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Identification of ALK-positive patients with advanced NSCLC and real-world clinical experience with crizotinib in Spain (IDEALK study)

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ABSTRACT

Objectives: To determine the incidence of ALK translocations in patients with advanced/metastatic NSCLC in Spain, to describe the clinical characteristics of these patients, and to evaluate the effectiveness and safety of treatment with crizotinib in a real-world setting.
Methods: This is an observational prospective and retrospective cohort study to determine the incidence of *ALK* translocations and to analyze the effectiveness and safety of crizotinib in a real-world setting. Patient characteristics, treatment patterns, time to best overall response, duration of treatment, objective response rates (ORR), rates of adverse events (AE), progression free survival (PFS) and overall survival (OS) were evaluated in the ALK study cohort of patients treated with crizotinib (prospective and retrospective). ALK incidence and quality of life (QoL) questionnaires were measured from patients included in the prospective cohort.

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Results: The incidence of *ALK* translocations was 5.5 % (31 of 559 patients). Compared with *ALK*-negative patients, *ALK*-positive patients were significantly younger, predominantly female, and non-smokers. In the crizotinib effectiveness and safety study, 91 patients (42 prospective, 49 retrospective) with *ALK*-positive NSCLC (43.9 % in first-line, 56.1 % in second or more lines) were included. The ORR was 59.3 % and the median duration of response was 13.5 months (IQR, 5.3–26.2). The median PFS was 15.8 months (95 % CI, 11.8–22.3) and the median OS was 46.5 months, with 53 patients (58.2 %) still alive at data cut-off date. Frequently reported AEs included elevated transaminases, gastrointestinal disorders, and asthenia. Most patients (76.5 %) reported improved or stable scores for global QoL during treatment.
Conclusions: The observed incidence of *ALK* translocations in NSCLC patients is aligned with published reports. This analysis of the real-world clinical experience in Spain confirms the therapeutic benefit and safety of crizotinib in advanced/metastatic *ALK*-positive NSCLC. Clinicaltrials.gov: NCT02679170.

1. Introduction

Lung cancer is the second-most commonly diagnosed malignant tumor worldwide and the leading cause of cancer death, with a mortality rate of 18.0 % of the total cancer deaths [1]. Increased mortality among lung cancer patients is often associated with advanced stage at diagnosis, which occurs in about half of all patients [2]. An increase in lung cancer mortality in recent years has been reported in Spain, reaching 20.3 % in 2020 [3].

Approximately 85 % of all new lung cancer diagnoses can be classified as non-small-cell lung cancer (NSCLC), a heterogeneous class of tumors [2]. In the last two decades the treatment of NSCLC has been revolutionized by the emergence of molecular-driven targeted therapies such as small molecule tyrosine kinase inhibitors and immunotherapy, which has led to greatly increased survival and quality of life in selected patients. Since its identification in 2007 [4], translocations of the anaplastic lymphoma kinase (*ALK*) gene, which lead to activation of the ALK tyrosine kinase domain and oncogenesis, have been identified in approximately 3–6 % of all NSCLC cases [5–9]. Patients with NSCLC driven by *ALK* rearrangements may benefit from ALK tyrosine kinase inhibitor (TKI) therapies [10].

Crizotinib is an orally available small-molecule TKI that suppresses the activity of ALK, ROS, and MET kinases. Crizotinib was the first TKI approved by the for the treatment of *ALK*-positive NSCLC [11]. The PROFILE 1001 and PROFILE 1005 clinical trials demonstrated significant objective response rates of about 60 % and longer progression-free survival (PFS) in previously treated *ALK*-positive patients [12,13]. Later, the PROFILE 1007 randomized trial demonstrated superior efficacy of crizotinib versus pemetrexed or docetaxel as second line after platinum-based chemotherapy [14] and the PROFILE 1014 study showed superiority of crizotinib to pemetrexed plus cisplatin or carboplatin in first line [15,16]. On the basis of these studies, crizotinib achieved a second-line approval in ALK-positive advanced NSCLC in 2012 and a first-line approval in 2015 from the EMA. Since that time, several new generation ALK inhibitors have changed the first-line treatment paradigm of ALK-positive NSCLC and continuation of ALK-inhibition by using sequential strategies such as mutational resistance profiling after second-generation ALK-TKI relapse, and the role of the 3rd-generation inhibitor lorlatinib have demonstrated to be an effective strategy in these oncogene-addicted patients [17,18].

Although the need for studies of oncologic therapies in the real-world setting is clear, the low-incidence *ALK* rearrangements makes the study of this small subgroup of patients with NSCLC challenging. The difficulty in the evaluation of effectiveness of specific therapies in real world clinical practice is increased by the heterogeneity of the patients and the complexity of the therapeutic pathways followed. Several real-world studies have been published in recent years analyzing cohorts from diverse countries. However, to date no analysis has been performed on *ALK*-positive NSCLC patients from Spain. The objective of this study was to evaluate the incidence of *ALK* translocations in patients with advanced/metastatic NSCLC in Spain, as well as to describe the clinical characteristics of these patients and the effectiveness and safety of crizotinib, the first locally approved ALK inhibitor, in routine clinical practice.

2. Materials and methods

This multicentric, non-interventional, observational study analyzed patients diagnosed with advanced/metastatic NSCLC in Spain. The study consisted of two sub-studies: the *ALK* incidence sub-study and the *ALK* treatment sub-study. An amendment of the study protocol was made to allow the inclusion and retrospective analysis of the ROS1 population in treatment with crizotinib from the date of marketing authorization for this indication in Spain in February 2017. The ROS1 cohort analysis will be presented separately.

The objective of the first sub-study was to determine the incidence of *ALK* translocations in patients with advanced/metastatic NSCLC in Spain; in the second sub-study the goal was evaluate the effectiveness of treatment with crizotinib in patients with advanced/metastatic NSCLC with *ALK* translocation in terms of progression-free survival (PFS). The study was conducted from November 2016 to June 2019 in 23 hospitals countrywide in Spain. All procedures of the study were carried out within the usual clinical practice guidelines for this patient population. The protocol was approved by the Ethics Research Committee of the Hospital Clínico San Carlos (Madrid, Spain). The study included patients recruited retrospectively and prospectively. In patients who were included prospectively, signed informed consent was obtained. The study was carried out in accordance with the protocol, the standards of Good Clinical Practice, the International Conference on Harmonization and the applicable local laws and requirements. In addition, the study complied with the principles contained in the Declaration of Helsinki [19].

2.1. Patients

All patients included were adults (>18 years) with a diagnosis of NSCLC. For the sub-study of *ALK* incidence, all patients with advanced/metastatic NSCLC who were diagnosed in the participating hospitals were registered. Subsequently, those patients in whom the *ALK* translocation status was determined were included (prospective recruitment).

For the sub-study on the effectiveness and safety of treatment with crizotinib, the patients included were those with confirmed *ALK* translocation-positive advanced/metastatic NSCLC who had been (retrospective recruitment, since January 2014) or were going to be (prospective recruiting) treated with crizotinib following standard clinical practice.

2.2. Endpoints and assessments

This study had two primary endpoints: the first was to determine the incidence of *ALK* translocations in patients with advanced/metastatic NSCLC in Spain. The second was to evaluate the effectiveness of treatment (PFS) with crizotinib in patients with advanced/metastatic NSCLC

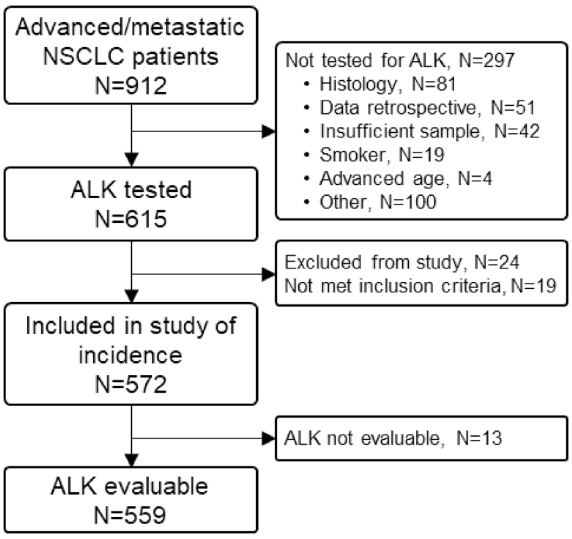

Fig. 1. Patient disposition. NSCLC, non-small cell lung cancer.

Table 1
Characteristics of patients with advanced/metastatic NSCLC.

Variable	ALK-negative (N = 528)	ALK-positive (N = 31)	Total (N = 559)
Age (years), median (IQR)	65.7 (59.3–73.2)	60.8 (45.2–68.1)	65.5 (59.0–72.9)
Sex (male), N (%)	352 (66.7)	12 (38.7)	364 (65.1)
Smoker, N (%)	208 (39.4)	6 (19.4)	214 (38.3)
Sample type, N (%)			
Biopsy	438 (83.0)	28 (90.3)	466 (83.4)
Cell block	27 (5.1)	0 (0.0)	27 (4.8)
Cytology	63 (11.9)	3 (9.7)	66 (11.8)
Histology, N (%)			
Adenocarcinoma	422 (79.9)	31 (100.0)	453 (81.0)
Squamous	71 (13.4)	0 (0.0)	71 (12.7)
Large cell	19 (3.6)	0 (0.0)	19 (3.4)
NOS	16 (3.0)	0 (0.0)	16 (2.9)
EGFR (positive)[1]	45 (8.5)	0 (0.0)	45 (8.1)
ALK diagnosis			
FISH	261 (49.4)	14 (45.2)	275 (49.2)
IHC	254 (48.1)	16 (51.6)	270 (48.3)
PCR	5 (0.9)	0 (0.0)	5 (0.9)
Other[2]	8 (1.5)	1 (3.2)	9 (1.6)
ROS1 (positive)	4 (0.8) *(N = 385)*	1 (3.2) *(N = 19)*	5 (0.9) *(N = 394)*
Other molecular alterations	104 (19.7)[3]	6 (19.4)[4]	110 (17.9)

EGFR, epidermal growth factor receptor; FISH, fluorescent in situ hybridization; IHC, immune histochemistry; IQR, interquartile range; NOS, not otherwise specified; PCR, polymerase chain reaction.

[1] Patients tested and with valid tests (N = 508 for ALK-negative; N = 27 for ALK-positive; N = 535 for Total).

[2] Includes tested by high-throughput sequencing (ThermoFisher Oncomine), CLART technology, and unspecified.

[3] Includes *KRAS*, PD-L1, *MET*, and *TP53*.

[4] All PD-L1 > 50 %.

with *ALK* translocation. As secondary endpoints, also the objective response rate (ORR), duration of response (DoR), time to best response, overall survival (OS, measured from the date of initiation of treatment with crizotinib until the date of death or last control), safety data, treatment post crizotinib, and patient-reported outcomes (PROs) –the latest only for the prospective cohort– were analyzed.

The incidence of patients with advanced/metastatic NSCLC and positive *ALK* translocation was defined as the percentage of *ALK*-positive patients out of the total number of patients with advanced/metastatic NSCLC included in the study. Information on the demographic and clinical characteristics of the patients, and the characteristics of the tumor, were also collected [20].

Radiological imaging control follow-up was performed according to standard clinical practice in Spain for both the retrospective and prospective population and in strict compliance with the Response Evaluation Criteria in Solid Tumors (RECIST) v1.1 diagnostic criteria.

The safety of crizotinib was evaluated by description of the incidence of all adverse events (AEs) and their severity graded according to NCI CTCAE version 4.0.

The quality of life of *ALK*-positive patients treated with crizotinib was measured in thirty patients included prospectively with the European Organization for Research and Treatment of Cancer (EORTC) questionnaires QLQ-C30 and QLQ-LC13, in their Spanish-validated versions [21–24]. A hundred percent of patients fulfilled both questionnaires at baseline, 77 % of them at the end of first cycle of treatment, 57 % after the third and only 17 % of those patients completed the last evaluation at the end of treatment. The results of the first 3 milestones will be assessed in this analysis.

2.3. Statistical analysis

To compare the differences in the characteristics of the population of patients according to *ALK* status, the Pearson's Chi-Square test was used (or Fisher's exact test if necessary) for qualitative variables, and the *T*-Student test, one-way ANOVA or their non-parametric equivalents U-Mann-Whitney and H-Kruskal-Wallis tests in the case of quantitative variables. To evaluate the effectiveness and safety of treatment with crizotinib, the Kaplan-Meier survival function was used to study PFS, DoR and OS. The ORR was analyzed with absolute and relative frequencies. For the comparison of survival functions, the Log-rank test was used. The incidence rate of specific adverse effects was described in the total population. To analyze the association between visits in the quality of life of the patients, the Wilcoxon test was used.

3. Results

3.1. Incidence of ALK-positive NSCLC

From 912 patients with advanced/metastatic NSCLC screened, a total of 559 patients were evaluated for *ALK* status (Fig. 1). The remaining 353 patients were not included in this analysis at the investigator's discretion for a variety of reasons, including histology not consistent with adenocarcinoma, previously performed ALK testing, scarce tissue sample, exitus, EGFR positive status, very old age, and smoking habit, among others. The demographic and clinical characteristics of the patients are shown in Table 1. Thirty-one patients (5.5 %) were *ALK*-positive.

ALK-positive patients were significantly younger than *ALK*-negative patients (p = 0.0017), and predominantly female (p = 0.0029). A higher proportion of non-smokers was found among *ALK*-positive patients (p = 0.0349). All *ALK*-positive patients (100 %) presented adenocarcinoma, and all were negative for EGFR mutations. Interestingly, of 19 *ALK*-positive patients also tested for ROS1, one was positive. PD-L1+ (PD-L1 > 50 %) was reported in 6 out of 110 patients for whom additional molecular alterations were reported, but no alterations in *KRAS*, *MET*, or *TP53* were found in *ALK*-positive patients (Table 1).

3.2. Characteristics of patients treated with crizotinib

A total of 91 patients with NSCLC with confirmed *ALK*-positive and treated with crizotinib were included in the study, 49 (53.8 %) retrospectively and 42 (46.2 %) prospectively. The baseline characteristics of these patients are shown in Table 2. The median age was 60.5 years

Table 2
Baseline characteristics of patients treated with crizotinib (N = 91).

Variable	N = 91
Age (years), median (IQR)	60.5 (44.4–67.2)
Sex (female)	58 (67.3)
Smoker	18 (19.8)
Stage at diagnosis	
IIIA	5 (5.5)
IIIB	8 (8.8)
IV	78 (85.7)
Number of metastases	
0	8 (8.8)
1–2	60 (65.9)
≥3	23 (23.0)
Histology	
Adenocarcinoma	84 (92.3)
Squamous	1 (1.1)
Large cell	3 (3.3)
NOS	3 (3.3)
Baseline ECOG[1]	
0–1	78 (96.3)
2–3	3 (3.7)
Brain metastasis	11 (12.1)
Line of treatment with crizotinib	
1st line	40 (44.0)
2nd line	35 (38.5)
3rd line	16 (17.6)

All data reported as N (%) unless otherwise indicated.
CR, complete response; ECOG, Eastern Cooperative Oncology Group; IQR, inter-quartil range; NOS, not otherwise specified; PD, progressive disease; PR, partial response; SD, stable disease.
[1] N = 81.

Table 3
Response to treatment with crizotinib (N = 91).

Variable	All patients (N = 91)
Best response with crizotinib, N (%)	
CR	8 (8.8)
PR	40 (44.0)
SD	22 (24.2)
PD	11 (12.1)
Not evaluable/unknown	10 (11.0)
ORR, N (%)	48 (52.8)
Time to best response,[1] months, median (IQR)	3.0 (2.3–5.8)
Duration of response,[2] months, median (IQR)	13.5 (5.3–26.2)
Concomitant radiotherapy, N (%)	6 (13.2)
Reason for crizotinib discontinuation, N (%)	
Progression	68 (84.8)
Adverse event	9 (11.4)
Other	3 (3.8)
Crizotinib dose reduction, N (%)	22 (24.2)
Time from start to 1st dose reduction, median (IQR), months	2.7 (1.1–9.1)
Crizotinib interruption, N (%)	33 (36.3)
Time to restart, days, median (IQR)	12.0 (7.0–20.0)

All data reported as N (%) unless otherwise indicated.
CR, complete response; IQR, interquartile range; ORR, overall response rate; PD, progressive disease; PR, partial response; SD, stable disease.
[1] N = 78.
[2] N = 32.

(IQR, 44.4–67.2; range, 22.2–85.0) and 63.7 % were female. Most patients (N = 81, 89.0 %) were initially diagnosed as advanced/metastatic, compared to 10 patients (11.0 %) who already had an early diagnosis of lung cancer. For the latter, the median time from early diagnosis to diagnosis of advanced/ metastatic cancer was 24.1 months (IQR, 9.3–59.3).

Crizotinib was administered to 40 patients (44 %) as first line treatment, 35 patients (38.5 %) as the second line treatment, and 16 patients (17.6 %) as third line treatment. For those patients who received the drug as ≥ 2nd line, median duration of prior therapies was 2.8 months (IQR, 1.4–8.5) (mostly chemotherapy).

3.3. Treatment with crizotinib

Most patients (90.1 %) started treatment with crizotinib at a dose of 250 mg twice daily; a few patients received a lower dose (5 patients received 200 mg/12 h and 3 patients 250 mg/24 h) at the discretion of the treating physician.

Tumor responses are shown in Table 3. Of 81 patients evaluated for efficacy, an objective response was achieved by 48 patients (ORR = 59.3 %), and clinical benefit by 70 patients (CBR = 86.4 %). The median time to best overall response since the start of crizotinib treatment was 3.0 months (IQR, 2.3–5.8; N = 78). The median duration of response was 13.5 months (IQR, 5.3–26.2; N = 35).

The median follow-up was 15.1 months (IQR, 4.2–27.5) for the prospective cohort (N = 42) and 36.8 months (IQR, 18.6–49.4) for the retrospective cohort (N = 49). Progression after treatment with crizotinib was observed in 68 patients (74.7 %). The global median PFS was 15.8 months (95 % CI, 11–8–22.3) (Fig. 2A). A total of 38 patients (41.8 %) died for any reason, compared to 53 patients (58.2 %) who remained alive at data cutoff date. Of the deceased patients, 30 patients had progressed to crizotinib treatment, 6 patients had terminated treatment due to toxicity (3 of them started another treatment) and 2 patients died while being treated with crizotinib. The global median OS was 46.5 months (Fig. 2B).

The median PFS was 11.8 months (95 % CI, 7.1–15.2) and 23.1 months (95 % CI, 14.9–31.6) if the patients had been recruited into the study prospectively or retrospectively, respectively. Similarly, median OS was 34.8 months in the prospective cohort and was not reached in the retrospective cohort (Fig. 2C and D).

3.4. Crizotinib use in 1st line or later lines

Forty patients received crizotinib as 1st line treatment and 51 patients as ≥ 2nd line. An objective response was achieved by 24 patients in 1st line (ORR = 68.6 %) and 24 patients in ≥ 2nd line (ORR = 52.2 %) (Table 4). The median time to best overall response since initiation of treatment was 3.0 months (IQR, 2.2–4.4) for patients in 1st line, and 3.2 months (IQR, 2.4–9.2) for patients in ≥ 2nd line of treatment with crizotinib. Of the patients achieving an objective response, 19 patients in 1st line progressed after a median 9.9 months (IQR, 2.9–20.5) and 17 patients in ≥ 2nd line progressed after 18.2 months (IQR, 6.2–32.4). There were significant differences in PFS and OS between patients in 1st versus in ≥ 2nd line (p = 0.0025 and p = 0.0424, respectively) (Fig. 2E and F). The PFS of patients in 1st line was similar if the patients were recruited retrospectively (N = 18; 16.8 months, 95 % CI: 4.5–25.4) or prospectively (N = 22; 12.1 months, 95 % CI: 2.1–15.2) (p = 0.4213). However, for patients in ≥ 2nd lines of treatment, the PFS was significantly different if the patients were recruited retrospectively (N = 27; 36.7 months, 95 % CI: 20.4–53.0) versus prospectively (N = 24; 10.5 months, 95 % CI: 7.0–20.2) (p = 0.0005).

3.5. Crizotinib efficacy in brain metastasis

Of the 91 patients, 10 patients (11.0 %) presented brain metastasis and were evaluated for treatment with crizotinib (Table 4). Of these, four achieved an objective response but 3 progressed after a median of 5.8 months (IQR, 2.5–20.8). Of the 44 patients without brain metastasis who reached objective response, 33 patients progressed after a median

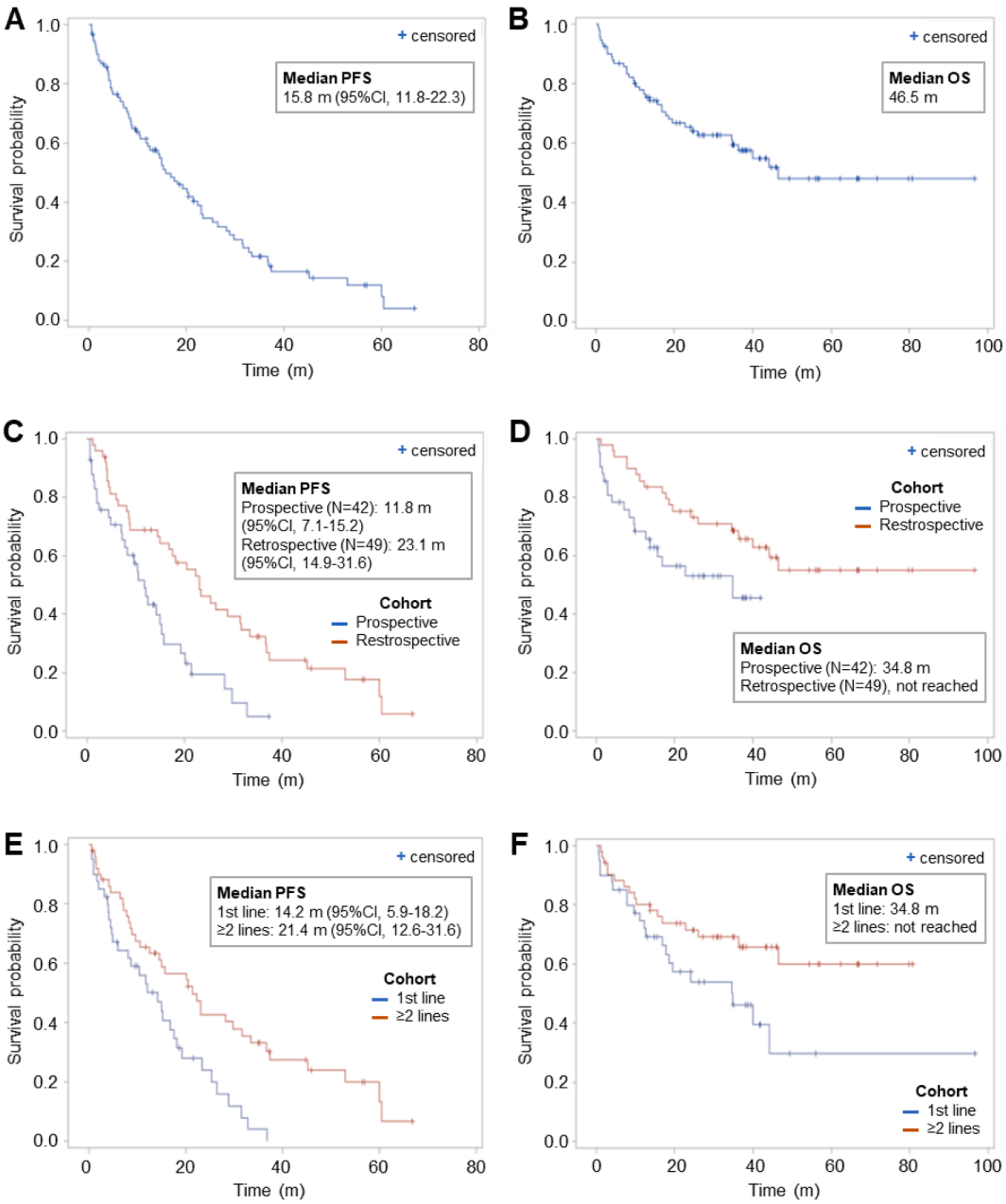

Fig. 2. Kaplan-Meier analysis of PFS and OS in patients treated with crizotinib. A. PFS for the overall population (N = 91); B. OS for the overall population (N = 91); C, PFS of patients according to cohort (retrospective or prospective); D, OS of patients according to cohort (retrospective or prospective)*; E, PFS of patients treated in 1st line (N = 40) or ≥ 2nd line (N = 51) with crizotinib; F, OS of patients treated in 1st line or ≥ 2nd line. CI, confidence interval; m, months; OS, overall survival; PFS, progression free survival (*). Patients included in prospective cohort (N = 42) had 15.1 months (IQR, 4.2–27.5) of median follow-up and for the retrospective cohort (N = 49) the median follow- up was 36.8 months (IQR, 18.6–49.4).

13.5 months (IQR, 6.2–26.4). Of those patients without brain metastases at baseline, 13 patients (22.4 %) developed brain metastases at progression after treatment. There were no significant differences between PFS and OR in patients with or without brain metastasis.

3.6. Crizotinib efficacy according to stage disease

Of 91 patients, 13 (14.3 %) had stage III unresectable disease (including stage IIIA and IIIB), and 78 patients (85.7 %) had stage IV disease. Patients initially diagnosed in early stage (stages IIIA and IIIB) underwent conventional radical chemo-radiotherapy treatment. After relapsing in advanced stage, they started treatment with crizotinib.

3.7. Safety and AEs with crizotinib

During crizotinib treatment, most patients (N = 52, 57.1 %) did not require dose reduction nor interruption. Dose reductions were observed in 22 (22.4 %) patients, and 17 patients (18.6 %) underwent interruptions with 12 days as median time to treatment restart (IQR 7.0–20.0) (Table 3).

A total of 57 patients (62.5 %) experienced at least one crizotinib-related AE at some point during treatment. Of these, 37 patients (39.6 %) had AEs grade 1–2, and 17 patients (19.8 %) AEs grade 3–4 (Table 5). Common AEs observed in this study included elevated transaminase levels (17.6 %), gastrointestinal disorders such as nausea, diarrhea, and constipation (22.6 %), asthenia (14.3 %), and visual disorders (12.2 %). One patient had to discontinue treatment two months

Table 4
Sub-group analysis of patients treated with crizotinib.

Subgroups	ORR[1]	Duration of response[2]	PFS[3]	OS[4]
Line of treatment				
1st line (N = 40)	24 (60.0)	9.9 (2.9–20.5)	14.2 (5.9–18.2)	39.8
≥2nd line (N = 51)	24 (47.0)	18.2 (6.2–32.4)	21.4 (12.6–31.6)	NR
Presence of brain metastasis at baseline				
Yes (N = 10)	4 (40.0)	5.8 (2.5–20.8)	5.9 (0.7–23.1)	40.1
No (N = 71)	44 (62.0)	13.5 (6.2–26.4)	18.2 (12.6–23.3)	NR
Sex				
Male (N = 33)	13 (46.4)	17.8 (13.1–29.8)	17.5 (4.5–28.3)	44.2
Female (N = 58)	35 (66.0)	11.4 (4.8–25.9)	15.2 (10.5–23.1)	46.5

CI, confidence interval; IQR, interquartile range; NR, not reached; PFS, progression-free survival; ORR, overall response rate; OS, overall survival.
[5] Includes IIIA and IIIB.
[1] N (%).
[2] Months, median (IQR).
[3] Months, median (95 % CI).
[4] Months, median.

Table 5
Adverse events reported in patients during crizotinib treatment.

Adverse event	Grade 1–2	Grade 3–4	Grade 5	Total
Elevated transaminase levels	12 (13.2)	3 (3.3)	1 (1.1)	16 (17.6)
Elevated amilase	1 (1.1)	–	–	1 (1.1)
Elevated alkaline phosphatase	1 (1.1)	–	–	1 (1.1)
Extended QT interval in EG	1 (1.1)	–	–	1 (1.1)
Infections and infestations	–	2 (2.2)	–	2 (2.2)
Benign neoplasia	–	1 (1.1)	–	1 (1.1)
Bradycardia	3 (3.3)	1 (1.1)	–	4 (4.4)
Skin disorders	1 (1.1)	1 (1.1)	–	2 (2.2)
Anemia	1 (1.1)	–	–	1 (1.1)
Leucopenia	–	1(1.1)	–	1 (1.1)
Neutropenia	4 (4.4)	4 (4.4)	–	8 (8.8)
Amenorrhea	1 (1.1)	–	–	1 (1.1)
Decreased appetite	6 (6.6)	–	–	6 (6.6)
Hypophosphatemia	–	–	–	1 (1.1)*
Dysgeusia	2 (2.2)	–	–	2 (2.2)
Dizziness	1 (1.1)	–	–	1 (1.1)
Nervous system disorders	1 (1.1)	1 (1.1)	–	2 (2.2)
Gastrointestinal disorders	25 (1.1)	4 (4.4)	–	29 (31.9)
Asthenia	13 (14.3)	–	–	13 (14.3)
Edema	15 (16.5)	–	–	15 (16.5)
Pyrexia	1 (1.1)	–	–	1 (1.1)
Visual disorders	11 (12.1)	1 (1.1)	–	12 (13.2)
Renal and urinary disorders	–	1 (1.1)	–	1 (1.1)
Respiratory disorders	2 (2.2)	2 (2.2)	–	5 (5.5)

* No grade recorded.
EG, electrocardiogram.

after initiation because of toxicity and died one month later due to treatment-related elevated transaminase levels causing liver failure. Five patients presented grade 5 AEs unrelated to treatment. SAEs were observed in 25 patients (27.5 %), which were treatment-related in 8 patients (8.8 %): increased transaminases in five patients, bradycardia in one patient, and neutropenia in two patients.

3.8. Treatment post-crizotinib

After treatment with crizotinib, 59/91 patients (64.8 % of the total) received subsequent treatment. Of these patients, 41 patients (69.5 %) received a single treatment, 14 (23.7 %) two more lines of therapy and 4 (6.8 %) >2 treatments (Table 6). A total of 50 patients (85 %) received a sequence of treatment with a next generation ALK inhibitor. Most patients received alectinib (68.5 %), but some were treated with ceritinib (10.2 %) or brigatinib (5.1 %). The median duration of treatment for the first drug used after crizotinib was 4.0 months (IQR,1.4–13.1) for 53 patients. The main reason for discontinuation was progression (77.4 % of the patients). Thirty patients were still on treatment at data cut-off date.

Table 6
Treatments post-crizotinib.

Treatment	1st treatment (N = 59)	2nd treatment (N = 17)	3rd treatment (N = 4)
Alectinib	40 (67.8)	5 (29.4)	–
Atezolizumab	–	1 (5.9)	–
Brigatinib	3 (5.1)	5 (29.4)	–
Carboplatin and gemcitabine	1 (1.7)	–	–
Carboplatin and pemetrexed	5 (8.5)	1 (5.9)	–
Ceritinib	6 (10.2)	1 (5.9)	–
Cisplatin and pemetrexed	1 (1.7)	–	–
Docetaxel and nintedanib	1 (1.7)	–	–
Erlotinib	–	1 (5.9)	–
Lorlatinib	1 (1.7)	–	2 (50.0)
Nivolumab	1 (1.7)	–	–
Paclitaxel	–	1 (5.9)	–
Pembrolizumab	–	–	1 (25.0)
Pemetrexed	–	1 (5.9)	1 (25.0)
Vinorelbine	–	1 (5.9)	–

All values are given as N (%).

3.9. Patient-reported outcomes

Of the 42 patients that had been enlisted prospectively, 30 completed the Spanish-validated versions of the EORTC QLQ-C30 and its lung cancer module QLQ-LC13 at baseline. After the third cycle of treatment with crizotinib, 17 and 16 patients completed the QLQ-C30 and the QLQ-LC13, respectively. The median (IQR) baseline score for the global QoL scale was 50.0 [33.3–66.7]. At the end of the first cycle of treatment, the median (IQR) score was 62.5 points [33.3–66.7] showing a numerical improvement in this category. The percentage of patients who reported an improvement, stable state, or a worsening of their functioning domains or symptoms after 3 cycles of treatment are shown in Fig. 3. The results show that most functioning domains or symptoms improved or remained stable for most patients. Dyspnea (in both questionnaires) and peripheral neuropathy in the QLQ-LC13 questionnaire were the symptoms that were reported as worsening by just under 50 %.

4. Discussion

This is the first study exploring real-world data of patients with *ALK*-positive NSCLC treated with crizotinib in Spain. We first evaluated incidence of *ALK* rearrangements in a prospective cohort of 559 patients diagnosed with NSCLC. Secondly, we analyzed the clinical effectiveness and safety of treatment with crizotinib in 91 *ALK*-positive patients. Our analysis complements and supports previous real-world studies carried out in other countries, which suggest that crizotinib has a positive impact on outcomes in this subgroup of molecularly defined patients.

In Spain, ALK is the second-most tested biomarker (80.1 % of NSCLC patients), only surpassed by EGFR testing (91.4 %) [9]. As in previous studies, almost all of the ALK determinations analyzed in our study were performed by IHC and FISH [8,9]. Two recent registry-based studies carried out in Spain showed that the rate of *ALK*-positivity among patients diagnosed with NSCLC was 3.0 %-3.4 % [8,9]. Our study showed a

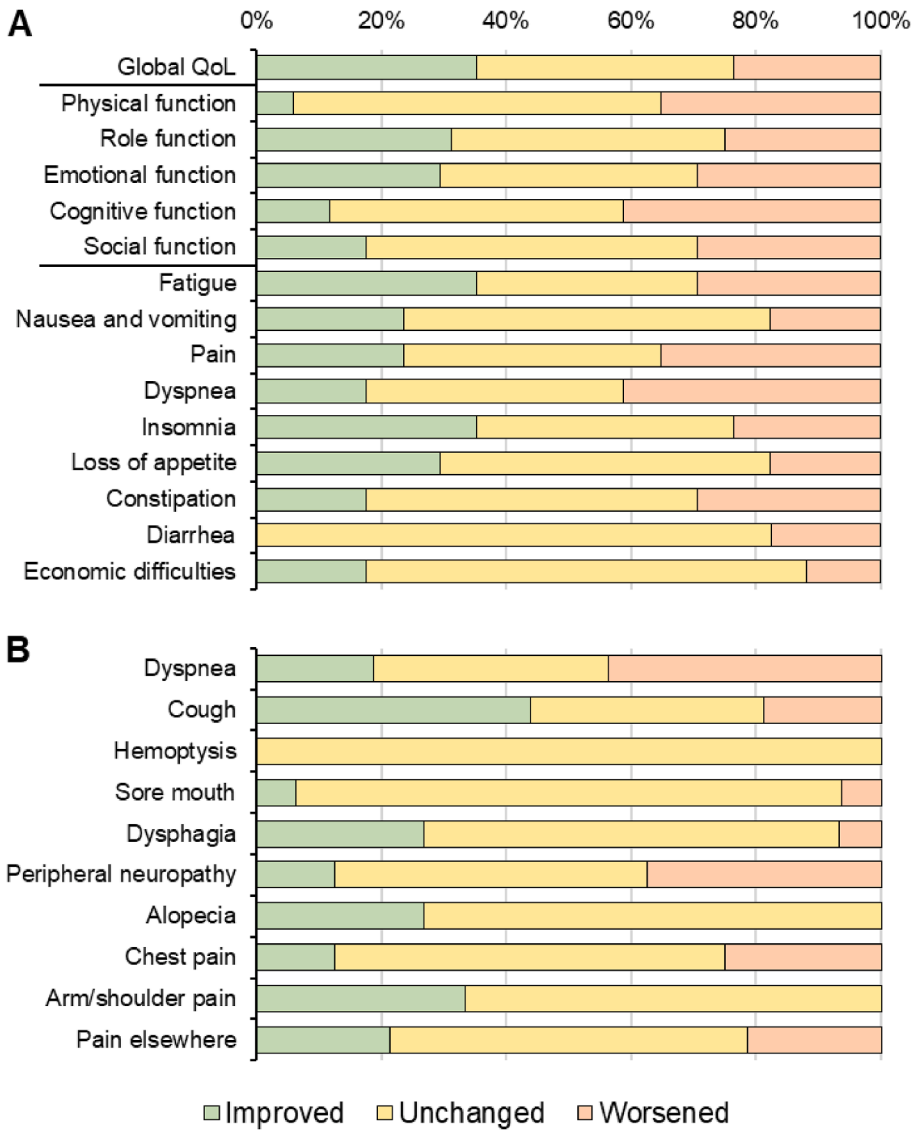

Fig. 3. Change from baseline to month 3 of treatment with crizotinib in the EORTC QLQ-C30 (A) and QLQ-LC13 (B) questionnaires. Bars represent the percentage of patients who at month 3 indicated that their status had improved, not changed, or worsened with respect to baseline for each of the areas and domains of the questionnaires.

slightly higher incidence of 5.5 %, comparable to the 4–6 % which has been observed in other countries [5,6,25–27]. This difference could be explained by the progressive acquisition of technical experience and knowledge in *ALK* testing over time. In this regard, it is important to point out that real world studies may have limitations in terms of study populations. In our incidence sub-study, the investigator excluded some patients for *ALK* testing because of inconsistent histology, smoking or advanced age, and this could have affected the observed incidence of *ALK*. However, consistent with previous studies, our cohort had detected an enrichment of *ALK*-positive among younger, female, adenocarcinoma histology, and non-smoking patients [26]. A rare case of a patient harboring co-occurring *ALK/ROS1* rearrangements was detected. Since *ROS1* testing in NSCLC samples is often considered only after negative results for *EGFR* and *ALK* owing a known mutual exclusion between biomarkers, it could be hypothesized that a *ROS1* subclone perhaps coexists along with the dominant *ALK* tumor. In any case, there are some publications on this regard suggesting that crizotinib could be a good therapeutic option for these rare cases [28,29].

In the phase III PROFILE 1014 study, which compared crizotinib in the first line setting compared with platinum-based doublet chemotherapy, crizotinib demonstrated a significantly higher ORR (74 vs 45 %, $p < 0.001$) and significantly extended PFS compared to chemotherapy (10.9 vs 7.4 months, $p < 0.0001$) [15]. In our study, the ORR of the overall population was 52.8 % (60.0 % if only patients in first line are considered), which is somewhat lower than in the clinical trial, although the median PFS was notably longer, 15.8 months (14.2 months for only patients in first line). However, the median PFS in our study is comparable to that of other real-world observational studies of *ALK*-positive NSCLC patients treated with crizotinib published in recent years, which ranged from 13.0 to 16.5 months [30–36].

It is worth mentioning that in our study, the median PFS and OS resulted markedly different in the cohort of patients included prospectively or retrospectively. The median PFS was 11.8 months vs 23.1 months ($p = 0.0014$) and the median OS 34.8 months vs not reached ($p = 0.0312$) in the prospective and retrospective cohort respectively. These differences could be due in part to the different follow-up times of these cohorts (15.1 vs 36.8 months), as the prospective data could be immature, but other factors such as previous lines of treatment before crizotinib or performance status of patients at the time of starting crizotinib could be involved as well. Interestingly, for patients treated with

Table A1
Real-world studies of crizotinib.

Study and reference	Country	Patients treated with crizotinib, N	Crizotinib in 1L, %	Brain metastasis, %	ORR, %	mDOT, m	mPFS (96% CI), m	mOS (96% CI), m
THIS STUDY	Spain	91	44	12.1	52.8	13.5	15.8 (11.8–22.3)	46.5
Gibson 2021 [36]	Canada	72	100	19.0	45.0	–	16.0 (12.1–22.0)	46.6 (25.1–NR)
Tsimafeyeu 2019 [37]	Russia	96	71	17.0	34	–	–	31.0 (28.5–33.5)
Yang 2019 [35]	China	201	53.2	21.9	62.2	20.7	13.2 (10.8–15.6)	50.5 (37.0–64.0)
Xing 2019 [33]	China	428	63.8	22.2	72.6	13.6	14.4 (12.4–16.4)	53.4 (33.7–73.1)
Flores del Valle 2019 [34]	Singapore	22	50.0	50.0	64.0	8.5	1L: 15.0	1L: NR
Ueno 2019 [44]	Japan	2,028	27.8	–	–	7.9	–	–
Gobbini 2019 [38]	Italy	244	28.0	25.4		10.2	1L: 10.7 (9.0–14.7) 2L: 8.1 (6.7–1.2)	1L: 61.6 (35.0–NR) 2L: 56.8 (42.0–77.9)
Liu 2019 [32]	China	104	60.0	38.5	82.7	16.0	13.0 (9.0–17.0)	36.0 (31.0–41.0)
Davis 2018 [48]	US, Canada	212	65	15.6	66	8.7	9.5 (8.7–10.1)	23.4 (19.5–NR)
Zhou 2018 [30]	China	32	100	25.5	78.1	–	16.1 (12.7–19.4)	–
Reynolds 2018 [49]	US	199	61.8	32.2	–	8.5	1L: 10.4 (7.3–12.3) 2L: 8.6 (4.5–15.8)	33.8 (24.3–38.8)
Duruisseaux 2017 [41]	France	318	54	34.9	50.2	–	6.8 (5.6–8.3)	16.6 (12.2–19.6)
Davis 2017 [50]	Europe	303	34	–	65.7	7	1L: 7.3 (6.6–8.5) 2L: 7.0 (6.6–8.0)	1L: 20.6 (17.0–25.2) 2L: 20.4 (18.2–22.9)
Cui 2016 [51]	China	80	–	–	1L: 73.3 2L: 65.4	–	1L: 13.3 (6.5–20.0) 2L: 9.9 (6.4–13.4)	–

1L, first line; 2L, second line; CI, confidence interval; m, months; mDOT, median duration of treatment; mOS, median overall survival; mPFS, median progression-free survival; ORR, overall response rate

crizotinib in first line there was no difference in median PFS if the data was retrospective or prospective (crizotinib entered the Spanish market in January 2017), but the difference in median PFS was very significant if patients were treated with crizotinib in ≥ 2nd line (retrospective vs prospective, 36.7 vs 10.5 months, p = 0.0005).

The median OS observed in this study was 46.5 months, consistent with that observed in other real-world studies (range: 31 months–not reached) [32–38]. Longer median OS figures across several series have been published so far (see Supplementary Information, Table 1). At this time, the current standard of care for first line in patients with *ALK*-positive NSCLC has changed to second-third generation ALK inhibitors. In the ALEX study, after a follow-up of 48.2 months, alectinib did not reach the median OS vs 57.4 months for crizotinib (stratified HR = 0.67, 95 % CI: 0.46–0.98) [38]. For its part, the median OS was not reached in either group at the brigatinib's ALTA 1L trial (HR = 0.81, 95 % CI: 0.53–1.22) [39]. Similarly, in the CROWN study comparing first line crizotinib and lorlatinib (third generation ALK TKI) [39], the OS data was still immature at the time of data cutoff, but the HR for death was 0.72 (95 % CI, 0.41 to 1.25), with a not significant difference in OS the between groups. Recently, this study submitted an update to its primary endpoint (PFS) after a median duration of follow-up by BICR of 36.7 months for lorlatinib and 29.3 months for crizotinib. Median PFS by BICR was NR (95 % CI, NR-NR) with lorlatinib and 9.3 months (95 % CI, 7.6–11.1 months) with crizotinib (HR, 0.27; 95 % CI, 0.184–0.388). Notably, PFS at 36 months was 63.5 % and 18.9 % for lorlatinib and crizotinib respectively [40].

In our study, the median OS in patients treated with crizotinib in first line was 34.8 months and was not reached in patients treated in ≥ 2nd line. This latest data is in line with the results of a real world French cohort of 318 highly pretreated *ALK*-positive patients receiving multiple lines of chemotherapy prior crizotinib as their first TKI, reaching a remarkable median OS of 89.6 months as assessed from diagnosis of metastatic disease [41]. In contrast, in PROFILE 1014, the longest OS was observed in patients treated with crizotinib in 1st line who received a subsequent ALK tyrosine kinase inhibitor [16], a result that was also confirmed in real-world cohorts [32,33,38]. Additionally, the type of prior chemotherapy in patients with 2nd line crizotinib could influence its effectiveness [30].

Brain metastases are frequent in *ALK*-positive NSCLC already at diagnosis [42], and in our study this was the case in 12.1 % of patients. Crizotinib has limited intracranial activity. In this study, the presence of brain metastasis at diagnosis was associated with worse response and survival outcomes compared with patients without brain metastasis, although the differences were not statistically significant. Brain metastasis in NSCLC patients develops in 15–40 % of *ALK*-positive patients after first diagnosis [43], and in our study 22.4 % of patients developed brain metastasis while receiving crizotinib. Progression of preexisting or development of new metastases while receiving therapy is a common manifestation of acquired resistance to crizotinib [43].

The most common reason for discontinuation in our study was progression owing to the inevitable development of resistance. Treatment with crizotinib appeared to be well tolerated, as 57.1 % of patients did not require a dose interruption or reduction. Nine patients (11.4 %) discontinued treatment because of toxicity. This was similar to the results from PROFILE 1014, in which 12 % of patients permanently discontinued crizotinib treatment resulting from AEs [16,15]. Gastrointestinal disorders, such as vomiting and diarrhea, followed by elevated transaminase levels, were the most common AE reported in our study and were consistent with previous reports [16,31,44]. Visual disorders were very commonly observed in numerous studies, as in PROFILE 1014, where they reached 73.1 % of patients [16], but they were only present in 13.2 % of our cohort. In this study, patients treated tended to have low-grade toxicity events (grades 1–2). Compared with chemotherapy, crizotinib has been associated with a greater reduction in lung cancer symptoms and a greater improvement in quality of life [14,45,46]. In our study, most patients reported an improvement in

terms of global QoL after the first cycle of treatment with crizotinib and after three months of treatment their functioning domains and symptom control status had improved or remained stable (60–80 % of the patients), although the low number of patients evaluated limited the analysis.

Likewise, a large proportion of patients were subsequently treated with another ALK TKI after crizotinib discontinuation (50/59, 85 %). In most cases the chosen ALK TKI was alectinib, but some patients were also treated with ceritinib or brigatinib. The relative proportions of use of the ALK TKIs could reflect in part their availability in Spain at the time of the study. Studies of real-world clinical practice show that alectinib is a common second ALK TKI used after crizotinib progression in 1st line of treatment [47].

Our study should be viewed within the context of the inherent limitations common to all real-world studies. Additionally, the relative immaturity of the data for the prospective cohort, which accounted for almost half of the population, may have affected the PFS and OS results. It is important to note that, as in all real-world studies, the lack of adequate control and standards for the reporting of information in relation to safety can make it possible that minor toxicities are not correctly reported or classified as adverse events related to the drug. Also, sub-groups were imbalanced, which limited statistical analyses. However, strengths of our study were that it was the first in Spain analyzing this group of patients and that it was multicentric.

5. Conclusions

Our study of the incidence and effectiveness of treatment of crizotinib reveals the real-world clinical experience from hospitals in Spain. It confirms the therapeutic benefit of crizotinib in patients with advanced *ALK*-positive NSCLC observed in previous clinical trials and retrospective studies. The sequential administration of crizotinib and new-generation TKIs might further improve patients' treatment options and outcomes, but observational studies are required to confirm this possibility in real-world populations.

CRediT authorship contribution statement

Carlos Aguado de la Rosa: Investigation. **Patricia Cruz Castellanos:** Investigation. **Martín Lázaro-Quintela:** Investigation. **Manuel Dómine:** Investigation. **Sergio Vázquez Estévez:** Investigation. **Guillermo López-Vivanco:** Investigation. **José Luis Fírvida Pérez:** Investigation. **José Luis Alonso Romero:** Investigation. **Lioba Ferrera Delgado:** Investigation. **Carlos García Girón:** Investigation. **Pilar Diz Taín:** Investigation. **Rosa Álvarez Álvarez:** Investigation. **Pilar Mut Sanchís:** Investigation. **Inmaculada Fernández Cantón:** Investigation. **Isabel Manrique Abós:** Investigation. **Maite Martínez Aguillo:** Investigation. **Lorenzo Gómez-Aldaraví Gutiérrez:** Investigation. **Ana Laura Ortega Granados:** Investigation. **Ruth Álvarez Cabellos:** Investigation. **Arancha García Sebastián:** Project administration. **Luis Fernando García Sifuentes:** Project administration, Writing – review & editing. **Noemí Reguart:** Conceptualization, Methodology, Investigation, Supervision, Writing – original draft.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A

References

[1] Hyuna Sung, Jacques Ferlay, Rebecca L. Siegel, Mathieu Laversanne, Isabelle Soerjomataram, Ahmedin Jemal, Freddie Bray, Global cancer statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries, CA Cancer J. Clin. 71 (3) (2021) 209–249.

[2] Cesare Gridelli, Antonio Rossi, David P. Carbone, Juliana Guarize, Niki Karachaliou, Tony Mok, Francesco Petrella, Lorenzo Spaggiari, Rafael Rosell, Non-small-cell lung cancer, Nat. Rev. Dis. Primers 1 (1) (2015), https://doi.org/10.1038/nrdp.2015.9.

[3] Las cifras de cáncer en España 2021, Sociedad Española de Oncología Médica (www.seom.org). Accessed at https://seom.org/images/Cifras_del_cancer_en_Espnaha_2021.pdf (Accessed on 10 September 2021). 2021.

[4] Manabu Soda, Young Lim Choi, Munehiro Enomoto, Shuji Takada, Yoshihiro Yamashita, Shunpei Ishikawa, Shin-ichiro Fujiwara, Hideki Watanabe, Kentaro Kurashina, Hisashi Hatanaka, Masashi Bando, Shoji Ohno, Yuichi Ishikawa, Hiroyuki Aburatani, Toshiro Niki, Yasunori Sohara, Yukihiko Sugiyama, Hiroyuki Mano, Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer, Nature 448 (7153) (2007) 561–566.

[5] L.A. Pikor, V.R. Ramnarine, S. Lam, W.L. Lam, Genetic alterations defining NSCLC subtypes and their therapeutic implications, Lung Cancer. 82 (2013) 179–189, https://doi.org/10.1016/j.lungcan.2013.07.025.

[6] Fiona H. Blackhall, Solange Peters, Lukas Bubendorf, Urania Dafni, Keith M. Kerr, Henrik Hager, Alex Soltermann, Kenneth J. O'Byrne, Christoph Dooms, Aleksandra Sejda, Javier Hernández-Losa, Antonio Marchetti, Spasenija Savic, Qiang Tan, Erik Thunnissen, Ernst-Jan M. Speel, Richard Cheney, Daisuke Nonaka, Jeroen de Jong, Miguel Martorell, Igor Letovanec, Rafael Rosell, Rolf A. Stahel, Prevalence and clinical outcomes for patients with ALK-positive resected stage I to III adenocarcinoma: results from the European Thoracic Oncology Platform Lungscape Project, J. Clin. Oncol. 32 (25) (2014) 2780–2787.

[7] María Sánchez-Ares, José M. Cameselle-Teijeiro, Sergio Vázquez-Estévez, Martín Lázaro-Quintela, Ángel Vázquez-Boquete, Francisco J. Afonso-Afonso, Joaquín Casal-Rubio, Ana L. González-Piñeiro, Yolanda Rico-Rodríguez, José L. Fírvida-Pérez, Juan Ruíz-Bañobre, Elena Couso, Lucía Santomé, Raquel Pérez-Becerra, Rosario García-Campelo, Margarita Amenedo, Cristina Azpitarte-Raposeiras, José Antúnez, Ihab Abdulkader, Fluorescence in situ hybridization

analysis of the ALK gene in 2,045 non-small cell lung cancer patients from North-Western Spain (Galicia), Oncol. Lett. 12 (2) (2016) 1403–1407.
[8] Mariano Provencio, Enric Carcereny, Delvys Rodríguez-Abreu, Rafael López-Castro, María Guirado, Carlos Camps, Joaquim Bosch-Barrera, Rosario García-Campelo, Ana Laura Ortega-Granados, José Luis González-Larriba, Joaquín Casal-Rubio, Manuel Domine, Bartomeu Massutí, María Ángeles Sala, Reyes Bernabé, Juana Oramas, Elvira del Barco, Lung cancer in Spain: information from the Thoracic Tumors Registry (TTR study), Transl. Lung Cancer Res. 8 (4) (2019) 461–475.
[9] Clara Salas, Javier Martín-López, Antonio Martínez-Pozo, Teresa Hernández-Iglesias, David Carcedo, Lucia Ruiz de Alda, J Francisco García, Federico Rojo, Real-world biomarker testing rate and positivity rate in NSCLC in Spain: prospective Central Lung Cancer Biomarker Testing Registry (LungPath) from the Spanish Society of Pathology (SEAP), J. Clin. Pathol. 75 (3) (2022) 193–200.
[10] Muhammad Khan, Jie Lin, Guixiang Liao, Yunhong Tian, Yingying Liang, Rong Li, Mengzhong Liu, Yawei Yuan, ALK Inhibitors in the Treatment of ALK Positive NSCLC, Front. Oncol. 8 (2018), https://doi.org/10.3389/fonc.2018.00557.
[11] R.S. Herbst, D. Morgensztern, C. Boshoff, The biology and management of non-small cell lung cancer, Nature 553 (2018) 446–454, https://doi.org/10.1038/nature25183.
[12] D Ross Camidge, Yung-Jue Bang, Eunice L Kwak, A John Iafrate, Marileila Varella-Garcia, Stephen B Fox, Gregory J Riely, Benjamin Solomon, Sai-Hong I Ou, Dong-Wan Kim, Ravi Salgia, Panagiotis Fidias, Jeffrey A Engelman, Leena Gandhi, Pasi A Jänne, Daniel B Costa, Geoffrey I Shapiro, Patricia LoRusso, Katherine Ruffner, Patricia Stephenson, Yiyun Tang, Keith Wilner, Jeffrey W Clark, Alice T Shaw, Activity and safety of crizotinib in patients with ALK-positive non-small-cell lung cancer: updated results from a phase 1 study, Lancet Oncol. 13 (10) (2012) 1011–1019.
[13] Fiona Blackhall, D. Ross Camidge, Alice T. Shaw, Jean-Charles Soria, Benjamin J. Solomon, Tony Mok, Vera Hirsh, Pasi A. Jänne, Yuankai Shi, Pan-Chyr Yang, Tommaso De Pas, Toyoaki Hida, Javier Castro De Carpeño, Silvana Lanzalone, Anna Polli, Shrividya Iyer, Arlene Reisman, Keith D. Wilner, Dong-Wan Kim, Final results of the large-scale multinational trial PROFILE 1005: efficacy and safety of crizotinib in previously treated patients with advanced/metastatic ALK-positive non-small-cell lung cancer, ESMO Open. 2 (3) (2017) e000219.
[14] Alice T. Shaw, Dong-Wan Kim, Kazuhiko Nakagawa, Takashi Seto, Lucio Crinó, Myung-Ju Ahn, Tommaso De Pas, Benjamin Besse, Benjamin J. Solomon, Fiona Blackhall, Yi-Long Wu, Michael Thomas, Kenneth J. O'Byrne, Denis Moro-Sibilot, D. Ross Camidge, Tony Mok, Vera Hirsh, Gregory J. Riely, Shrividya Iyer, Vanessa Tassell, Anna Polli, Keith D. Wilner, Pasi A. Jänne, Crizotinib versus chemotherapy in advanced ALK-positive lung cancer, N. Engl. J. Med. 368 (25) (2013) 2385–2394.
[15] Benjamin J. Solomon, Tony Mok, Dong-Wan Kim, Yi-Long Wu, Kazuhiko Nakagawa, Tarek Mekhail, Enriqueta Felip, Federico Cappuzzo, Jolanda Paolini, Tiziana Usari, Shrividya Iyer, Arlene Reisman, Keith D. Wilner, Jennifer Tursi, Fiona Blackhall, First-line crizotinib versus chemotherapy in ALK-positive lung cancer, N. Engl. J. Med. 371 (23) (2014) 2167–2177.
[16] Benjamin J. Solomon, Dong-Wan Kim, Yi-Long Wu, Kazuhiko Nakagawa, Tarek Mekhail, Enriqueta Felip, Federico Cappuzzo, Jolanda Paolini, Tiziana Usari, Yiyun Tang, Keith D. Wilner, Fiona Blackhall, Tony S. Mok, Final overall survival analysis from a study comparing first-line crizotinib versus chemotherapy in ALK-mutation-positive non–small-cell lung cancer, JCO 36 (22) (2018) 2251–2258.
[17] Alice T. Shaw, Benjamin J. Solomon, Benjamin Besse, Todd M. Bauer, Chia-Chi Lin, Ross A. Soo, Gregory J. Riely, Sai-Hong Ignatius Ou, Jill S. Clancy, Sherry Li, Antonello Abbattista, Holger Thurm, Miyako Satouchi, D. Ross Camidge, Steven Kao, Rita Chiari, Shirish M. Gadgeel, Enriqueta Felip, Jean-François Martini, ALK resistance mutations and efficacy of lorlatinib in advanced anaplastic lymphoma kinase-positive non-small-cell lung cancer, J. Clin. Oncol. 37 (16) (2019) 1370–1379.
[18] E. Felip, A.T. Shaw, A. Bearz, D.R. Camidge, B.J. Solomon, J.R. Bauman, T. M. Bauer, S. Peters, F. Toffalorio, A. Abbattista, H. Thurm, G. Peltz, R. Wiltshire, B. Besse, Intracranial and extracranial efficacy of lorlatinib in patients with ALK-positive non-small-cell lung cancer previously treated with second-generation ALK TKIs, Ann. Oncol. 32 (5) (2021) 620–630.
[19] World Medical Association, World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects, JAMA 310 (2013) 2191–2194, https://doi.org/10.1001/jama.2013.281053.
[20] M. Amin, AJCC Cancer Staging Manual, 8th Edition, 8th ed., Springer, Switzerland, 2017.
[21] M.A.G. Sprangers, A. Cull, K. Bjordal, M. Groenvold, N.K. Aaronson, The European organization for research and treatment of cancer. Approach to quality of life assessment: guidelines for developing questionnaire modules. EORTC Study Group on Quality of Life, Qual. Life Res. 2 (4) (1993) 287–295.
[22] B. Bergman, N.K. Aaronson, S. Ahmedzai, S. Kaasa, M. Sullivan, The EORTC QLQ-LC13: a modular supplement to the EORTC Core Quality of Life Questionnaire (QLQ-C30) for use in lung cancer clinical trials. EORTC Study Group on Quality of Life, Eur. J. Cancer 30A (1994) 635–642, https://doi.org/10.1016/0959-8049(94)90535-5.
[23] J. Arrarás, E. Prujá, M. Marcos, M. Tejedor, J. Illarramendi, R. Vera, et al., El cuestionario de calidad de vida para cáncer de pulmón de la EORTC QLQ-LC13: Estudio de validación para nuestro país, Oncología 23 (2000) 127–134.
[24] J.I. Arraras, F. Arias, M. Tejedor, E. Pruja, M. Marcos, E. Martínez, J. Valerdi, The EORTC QLQ-C30 (version 3.0) Quality of Life questionnaire: validation study for Spain with head and neck cancer patients, Psychooncology 11 (3) (2002) 249–256.
[25] Fabrice Barlesi, Julien Mazieres, Jean-Philippe Merlio, Didier Debieuvre, Jean Mosser, Hervé Lena, L'Houcine Ouafik, Benjamin Besse, Isabelle Rouquette, Virginie Westeel, Fabienne Escande, Isabelle Monnet, Antoinette Lemoine, Rémi Veillon, Hélène Blons, Clarisse Audigier-Valette, Pierre-Paul Bringuier, Régine Lamy, Michèle Beau-Faller, Jean-Louis Pujol, Jean-Christophe Sabourin, Frédérique Penault-Llorca, Marc G Denis, Sylvie Lantuejoul, Franck Morin, Quân Tran, Pascale Missy, Alexandra Langlais, Bernard Milleron, Jacques Cadranel, Jean-Charles Soria, Gérard Zalcman, Routine molecular profiling of patients with advanced non-small-cell lung cancer: results of a 1-year nationwide programme of the French Cooperative Thoracic Intergroup (IFCT), Lancet 387 (10026) (2016) 1415–1426.
[26] P.L. Chia, P. Mitchell, A. Dobrovic, T. John, Prevalence and natural history of ALK positive non-small-cell lung cancer and the clinical impact of targeted therapy with ALK inhibitors, Clin. Epidemiol. 6 (2014) 423–432, https://doi.org/10.2147/CLEP.S69718.
[27] J.A. Barta, C.A. Powell, J.P. Wisnivesky, Global epidemiology of lung cancer, Ann. Glob Health 85 (2019) 8, https://doi.org/10.5334/aogh.2419.
[28] A. Uguen, U. Schick, G. Quéré, A rare case of ROS1 and ALK double rearranged non-small cell lung cancer, J. Thoracic Oncol. 12 (2017) e71–e72, https://doi.org/10.1016/j.jtho.2017.02.007.
[29] Mustafa Korkmaz, Melek Karakurt Eryilmaz, Fifty-five months progression-free survival with crizotinib treatment in coexistence of ALK and ROS1 rearrangements in lung adenocarcinoma: an extremely rare case and review of the literature, Anticancer Drugs 33 (1) (2022) e799–e801.
[30] Jianya Zhou, Jing Zheng, Xiaochen Zhang, Jing Zhao, Yanping Zhu, Qian Shen, Yuehong Wang, Ke Sun, Zeying Zhang, Zhijie Pan, Yihong Shen, Jianying Zhou, Crizotinib in patients with anaplastic lymphoma kinase-positive advanced non-small cell lung cancer versus chemotherapy as a first-line treatment, BMC Cancer 18 (1) (2018), https://doi.org/10.1186/s12885-017-3720-8.
[31] A. Mohieldin, A. Rasmy, M. Ashour, M. Al-Nassar, R.H. Ali, F.G. El-Enezi, Efficacy and safety of crizotinib in patients with anaplastic lymphoma kinase-positive advanced-stage non-small-cell lung cancer, Cancer Manage. Res. 10 (2018) 6555–6561, https://doi.org/10.2147/CMAR.S173084.
[32] Chang Liu, Hui Yu, Qianqian Long, Haiquan Chen, Yuan Li, Weixin Zhao, Kuaile Zhao, Zhengfei Zhu, Si Sun, Min Fan, Jianhua Chang, Jialei Wang, Real world experience of crizotinib in 104 patients with ALK rearrangement non-small-cell lung cancer in a single chinese cancer center, Front. Oncol. 9 (2019), https://doi.org/10.3389/fonc.2019.01116.
[33] Puyuan Xing, Di Ma, Qiang Wang, Xuezhi Hao, Mengzhao Wang, Yan Wang, Li Shan, Tao Xin, Li Liang, Hongge Liang, Yang Du, Zhaohui Zhang, Junling Li, Impact of crizotinib on long-term survival of ALK-positive advanced non-small-cell lung cancer: a Chinese multicenter cohort study, Chin. J. Cancer Res. 31 (3) (2019) 481–488.
[34] M.F.F. Del Valle, A.Y.-C. Chang, Real world experience on treatment, outcome and toxicity of crizotinib in patients with anaplastic lymphoma kinase positive advanced non-small cell lung cancer, J. Thorac Dis. 11 (2019) 3864–3873, https://doi.org/10.21037/jtd.2019.09.15.
[35] Guangjian Yang, Di Ma, Haiyan Xu, Lu Yang, Junling Li, Puyuan Xing, Xuezhi Hao, Yan Wang, Treatment duration as a surrogate endpoint to evaluate the efficacy of crizotinib in sequential therapy for patients with advanced ALK-positive non-small cell lung cancer: a retrospective, real-world study, Cancer Med. 8 (13) (2019) 5823–5830.
[36] Amanda J.W. Gibson, Adrian Box, Michelle L. Dean, Anifat A. Elegbede, Desiree Hao, Randeep Sangha, D. Gwyn Bebb, Retrospective real-world outcomes for patients with ALK-rearranged lung cancer receiving ALK receptor tyrosine kinase inhibitors, JTO Clin. Res. Rep. 2 (4) (2021) 100157.
[37] Ilya Tsimafeyeu, Fedor Moiseenko, Sergei Orlov, Elena Filippova, Alexander Belonogov, Aleksey Nebesnykh, Amir Khalimov, Elena Karabina, Valentina Shikina, Ahmed Abdelgafur, Galina Statsenko, Irina Titova, Dmitry Isaichikov, Galina Makarnyaeva, Aleksey Mordovskiy, Oksana Barkovskaya, Aleksey Smirnov, Marina Gikalo, Nikita Savelov, Dmitry Kosov, Evgeny Imyanitov, Irina Demidova, Sergei Tjulandin, Overall survival of patients with ALK-positive metastatic non-small-cell lung cancer in the russian federation: nationwide cohort study, J. Glob Oncol. (5) (2019) 1–7.
[38] E. Gobbini, R. Chiari, P. Pizzutillo, P. Bordi, L. Ghilardi, S. Pilotto, G. Osman, F. Cappuzzo, F. Cecere, F. Riccardi, V. Scotti, O. Martelli, G. Borra, E. Maiello, A. Rossi, P. Graziano, V. Gregorc, C. Casartelli, C. Sergi, A. Del Conte, A. Delmonte, C. Bareggi, D. Cortinovis, P. Rizzo, F. Tabbò, G. Rossi, E. Bria, D. Galetta, M. Tiseo, M. Di Maio, S. Novello, Real-world outcomes according to treatment strategies in ALK-rearranged non-small-cell lung cancer (NSCLC) patients: an Italian retrospective study, Clin. Transl. Oncol. 22 (3) (2020) 294–301.
[39] Alice T. Shaw, Todd M. Bauer, Filippo de Marinis, Enriqueta Felip, Yasushi Goto, Geoffrey Liu, Julien Mazieres, Dong-Wan Kim, Tony Mok, Anna Polli, Holger Thurm, Anna M. Calella, Gerson Peltz, Benjamin J. Solomon, First-line lorlatinib or crizotinib in advanced *ALK* -positive lung cancer, N. Engl. J. Med. 383 (21) (2020) 2018–2029.
[40] B.J. Solomon, T.M. Bauer, T. Mok, G. Liu, J. Mazieres, F. de Marinis, et al., Updated Efficacy and Safety From the Phase 3 CROWN Study of First-Line Lorlatinib vs Crizotinib in Advanced Anaplastic Lymphoma Kinase (ALK)–Positive Non-Small Cell Lung Cancer (NSCLC), AACR Annual Meeting. New Orleans, Louisiana, USA, 2022.
[41] Michaël Duruisseaux, Benjamin Besse, Jacques Cadranel, Maurice Pérol, Bertrand Mennecier, Laurence Bigay-Game, Renaud Descourt, Eric Dansin, Clarisse Audigier-Valette, Lionel Moreau, José Hureaux, Remi Veillon, Josiane Otto, Anne Madroszyk-Flandin, Alexis Cortot, François Guichard, Pascaline Boudou-Rouquette, Alexandra Langlais, Pascale Missy, Franck Morin, Denis Moro-Sibilot, Overall survival with crizotinib and next-generation ALK inhibitors in *ALK* -positive non-small-cell lung cancer (IFCT-1302 CLINALK): a

French nationwide cohort retrospective study, Oncotargetics 8 (13) (2017) 21903–21917.
[42] S. Ceddia, G. Codacci-Pisanelli, Treatment of brain metastases in ALK-positive non-small cell lung cancer, Crit. Rev. Oncol. Hematol. 165 (2021), 103400, https://doi.org/10.1016/j.critrevonc.2021.103400.
[43] Daniel B. Costa, Alice T. Shaw, Sai-Hong I. Ou, Benjamin J. Solomon, Gregory J. Riely, Myung-Ju Ahn, Caicun Zhou, S. Martin Shreeve, Paulina Selaru, Anna Polli, Patrick Schnell, Keith D. Wilner, Robin Wiltshire, D. Ross Camidge, Lucio Crinò, Clinical experience with crizotinib in patients with advanced ALK-rearranged non-small-cell lung cancer and brain metastases, J. Clin. Oncol. 33 (17) (2015) 1881–1888.
[44] Naomi Ueno, Shigeo Banno, Yutaka Endo, Motoko Tamura, Katsuko Sugaya, Satoshi Hashigaki, Emiko Ohki, Akinobu Yoshimura, Akihiko Gemma, Treatment status and safety of crizotinib in 2028 Japanese patients with ALK-positive NSCLC in clinical settings, Jpn. J. Clin. Oncol. 49 (7) (2019) 676–686.
[45] Fiona Blackhall, Dong-Wan Kim, Benjamin Besse, Hiroshi Nokihara, Ji-Youn Han, Keith D. Wilner, Arlene Reisman, Shrividya Iyer, Vera Hirsh, Alice T. Shaw, Patient-reported outcomes and quality of life in PROFILE 1007: a randomized trial of crizotinib compared with chemotherapy in previously treated patients with ALK-positive advanced non-small-cell lung cancer, J. Thorac. Oncol. 9 (11) (2014) 1625–1633.
[46] Quan Zhang, Na Qin, Jinghui Wang, Jialin Lv, Xinjie Yang, Xi Li, Jingying Nong, Hui Zhang, Xinyong Zhang, Yuhua Wu, Shucai Zhang, Crizotinib versus platinum-based double-agent chemotherapy as the first line treatment in advanced anaplastic lymphoma kinase-positive lung adenocarcinoma, Thorac. Cancer 7 (1) (2016) 3–8.
[47] Mohammad Jahanzeb, Huamao M. Lin, Xiaoyun Pan, Yu Yin, Yanyu Wu, Beth Nordstrom, Mark A. Socinski, Real-world treatment patterns and progression-free survival associated with anaplastic lymphoma kinase (ALK) tyrosine kinase inhibitor therapies for ALK+ non-small cell lung cancer, Oncologist 25 (10) (2020) 867–877.
[48] K.L. Davis, J.A. Kaye, E.T. Masters, S. Iyer, Real-world outcomes in patients with ALK-positive non-small cell lung cancer treated with crizotinib, Curr. Oncol. 25 (2018) e40–e49, https://doi.org/10.3747/co.25.3723.
[49] Craig Reynolds, Elizabeth Masters, Jenny Black-Shinn, Marley Boyd, Jack Mardekian, Janet Espirito, Marc Chioda, Real-world use and outcomes of ALK-positive crizotinib-treated metastatic NSCLC in US community oncology practices: a retrospective observational study, J Clin Med. 7 (6) (2018) 129.
[50] K.L. Davis, C. Lenz, K. Houghton, J.A. Kaye, Clinical outcomes of crizotinib in real-world practice settings for patients with advanced ALK-Positive non-small cell lung cancer, Int. J. Radiat. Oncol. Biol. Phys. 98 (2017) 238–239, https://doi.org/10.1016/j.ijrobp.2017.01.168.
[51] Shaohua Cui, Yizhuo Zhao, Lili Dong, Aiqin Gu, Liwen Xiong, Jialin Qian, Wei Zhang, Yanjie Niu, Feng Pan, Liyan Jiang, Is there a progression-free survival benefit of first-line crizotinib versus standard chemotherapy and second-line crizotinib in ALK-positive advanced lung adenocarcinoma? A retrospective study of Chinese patients, Cancer Med. 5 (6) (2016) 1013–1021.