dL)
	Before alectinib	After alectinib	Before alectinib	After alectinib			
1	11.5	12.4	272	289	2.27	1	0.42
2	14.6	10.2	295	251	4.23	0.17	0.9
3	11.9	13.0	163	213	2.47	0.88	0.4
4	12.8	8.5	212	626	2.9	0.05	0.31
5	14.4	12.4	156	199	2.87	0.56	0.33
6	14.1	11.7	213	191	3.01	0.05	1.29
7	9.9	11.0	232	306	0.77	6.01	0.05
8	12.1	9.3	-	221	1.83	1.57	0.07
9	12.4	11.9	-	278	2.27	0.1	0.44
10	16.2	12.9	145	173	1.41	1.4	0.36
11	9.4	10.4	109	150	2.31	1.69	0.20
12	10.4	11.5	197	195	2.19	0.43	0.48
13	12.1	8.5	210	203	5.32	0.1	1.07

Normal reference values for haptoglobin (0.3-2.0 g/L), lactate dehydrogenase (LDH) (135-214 U/L), reticulocyte count (0.2-2%)
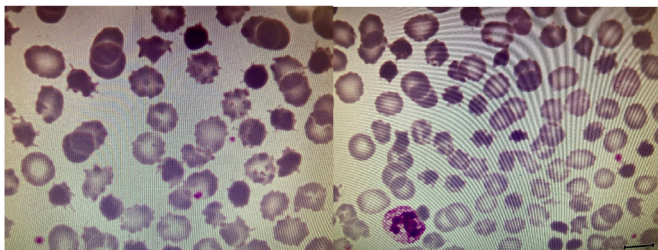
Figure 1. Peripheral blood smear samples showing numerous microsphero-acanthocytes, some echinocyte, rare fragmented erythrocytes, and general anisopoikilocytosis

alectinib dose reduction or cessation due to anemia. (iii) RBC morphological alterations were observed in all patients. (iv) Hemolytic anemia was nonimmune in all alectinib users. (v) The most prominent morphologic change in RBCs was the development of acanthocytosis.

We evaluated the presence and characteristics of anemia in patients with stage IV NSCLC. Our results showed the occurrence of anemia in all alectinib users. In contrast to the universal development of hemolytic anemia in our cohort related to alectinib use, clinical trials reported the development of anemia between 14-22% of patients in whom alectinib was used (12,17). Unfortunately, these trials did not provide details of the anemia type. One case series by Gullapalli et al. (18) reported alectinib-related hemolytic anemia in six patients, but they did not reveal among how many alectinib users they selected these six patients. Kunz et al. (15) conducted a prospective observational study in which they reported the development of reticulocytosis and abnormal erythrocyte morphology with anisocytosis in all 24 patients who were treated with alectinib within 2 weeks of its use. Our results are in agreement with those of the latter authors in that alectinib caused RBC morphologic changes and nonimmune hemolytic anemia in all treated patients.

We report the third largest case series after Kuzich et al. (13) and Kunz et al. (15), in which alectinib-related hemolytic anemia was characterized. To the best of our knowledge, 64 cases have been reported in the literature to date.

Alectinib-induced RBC membrane changes were first described by Kuzich et al. (13). The authors retrospectively evaluated 43 patients with advanced NSCLC and observed marked acanthocytosis, echinocytosis, and/ or spheroacanthocytosis in 95% of the alectinib-treated patients. However, anemia developed only in 73% of the patients and was mild to severity. Serum hemoglobin values were <10 g/dL in 38% and <8 g/dL in only 8% of the patients. Kunz et al. (15) found RBC membrane changes in all treated patients. The most commonly encountered morphological abnormality was acathopcytes, followed by echinocyte, spherocytes, dacrocytes, and fragmentocytes. Anemia was

present in only 68% of the patients treated with alectinib in Kunz et al. (15). The peripheral blood film of our patients showed widespread acanthocytosis, except in one patient. Among our patients, anemia was present in 61.5% of the patients with alectinib use. However, the *de novo* anemia rate related to alectinib use was 46%. There was no severe anemia, and only two patients had serum hemoglobin levels below 10 g/dL. None of our patients required alectinib dose reduction or discontinuation because of the development of anemia.

The exact mechanism of alectinib-induced alterations in the RBC membrane is yet to be elucidated. However, Kuzich et al. (13) and others (15) showed reduced eosin-5-maleimide staining (EMA) binding in affected patients. This finding, along with apparent morphologic changes, implies that alectinib leads to erythrocyte cytoskeletal changes. Apart from one case by Okumoto et al. (19), all reported alectinib-induced anemia cases in the literature were nonimmune and extravascular. The direct antiglobulin (Coombs) test was consistently found to be negative in reported cases. Okumoto et al. (19) reported alectinib-induced hemolytic anemia, and the Coombs test was negative. Nevertheless, the authors concluded that their case was Coombs-negative immune hemolytic anemia. We believe that the authors actually detected non-immune hemolytic anemia precipitated by alectinib-induced RBC membrane changes but erroneously labeled this as Coombs-negative immune hemolytic anemia. In our cohort, similar to the literature, all patients had negative direct and indirect Coombs tests.

In our study, reticulocyte counts were increased in 76.9% of the patients. The highest reticulocyte count was 4.23%. The results of Kuzich et al. (13) were similar to ours in terms of reticulocyte counts. It was available for seven of their patients (elevated in four), and the maximum value was 3.6%. On the other hand, in the study by Kunz et al. (15), 87.5% of the patients had elevated reticulocyte counts, and the maximum value was 8.3%. In our opinion, the discrepancy between the reticulocyte counts between our results and those by Kunz et al. (15) results from the study design difference. Kunz et al. (15) conducted a prospective study and had the opportunity to prospectively evaluate the patients at monthly intervals after alectinib initiation. Thus, they might have caught the most manifest time of alectinib-induced hemolysis. In contrast, in our evaluation, some patients had been on alectinib treatment for several months. Interestingly, reticulocyte elevation was still evident in three-fourths of our patients, and the counts were somehow smaller compared with those of patients by Kunz et al. (15).

Some limitations of this study deserve mention: First, ours was a retrospective study. Thus, we may have missed some relevant data due to the progress of RBC changes and resultant hemolysis. Second, we did not perform an EMA binding test or other investigations related to ultrastructural changes in the erythrocyte membrane. Thus, we cannot draw any conclusions as to the exact mechanism of the RBC membrane changes that were evident in the light microscopic evaluation. However, studies of some groups mentioned earlier shed some light on the underpinnings of acathocyte development with alectinib use. EMA binging was uniformly diminished, pointing to a possible effect of alectinib on the RBC membrane structure. However, we still do not know by what mechanism alectinib can alter the membrane structure. Third, we cannot determine whether this observed effect of alectinib is specific to alectinib or a class effect. Other studies in the literature have not observed changes in RBCs similar to alectinib with the use of other ALK inhibitors such as crizotinib, brigatinib, and lorlatinib (15). Lastly, we cannot determine whether alectinib-related changes in RBC membranes are temporary because all of our patients were still using their alectinib treatments at the time of the evaluation. However, other researchers have reported that RBC membrane changes were corrected upon discontinuation of alectinib use (15).

CONCLUSION

In conclusion, in this third-largest study regarding the effects of alectinib on RBCs, we showed that alectinib caused erythrocyte membrane changes and nonimmune hemolyses in almost all patients who used it. Hemolytic anemia was not severe, necessitating drug dose reduction or discontinuation. However, it is of utmost importance to know alectinib-related hematologic changes beforehand so that lengthy and expensive studies can be undertaken to investigate the cause of anemia and hemolysis in alectinib-treated patients.

ETHICS

Ethics Committee Approval: The University of Health Sciences Türkiye, Bakırköy Dr. Sadi Konuk Training and Research Hospital Clinical Research Ethics Committee approved the study protocol (decision no: 2022-17-01, date: 05.09.2022).

Informed Consent: Retrospective study.

Authorship Contributions

Surgical and Medical Practices: E.G., S.Y.T., G.A., F.H., Concept: E.G., Design: E.G., Data Collection or Processing:

E.G., E.A., K.K., S.Y.T., G.A., M.K.D., Analysis or Interpretation: E.G., Literature Search: E.G., Writing: E.G.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: The authors declared that this study received no financial support.

REFERENCES

1. Global Burden of Disease Cancer Collaboration; Fitzmaurice C, Abate D, Abbasi N, Abbastabar H, Abd-Allah F, Abdel-Rahman O, et al. Global, Regional, and National Cancer Incidence, Mortality, Years of Life Lost, Years Lived With Disability, and Disability-Adjusted Life-Years for 29 Cancer Groups, 1990 to 2017: A Systematic Analysis for the Global Burden of Disease Study. JAMA Oncol 2019;5:1749-68.

2. Thandra KC, Barsouk A, Saginala K, Aluru JS, Barsouk A. Epidemiology of lung cancer. Contemp Oncol (Pozn) 2021;25:45-52.

3. Chen J, Xu C, Lv J, Lu W, Zhang Y, Wang D, et al. Clinical characteristics and targeted therapy of different gene fusions in non-small cell lung cancer: a narrative review. Transl Lung Cancer Res 2023;12:895-908.

4. El Yacoubi H, Sow ML, Kettani F, Gamra L, Mestari A, Jabri L, et al. Frequency of anaplastic lymphoma kinase rearrangements in Moroccan patients with non small cell lung cancer: a multi-institutional national retrospective study. BMC Cancer 2020;20:479.

5. Soda M, Choi YL, Enomoto M, Takada S, Yamashita Y, Ishikawa S, et al. Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer. Nature 2007;448:561-6.

6. Chevallier M, Borgeaud M, Addeo A, Friedlaender A. Oncogenic driver mutations in non-small cell lung cancer: Past, present and future. World J Clin Oncol 2021;12:217-37.

7. Camidge DR, Bang YJ, Kwak EL, Iafrate AJ, Varella-Garcia M, Fox SB, et al. Activity and safety of crizotinib in patients with ALK-positive non-small-cell lung cancer: updated results from a phase 1 study. Lancet Oncol 2012;13:1011-9.

8. McCusker MG, Russo A, Scilla KA, Mehra R, Rolfo C. How I treat ALK-positive non-small cell lung cancer. ESMO Open 2019;4:e000524.

9. Chazan G, Solomon BJ. Optimal first-line treatment for metastatic ALK+ non-small cell lung cancer-a narrative review. Transl Lung Cancer Res 2023;12:369-78.

10. Chuang CH, Chen HL, Chang HM, Tsai YC, Wu KL, Chen IH, et al. Systematic Review and Network Meta-Analysis of Anaplastic Lymphoma Kinase (ALK) Inhibitors for Treatment-Naive ALK-Positive Lung Cancer. Cancers (Basel) 2021;13:1966.

11. Dziadziuszko R, Peters S, Ruf T, Cardona A, Guerini E, Kurtsikidze N, et al. Clinical experience and management of adverse events in patients with advanced ALK-positive non-small-cell lung cancer receiving alectinib. ESMO Open 2022;7:100612.

12. Peters S, Camidge DR, Shaw AT, Gadgeel S, Ahn JS, Kim DW, et al. Alectinib versus Crizotinib in Untreated ALK-Positive Non-Small-Cell Lung Cancer. N Engl J Med 2017;377:829-38.

13. Kuzich JA, Heynemann S, Geoghegan N, Evelyn C, O'Mahoney S, Wilson S, et al. Alectinib induces marked red cell spheroacanthocytosis in a near-ubiquitous fashion and is associated with reduced eosin-5-maleimide binding. Pathology 2021;53:608-12.

14. El Sayed R, Tehfe M, Blais N. Successful Treatment with Brigatinib after Alectinib-Induced Hemolytic Anemia in Patients with

Metastatic Lung Adenocarcinoma-A Case Series. Curr Oncol 2022;30:518-28.

15. Kunz J, Wiedemann C, Grosch H, Kriegsmann K, Gryzik S, Felden J, et al. Early Development of Ubiquitous Acanthocytosis and Extravascular Hemolysis in Lung Cancer Patients Receiving Alectinib. Cancers (Basel) 2022;14:2720.

16. Cancer Therapy Evaluation Program, Common Terminology Criteria for Adverse Events, Version 3.0, DCTD, NCI, NIH, DHHS, March 31, 2003, https://ctep.cancer.gov/protocoldevelopment/electronic_applications/docs/ctcaev3.pdf.

17. Camidge DR, Dziadziuszko R, Peters S, Mok T, Noe J, Nowicka M, et al. Updated Efficacy and Safety Data and Impact of the EML4-ALK Fusion Variant on the Efficacy of Alectinib in Untreated ALK-Positive Advanced Non-Small Cell Lung Cancer in the Global Phase III ALEX Study. J Thorac Oncol 2019;14:1233-43.

18. Gullapalli V, Xu W, Lewis CR, Anazodo A, Gerber GK. A multi-centre case series of alectinib-related erythrocyte membrane changes and associated haemolysis. J Hematop 2021;14:131–6.

19. Okumoto J, Sakamoto S, Masuda T, Yamaguchi K, Horimasu Y, Miyamoto S, et al. Alectinib-induced Immune Hemolytic Anemia in a Patient with Lung Adenocarcinoma. Intern Med 2021;60:611-15.