Original Study

Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With *EGFR/ALK/ROS* Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis

Antoine Bronstein,[1] Hubert Curcio,[2] Isabelle Monnet,[3] Charles Ricordel,[4] Laurence Bigay-Game,[5] Margaux Geier,[6] Chantal Decroisette,[7] Catherine Daniel,[8] Florian Guisier,[9] Aurélie Swalduz,[10] Anne Claire Toffart,[11] Hélène Doubre,[12] Jean Michel Peloni,[13] Dominique Arpin,[14] Hugues Morel,[15] Remi Veillon,[16] Benedicte Clarisse,[17] Christos Chouaïd,[3] Laurent Greillier,[18] Olivier Bylicki[1], GFPC

Abstract

Feasibility and clinical impact of rebiopsy for a new mutational profile in metastatic non-small-cell lung cancer patients with oncogenic driver progressing after tyrosine kinase inhibitors (TKIs) is poorly studied. In this retrospective multicentric analysis, rebiopsy was performed in 53.4% of cases, providing a new information in 40.3% of cases and enabled identification of resistance mechanisms in 20% of cases. Overall survival did not differ between patients with and without rebiopsy.

Background: For metastatic non-small-cell lung cancer (mNSCLC) patients with oncogenic driver progression after tyrosine kinase inhibitors (TKIs), obtaining a new mutational profile is recommended to assess the mechanism of resistance. The feasibility of that recommendation and its clinical impact remain poorly studied. **Methods:** mNSCLC patients with *EGFR* mutation and *ALK* or *ROS* translocation progressing on optimal TKI therapy were screened for inclusion in an immunochemotherapy trial not requiring a new molecular profile determination. This analysis evaluated the rebiopsy rate and its clinical impact. **Results:** Among 148 patients, 79 (53.4%) analyzable re-biopsies showed 72/132 (54.6%) with *EGFR* mutations, 7/13 (53.8%) had *ALK* translocations and no (0/5) *ROS* translocations. Seventy-nine re-biopsies were tissue (37, 46.8%), liquid (26, 32.9%) or both samples (16, 20.3%). For patients harboring *EGFR* mutations, the rebiopsy was not contributive for 12/72 (16.7%), the initial mutation was not found for 9/72 (12.5%) and

[1]Department of Pneumology, Hôpital d'Instruction des Armées Saint-Anne, Toulon, France
[2]Department of Medical Oncology, Comprehensive Cancer Center François- Baclesse, Caen, France
[3]Department of Pneumology, Centre Hospitalier Intercommunal de Créteil, Créteil, France
[4]Department of Pneumology, CHU Rennes, Univ Rennes 1, INSERM, OSS (Oncogenesis Stress Signaling), Rennes, France
[5]Department of Pneumology & Thoracic Oncology, CHU Toulouse, Hôpital Larrey, Toulouse, France
[6]Department of Thoracic Oncology, CHRU Morvan, Brest, France
[7]Department of Pneumology & Thoracic Oncology, CH Annecy-Genevois, Metz-Tessy, France
[8]Thoracic Oncology Service, Thorax Institute Curie Montsouris, Institut Curie, Paris, France
[9]CHU Rouen, Service de Pneumologie, Oncologie Thoracique et Soins Intensifs Respiratoires, Normandie Univ, UNIROUEN, EA4108 LITIS Lab, Rouen, France
[10]Department of Pneumology, Comprehensive Cancer Centre Léon-Bérard, Lyon, France
[11]Pneumology, CHU Grenoble, Grenoble, France
[12]Thoracic Oncology, Hopital Foch, Suresnes, France
[13]Department of Pneumology, Maison de Santé Protestante de Bordeaux-Bagatelle, Talence, France
[14]Department of Pneumology, Hôpital Nord-Ouest, Villefranche-sur-Saône, France
[15]Department of Pneumology, CHR Orléans, Orléans, France
[16]Department of Pulmonology, Bordeaux University Hospital, Bordeaux, France
[17]Clinical Research Department, Comprehensive Cancer Centre François-Baclesse, Caen, France
[18]Aix–Marseille Univ, APHM, INSERM, CNRS, CRCM, Hôpital Nord, Multidisciplinary Oncology and Therapeutic Innovations, Marseille, France

Submitted: Apr 21, 2025; Revised: Aug 17, 2025; Accepted: Aug 18, 2025; Epub: xxx

Address for correspondence: Christos Chouaïd, MD, PhD, Service de Pneumologie, Centre Hospitalier Intercommunal de Créteil, 40, avenue de Verdun, 94010 Créteil, France
E-mail contact: christos.chouaid@chicreteil.fr

https://doi.org/10.1016/j.cllc.2025.08.009

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https:

only the unchanged initial profile was detected for 22/72 (30.6%); new information was provided for 29/72 (40.3%). Among patients with *ALK*-translocated mNSCLCs, re-biopsies enabled identification of resistance mechanisms for 20%. Overall survival did not differ between patients with rebiopsy and those without. **Conclusions:** In this population of patients with oncogenic driver progression under optimal targeted TKIs and in sufficiently good general condition to be included in an immunochemotherapy trial, only half were re-biopsied. Rebiopsy does not seem to improve the outcomes of these patients.

Clinical Lung Cancer, Vol. 000, No.xxx, 1–7
Keywords: Outcomes, Biopsy, Target therapy, Management, Chemotherapy

Introduction

Over the past several years, the demonstration of oncogene mutations in certain patients with metastatic non-small-cell lung cancers (NSCLCs) has transformed their prognoses.[1] The most frequent activating mutation was in the epidermal growth factor receptor (*EGFR*), ranging according to the country from 12% to > 50% of the patients.[1,2] Those patients received first-line therapy with first- or second-generation tyrosine-kinase inhibitors (TKIs) and, more recently, since the publication of the FLAURA trial results, third-generation TKIs.[3] Patient outcomes of that phase 3 trial, comparing first-line osimertinib to first-generation erlotinib or gefitinib TKI for patients with an exon-19 deletion (exon19del) or exon-21 *L858R* mutation, showed osimertinib significantly prolonged progression-free survival (18.9 vs. 10.2 months; hazard ratio (HR) 0.46; $P < .001$) and overall survival (OS; 38.6 vs. 31.8 months; HR 0.799, $P = .0462$).[4] More recently, first-line treatment options for patients with EGFR-positive NSCLC are evolving and now include also combination therapy with osimertinib and chemotherapy or amivantamab and Lazertinib.[5]

Although only 6% to 8% of the patients had metastatic NSCLCs with an anaplastic lymphoma kinase (*ALK*) or proto-oncogene tyrosine-protein kinase (*ROS*) translocation, they are now treated with TKIs.[6-12] Current recommendations start their first-line therapy with second- or third-generation TKI.[1]

Despite the major progress targeted TKIs achieved, the majority of patients' cancers will progress, with various progression mechanisms, linked or not to the initial oncogenic pathway or by histological transformation.[10] Thus, although osimertinib-resistance mechanisms predominantly arise via mesenchymal-to-epithelial transition (*MET*) amplification, numerous other resistance-acquisition possibilities have been described.[13,14] The multiplicity and complexity of those resistance mechanisms suggest that redetermination of the "progression" molecular profile would help orient subsequent management.[10,13-16]

Indeed, the rapidly expanding diagnostic and therapeutic landscape of precision oncology for NSCLCs may provide therapeutic options at disease progression that simply were not available at diagnosis. However, rebiopsy rates for patients with NSCLC progression remain low, between 15% and 40%, for a variety of reasons.[17-19] Moreover, information on feasibility of establishing a new molecular profile at progression after optimal TKI administration is scarce.[20-23] Hence, patients and physicians are increasingly requesting postprogression (re) biopsies and their analysis with the aim of identifying options for targeted therapy to control the lung cancer. While this strategy is based on good clinical and scientific reasoning, few comprehensive real-world data supporting this concept are available.

This study was undertaken to analyze the rebiopsy rate and its clinical impact on patients with metastatic NSCLCs harboring *EGFR*/*ALK*/*ROS* alterations that progressed under optimal TKI administration. The study population comprised patients screened to be included in an immunochemotherapy trial for which a new molecular biology work-up was not required.[24]

Materials and Methods

The analysis was based on patients with stage IIIB/IV nonsquamous NSCLCs with *EGFR* mutation or *ALK*/*ROS1* translocation screened for entry into the multicenter, open-label, nonrandomized phase II GFPC trial 06-2018.[23] Patients' NSCLCs had to have progressed during or after treatment with one or more EGFR, ALK or ROS1 TKIs, with an Eastern Cooperative Oncology Group performance status (ECOG PS) of 0 or 1 and measurable disease (RECIST v1.1), no prior chemotherapy, and adequate hematological and organ function. Patients with inactive brain metastases were eligible for screening. For patients included after 2 lines of TKI only the period of the last TKI progression was analyzed.

Information collected from patients' medical records stored in electronic case-report forms included sociodemographic characteristics, medical and surgical history, previous oncological treatments, histology, somatic genetic alterations (*EGFR* mutations, *ALK* or *ROS1* translocation or other relevant alterations), and numbers and locations of metastatic sites. All results of tissue and/or liquid samples taken for molecular biology purposes and their source results were retrieved.

Outcomes

The primary objective of this real-world study was to determine the percentage of re-biopsies after progression with optimal TKI administration. The secondary objective was to describe the TKI-resistance mechanisms, according to the therapeutic sequences and rebiopsy impact of subsequent management choices on median OS.

Statistical Analyses

Dichotomous variables are expressed as number (%) with 95% confidence interval (CI), and continuous variables as mean or median with range, 95% CI and/or interquartile range (IQR).

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https:

Statistical analyses were computed with R Studio statistical software (version 2022.12.0 + 353). Each group's OS rate was estimated with the Kaplan–Meier method. OS was defined as the time from the date of progression under last TKI line to death from any cause.

Results

Between September 2019 and October 2021, 148 patients followed in 27 centers were screened: 131 harbored *EGFR* mutations, 13 carried an *ALK* translocation and 5 a *ROS* translocation. The median age of patients was 63 years, 60.8% were female, and 52.7% were never-smokers. The median interval between stage IIIb/IV NSCLC diagnosis and screening was 2.4 years. Among the 148 patients analyzed, 79 (53.4%) had 1 rebiopsy at progression and 6 (9%) had 2. Rebiopsy rates were 54.6% (72/132) for patients with *EGFR* mutations, 53.8.% (7/13) for those with *ALK* translocation and none (0/5) had a *ROS* translocation.

According to management rebiopsy-rate rate varies from 0 to 100% (Supplemental Table 1). Rebiopsy samples were tissue, liquid or both, respectively for 46.8% (37/79), 32.9% (26/79) or 20.3% (16/79). Biopsy sites were mostly the lung (29/79, 37%) and pleura (6/79, 8%) (Table 1). No differences between patients with rebiopsy or not were observed for initial characteristics, the numbers of metastatic sites, or initial molecular profiles or therapeutic strategy.

Rebiopsy results are reported in Table 2 for the 72 patients with NSCLCs harboring *EGFR* mutations: 16.7% (12/72) had no interpretable data and, for 9 (12.5%) of them, the initial mutation was no longer detected. Intriguingly, the same profile as initially was found for 22 (30.6%), while 29 (40.3%) patients' rebiopsies had new markers. Median OS did not differ significantly between patients with rebiopsy or without: 6.6 vs.14.6 months ($P = .85$), respectively. The re biopsy of the 7 ALK patients showed the same histology compare to baseline, with always ALK translocation and no evidence of a resistance mechanism.

Discussion

The rebiopsy rate was 53.4% for patients with an initial oncogenic anomaly whose NSCLCs progressed under optimal latest-generation TKI administration. No significant differences were found according to rebiopsy status between patients' characteristics or the centers where they were managed. Rebiopsies provided new information compared to the initial sample for 40.3% of those harboring alterations but analysis did not reveal any impact on OS according to rebiopsy status.

Our rebiopsy rate was higher than that previously reported. In the prospective ELIOS study, whose primary objective was to compare molecular profiles of advanced NSLCL patients with *EGFR* mutations before and after progression on first-line osimertinib, the rebiopsy rate was only 34%.[20] Those authors explained that low rate to patient refusals, difficulty obtaining specimens, and in association with patients' general clinical condition, lesion location but also the managing team's motivation (120). For patients without identified oncogene anomalies at the time first-line therapy was decided, the rebiopsy rate was even lower. Among 17,477 nonselected stage IV NSCLC patients, only 403 (2.3%) had at least 1 rebiopsy of a primary tumor or metastasis.[23] Biomarker profile

Table 1 Patient Characteristics According to Rebiopsy Status

Characteristic	No Biopsy $n = 69$	Rebiopsy $n = 79$
Age, years, median (SD)	63(11)	63.(10)
Female	41 (59.4%)	49 (62.0%)
ECOG performance status		
0	34 (49.3%)	37 (46.8%)
1	35 (50.7%)	42 (53.2%)
Smoking status		
Former	28 (40.6%)	36 (45.6%)
Current	2 (2.9%)	4 (5.1%)
Never-smoker	39 (56.5%)	39 (49.4%)
Adenocarcinoma	68 (98.6%)	78 (98.7%)
Molecular profile		
EGFR mutation	59 (85.5%)	72 (91.1%)
Exon 19	35 (50.7%)	47 (59.5%)
L858R	14 (20.3%)	22 (27.8%)
Exon 18	0	6 (7.6%)
Exon 20 Insertion	0	2 (2.5%)
C797S	0	2 (2,5%)
T790M	9 (13.0%)	9 (11.4%)
ALK rearrangement	6 (8.7%)	7 (8.9%)
ROS1 fusion,	5 (5.8%)	0
Stage IIIB	5 (7.2%)	0
Stage IV	64 (92.8%)	79 (100%)
Metastatic disease		
1	19 (27.5%)	30 (38%)
2	25 (36,2%)	22 (27.8%)
> 3	25 (36.2%)	27 (34.2%)
Metastasis sites		
Bone	46 (66.7%)	40 (50.6)
Lung	23 (33.3%)	30 (40%)
Pleura	8 (11.6%)	22 (31.9%)
Brain	27 (39.1%)	18 (22.8%)
Lymph nodes	24 (34.8%)	21 (26.6%)
Liver	13 (18.8%)	19 (24.1%)
Adrenal gland	10 514.5%)	11 (13.9%)
Skin	0	1 (1.3%)
Pericardium	3 (4.3%)	3 (3.8%)
Peritoneum	1 (1.4%)	3 (3.8%)
Biopsy site		
Lung		29 (36.7%)
Pleura		6 (7.8%)
Mediastinal node		4 (5.1%)
Bone		2 (2.5%)
Liver		1 (1.3%)
Breast		1 (1.3%)
Skin		1 (1.3%)

Values are n (%), unless stated otherwise.

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https://doi.org/10.1016/j.cllc.2025.09.009

changes, compared to baseline, were observed in 48.9% and 31.3% of patients, revealing findings of potential therapeutic relevance, including a targetable marker, detected in only in 4.4% of the patients' rebiopsies. The greatest impact of rebiopsy in that study was found in the subgroup of patients harboring *EGFR*/*ALK*/*ROS1* alterations. In contrast to our results, those authors found that OS for rebiopsied patients was significantly longer that for controls, regardless of the presence or absence of therapeutically targetable anomalies.

Rebiopsies in this setting pose several challenges in clinical practice. The new specimen may contain insufficient material to obtain a new molecular profile of good quality, as was the case for 16.7% of our patients who had inconclusive results. That eventuality was also reported for a prospective study on 86 NSCLC patients with first-line EGFR-TKI resistance who underwent tissue rebiopsy with 30% having inadequate samples. Rebiopsies were also a source of morbidity in a prospective study[22] that enrolled 113 NSCLC patients who underwent lung biopsy initially and were

Table 2 Results of Rebiopsy According to the Initial Molecular Profile and Therapeutic Sequence

A. Only 1 line of tyrosine kinase inhibitor.

Patient	Re biopsy	Site	Treatement	Exon 18	exon19	L858R			
1	Solid	Lung	Gefitinib				HRAS	RET	
2	Solid	Lung	Gefitinib						
3	Solid	Lung	Gefitinib				TP53		
4	Solid	node	Gefitinib						
5	Liquid	x	Gefitinib						
6	Both	Lung	Gefitinib						
7	Solid	node	Erlotinib						
8	Solid	Lung	Erlotinib						
9	Both	Lung	Erlotinib						
10	Both	Lung	Erlotinib						
11	Solid	Liver	Erlotinib						
12	Both	plevra	Afatinib						
13	Solid	Lung	Afatinib						
14	Solid	Lung	Afatinib						
15	Solid	Plevra	Afatinib				TP53		
16	Sol Liquid id	Lung	Osimertinib				TP53		
17	Liquid	x	Osimertinib						
18	Liquid	x	Osimertinib						
19	Solid	Lung	Osimertinib						
20	Both	Lung	Osimertinib						
21	Both	Lung	Osimertinib				TP53		
22	Liquid	x	Osimertinib				TP53		
23	Solid	Bone	Osimertinib				Amp MET		
24	Liquid	x	Osimertinib						
25	Liquid	x	Osimertinib				Amp MET		
26	Both	Lung	Osimertinib				ROS1	HER2	
27	Solid	Node	Osimertinib						
28	Solid	Lung	Osimertinib				PIK3A		
29	Both	Lung	Osimertinib						
30	Solid	Lung	Osimertinib						
31	Solid	Plevra	Osimertinib						
32	Both	ganglions	Osimertinib				TP53	TET2	CTMNB1
33	Both	Node	Osimertinib				TP53		
34	Solid	Lung	Osimertinib						
35	Solid	Lung	Osimertinib						
36	Liquid	x	Osimertinib				TP53	CTMNB1	BRCA2
37	Solid	Lung	Osimertinib						
38	Solid	Lung	Osimertinib				PIK3A	KRAS	NRAS
39	Solid	Lung	Osimertinib				STK11		
40	Solid	Lung	Osimertinib						
41	LIquid	x	Osimertinib				PTEN	APC	
42	Liquid	x	Osimertinib				TP53	STK11	
43	Liquid	x	Osimertinib				STK11		
44	Liquid	x	Osimertinib						
45	Solid	Lung	Osimertinib				TP53		
48	LIquid	x	Osimertinib						

(*continued on next page*)

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https:

Table 2 (*continued*)

B. Two lines or more.

Patient	Biopsy	Site	Line 1	Line 2	Line 3	Exon 18	exon19	L858R	T790M		
1	Liquid	x	Erlotinib	Osimertinib							
2	Solid	Lung	Gefitinib	Osimertinib						Amp MET	
3	Solid	Node	Afatinib	Osimertinib						C797S	
4	Solid	Breast	Gefitinib	Osimertinib						TP53	
5	Liquid	x	Afatinib	Osimertinib							
6	Solid	Lung	Erlotinib	Osimertinib	Erlotinib					Exon 20ins	CTMNB1
7	Both	Liver	Gefitinib	Osimertinib							
8	Both	Bone	Erlotinib	Osimertinib							
9	Both	Lung	Erlotinib	Osimertinib						PIK3A	
10	Solid	Spinal fluid	Gefitinib	Osimertinib						TP53	
11	Both	Lung	Gefitinib	Osimertinib							
12	Both	plevra	Afatinib	Erlotinib	Osimertinib					C797S	
13	Liquid	x	Erlotinib	Osimertinib						TP53	
14	Liquid	x	Erlotinib	Osimertinib							
15	Solid	Bone	Erlotinib	Osimertinib						TP53	
16	Both	Bone	Erlotinib	Erlotinib						TP53	
17	Liquid	x	Afatinib	Osimertinib							
18	Liquid	x	Erlotinib	Osimertinib						Amp MET	
19	Solid	Lung	Erlotinib	Afatinib	Osimertinib					STK11	
20	Solid	Lung	Erlotinib	Osimertinib							
21	Liquid	x	Afatinib	Gefitinib							
22	Solid	Node	Afatinib	Osimertinib						Amp MET	
23	Liquid	x	Erlotinib	Osimertinib							
24	Liquid	x	Osimertinib	Capmatinib							
25	Solid	Pleuvra	Erlotinib	Osimertinib						Exon 20ins	
26	Solid	Lung	Osimertinib	Crizotinib						TP53	ROS1
27	Liquid	x	Erlotinib	Osimertinib						PIK3A	
28	Liquid	x	Gefitinib	Osimertinib					C797S		

APC, adenomatous polyposis coli; BRCA2, breast cancer-2; CTNNB1, β-catenin;
HER2, human epidermal growth factor receptor-2; HRAS, Harvey rat sarcoma;
KRAS, Kirsten rat-sarcoma viral oncogene;
MET amp, mesenchymal-to-epithelial transition amplification;
NRAS, neuroblastoma rat-sarcoma viral oncogene; PBRM1, polybromo-1;
PIKC3A, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit-alpha;
PTEN, phosphatase and TENsin homolog; tumor-suppressor gene;
RET, rearranged-during-transfection translocation; ROS1, proto-oncogene tyrosine-protein kinase-1;
STK11, serine/threonine kinase-11; TET2, ten-eleven-translocation; TP53, tumor protein-53.

Pre existing	New mut, Solid biopsy	New Mut, Liquid biospy	Mut disapeared

rebiopsied after progression on EGFR-TKIs and/or chemotherapy, with respective complication rates of 22.1% and 32.7%. The pulmonary hemorrhage frequency rose from 7.1% initially to 10.6% at rebiopsy, while that of pneumothorax increased from 14.2% to 20.4%. Compared to the initial biopsy, the respective overall complication, parenchymal hemorrhage and pneumothorax rates rose by 10.6%, 3.5% and 6.2%. Their multivariate logistic-regression analysis retained male sex, tumor size $\leq$ 2 cm, combination EGFR-TKI–chemotherapy and trans-fissural approach as independent risk factors for overall complications after rebiopsy. The discussion of the indication for these re-biopsies in a multidisciplinary board meeting, the careful choice of the technique and the site of biopsy according to progression modalities and the wider use of liquid biopsies should reduce the morbidity of these re-biopsies. At the time of the study, access to liquid biopsy was limited in most centers, making comparison between tissue and liquid biopsy difficult. While the patterns of resistance mutations and other alterations reported in this study are consistent with existing work, the clinical impact of these rebiopsies is difficult to establish. It depends on treatment accessibility, which varies according to country. The appearance of a targetable alteration, known or a new oncogene mutation, theoretically guides the therapeutic choices. That is particularly true when the tumor progresses under first- or second-generation EGFR-TKIs, with the most frequently appearing resistance mechanism being *T790M,* for which third-generation TKI efficacy has been documented.[25]

Acquired resistance to all 3 EGFR-TKI generations by activation of bypass signaling through the selection of *MET* amplification has also been clinically validated. That amplification was effectively targeted in prospective clinical studies combining EGFR- and MET-TKI.[2,25] In fact, rebiopsy molecular-profile changes outside the main oncogenic driver showed greater variety with a specific spectrum for some molecular subclasses and statistical noise for others. Among the more frequently observed acquisitions were tumor protein 53 (*TP53*) mutations, high-level *MET* or membrane-bound Erb-B2–receptor tyrosine kinase-3 *EGFR* family of receptor tyrosine kinase-2 (*ERBB2*) amplifications. The loss of previously detected, accompanying alterations, such as *TP53* mutations,

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https://doi.org/10.1016/j.cllc.2025.09.009

intermediate-level *MET* amplifications or phosphatase and TENsin homolog (*PTEN*) alterations, was also a common phenomenon. However, for the time being, the clinical and therapeutic repercussions of this modified molecular profile are limited, especially because interpretation of this new information is sometimes difficult.

Some alterations, such as *MET* and *ERBB2* amplifications, are well-known resistance mechanisms of EGFR-targeted treatment. Others should be considered putative resistance mechanisms still lacking convincing clinical validation (acquisition of β-catenin (*CTNNB1*), phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha (*PIK3CA*) or *PTEN* mutations). *TP53*-mutation acquisition is a common phenomenon. Indeed, it apparently is an epiphenomenon of resistance to EGFR-TKIs. The loss of a preexisting *TP53* mutation that we observed in some patients could reflect technical problems or subclonal evolution.[26,27] In the future, there will be more therapeutic options, whether combinations of targeted therapy, such as the association of savolitinib or tepotinib with osimertinib[28,29] or new drugs such datopotamab- deruxtecan, which already encouraging antitumor activity in heavily pretreated EGFR NSCLC patients.[30] The situation is also complex for patients with *ALK* or *ROS* translocations.[10,27] According to a real-world analysis of rebiopsies obtained from patients with *ALK*-harboring NSCLCs progressing on second-generation anti-ALK-TKIs, the dominant resistance mechanism was secondary mutation in the *ALK* domain (56.8%, 25/44), with *G1202R* being the most frequent (27.2%, 12/44), but many other potential resistance mechanisms were also identified: *MET* amplification, A-kinase anchoring protein-9–v-*RAF* murine sarcoma viral oncogene homolog B (*AKAP9–BRAF*) fusion, $BRAF^{V600E}$ mutation, or Kirsten rat-sarcoma viral oncogene (*KRAS*) amplification and $KRAS^{G12A}$ mutation.[10]

Our study has several limitations, primarily its retrospective design, nonstandardization of the molecular analyses run according to each center's local practices. Furthermore, for a given center, the period between initial diagnosis and progression is sometimes long, so the tests may have evolved, generally with in-depth research. Likewise, liquid biopsy uses the same targets than tissue for a given center, but again, there may be differences between centers. In addition, the first-line therapeutic strategies relied, as much for patients with *EGFR* mutations as those with *ALK* or *ROS* translocations, on last-generation TKIs. It must also be emphasized that test performances have evolved and more relevant information is probably obtained today Finally, this is a selected population, remaining at progression in a performans status which allows platinum-based chemotherapy to be considered. In conclusion, this analysis of practices showed that more than half of the metastatic NSCLCs harboring *EGFR, ALK* or *ROS* oncogenic anomalies that progressed on TKIs were rebiopsied. Those new samples provided new information compared to the initial biopsies for 40.3% of them but with no impact on median OS according to rebiopsy status.

Disclosure

Personal fees, grants, and nonfinancial support: G Radj, AstraZeneca, Roche, Bristol-Myers Squibb, Merck Sharp & Dohme, and Janssen, C Ricordel, AstraZeneca, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Takeda, Laurence Bigay Gamé, MSD, BMS, Astra Zeneca, Pfizer, Amgen, Roche, Janssen Cilag, M Geier, MSD, BMS, Astra Zeneca, Pfizer, Amgen, Roche, Janssen Cilag, Chantal Decroisette, MSD, BMS, Astra Zeneca, Novocure, Pfizer, Amgen, Roche, Regeneron, Takeda, Janssen Cilag, Florian Guisier, AstraZeneca, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Takeda, A Swalduz, AstraZeneca, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Takeda, R Veillon, AstraZeneca, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Takeda, C Chouaïd, Chugai, Daïchi-Sankyo, and Abbvie AstraZeneca, Boehringer Ingelheim, GSK, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Lilly, Novartis, Pfizer, Takeda, Bayer, and Amgen,L Greillier, AstraZeneca, Boehringer Ingelheim, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Lilly, Novartis, Pfizer, Takeda, Bayer, and Amgen, O Bylicki, AstraZeneca, Roche, Sanofi Aventis, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Takeda. A Bronstein, I Monnet, Catherine Daniel, AC Toffart, H Doubre, JM Peloni, D Arpin,H Morel, B Clarisse declare no conflict or interest.

CRediT authorship contribution statement

Antoine Bronstein: Formal analysis, Data curation, Conceptualization. **Hubert Curcio:** Formal analysis, Data curation, Conceptualization. **Isabelle Monnet:** Formal analysis, Data curation, Conceptualization. **Charles Ricordel:** Investigation, Formal analysis, Data curation. **Laurence Bigay-Game:** Investigation, Formal analysis. **Margaux Geier:** Investigation, Formal analysis, Data curation. **Chantal Decroisette:** Investigation, Funding acquisition. **Catherine Daniel:** Investigation, Formal analysis, Data curation. **Florian Guisier:** Funding acquisition, Formal analysis, Data curation. **Aurélie Swalduz:** Validation, Investigation, Formal analysis. **Anne Claire Toffart:** Validation, Investigation. **Hélène Doubre:** Investigation, Formal analysis, Data curation. **Jean Michel Peloni:** Investigation, Data curation. **Dominique Arpin:** Formal analysis, Data curation. **Hugues Morel:** Formal analysis, Data curation. **Remi Veillon:** Investigation, Formal analysis, Data curation. **Benedicte Clarisse:** Methodology. **Christos Chouaïd:** Writing – review & editing, Writing – original draft, Data curation, Conceptualization. **Laurent Greillier:** Project administration, Methodology, Funding acquisition, Formal analysis, Conceptualization. **Olivier Bylicki:** Writing – original draft, Validation, Supervision, Project administration, Methodology, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Acknowledgments

This research received grant from GFPC, an academic group. Academic grant from JANSSEN–CILAG.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.cllc.2025.08.009.

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https://doi.org/10.1016/j.cllc.2025.08.009

References

1. Suay G, Aparisi F, Juan-Vidal O. European Society of Medical Oncology (ESMO) guidelines for oncogene-addicted metastatic non-small cell lung cancer (NSCLC): a personalized treatment for each patient. *Chin Clin Oncol*. 2024;13:46.
2. Rosell R, Moran T, Queraltet C, et al. Screening for epidermal growth factor receptor mutations in lung cancer. *N Engl J Med*. 2009;361:958–967.
3. Soria JC, Ohe Y, Vansteenkiste J, et al. Osimertinib in untreated EGFR-mutated advanced non-small-cell lung cancer. *N Engl J Med*. 2018;378:113–125.
4. Ramalingam SS, Vansteenkiste J, Planchard D, et al. Overall survival with osimertinib in untreated, EGFR-mutated advanced NSCLC. *N Engl J Med*. 2020;382:41–50.
5. La Cava G, Cortellini A, Rotow JK, Bauman JR. Navigating first-line treatment options for patients with epidermal growth factor receptor-positive non-small cell lung cancer. *Am Soc Clin Oncol Educ Book*. 2025;45(3):e472784.
6. Schneider JL, Lin JJ, Shaw AT. ALK-positive lung cancer: a moving target. *Nat Cancer*. 2023;4:330–343.
7. Solomon BJ, Liu G, Felip E, et al. Lorlatinib versus Crizotinib in patients with advanced ALK-positive non-small cell lung cancer: 5-year outcomes from the phase III CROWN study. *J Clin Oncol*. 2024;42:3400–3409.
8. Mok T, Camidge DR, Gadgeel SM, et al. Updated overall survival and final progression-free survival data for patients with treatment-naive advanced ALK-positive non-small-cell lung cancer in the ALEX study. *Ann Oncol*. 2020;31:1056–1064.
9. Ahn MJ, Kim HR, Yang JCH, et al. Efficacy and safety of Brigatinib compared with Crizotinib in Asian vs. Non-Asian patients with locally advanced or metastatic ALK-inhibitor-naive ALK+ Non-small cell lung cancer: final results from the phase III ALTA-1L study. *Clin Lung Cancer*. 2022;23:720–730.
10. Wu L, Zou Z, Li Y, et al. Progression patterns, resistant mechanisms and subsequent therapy for ALK-positive NSCLC in the era of second-generation ALK-TKIs. *J Transl Med*. 2024;22:585.
11. Gendarme S, Bylicki O, Chouaid C, Guisier F. ROS-1 fusions in non-small-cell lung cancer: evidence to date. *Curr Oncol*. 2022;29:641–658.
12. Leonetti A, Sharma S, Minari R, Perego P, Giovannetti E, Tiseo M. Resistance mechanisms to osimertinib in EGFR-mutated non-small cell lung cancer. *Br J Cancer*. 2019;121:725–737.
13. Schmid S, Li JNJ, Leighl NB. Mechanisms of osimertinib resistance and emerging treatment options. *Lung Cancer*. 2020;147:123–129.
14. Papadimitrakopoulou V, Wu YL, Han JY, et al. Analysis of resistance mechanisms to osimertinib in patients with EGFR T790M advanced NSCLC from the AURA3 study. *Ann Onco*. 2018;29:viii741.
15. Oxnard GR, Hu Y, Mileham KF, et al. Assessment of resistance mechanisms and clinical implications in patients with EGFR T790M-positive lung cancer and acquired resistance to osimertinib. *JAMA Oncol*. 2018;4:1527–1534.
16. Ramalingam SS, Cheng Y, Zhou C, et al. Mechanisms of acquired resistance to first-line osimertinib: preliminary data from the phase III FLAURA study. *Ann Oncol*. 2018;29:viii740.
17. Swalduz A, Souquet PJ, Pérol M, et al. Compliance to regional recommendations for molecular analyses and management of advanced lung cancer patients. *Future Oncol*. 2019;15 213–149.
18. Bosc C, Ferretti GR, Cadranel J, et al. Rebiopsy during disease progression in patients treated by TKI for oncogene-addicted NSCLC. *Target Oncol*. 2015;10:247–253.
19. Chouaid C, Dujon C, Do P, et al. Feasibility and clinical impact of re-biopsy in advanced non small-cell lung cancer: a prospective multicenter study in a real-world setting (GFPC study 12-01). *Lung Cancer*. 2014;86:170–173.
20. Piotrowska Z, Ahn MJ, Pang YP, et al. A multicentre, molecular profiling study of patients (pts) with epidermal growth factor receptor-mutated (EGFRm) advanced NSCLC treated with first-line osimertinib. *Ann Oncol*. 2022;33:1420–1421.
21. Wang Y, Zhang Y, Ren N, et al. Repeat biopsy versus initial biopsy in terms of complication risk factors and clinical outcomes for patients with non-small cell lung cancer: a comparative study of 113 CT-guided needle biopsy of lung lesions. *Front Oncol*. 2024;14:1367603.
22. Lin YT, Ho CC, Hsu WH, et al. Tissue or liquid rebiopsy? A prospective study for simultaneous tissue and liquid NGS after first-line EGFR inhibitor resistance in lung cancer. *Cancer Med*. 2023;13:e6870.
23. Scheffler M, Wiesweg M, Michels S, et al. Rebiopsy in advanced non-small cell lung cancer, clinical relevance and prognostic implications. *Lung Cancer*. 2022;168:10–20.
24. Bylicki O, Tomasini P, Radj G, et al. Atezolizumab with or without bevacizumab and platinum-pemetrexed in patients with stage IIIB/IV non-squamous non-small cell lung cancer with EGFR mutation, ALK rearrangement or ROS1 fusion progressing after targeted therapies: a multicentre phase II open-label non-randomised study GFPC 06-2018. *Eur J Cancer*. 2023;83:38–48.
25. Gomatou G, Syrigos N, Kotteas E. Osimertinib resistance: molecular mechanisms and emerging treatment options. *Cancers*. 2023;15:841.
26. Schoenfeld AJ, Chan JM, Rizvi H, et al. Tissue-based molecular and histological landscape of acquired resistance to osimertinib given initially or at relapse in patients with EGFR-mutant lung cancers. *J Clin Oncol*. 2019;37:9028.
27. Rotow J, Bivona TG. Understanding and targeting resistance mechanisms in NSCLC. *Nat Rev Cancer*. 2017;17:637–658.
28. Ahn MJ, De Marinis F, Bonanno L, et al. Biomarker-based preliminary efficacy analysis in SAVANNAH: savolitinib+osimertinib in EGFRm NSCLC Post-Osimertinib. *J Thoracic Oncol*. 2022;7:S469–S470.
29. Mazieres J, Kim TM, Lim BK, et al. Tepotinib + osimertinib for EGFRm NSCLC with MET amplification (METamp) after progression on first-line (1L) osimertinib: initial results from the INSIGHT 2 study. *Ann Oncol*. 2022;33:S1419–S1420.
30. Kitazono S, Paz-Ares L, Ahn MJ, et al. TROPION-Lung05: datopotamab deruxtecan (Dato-DXd) in previously treated non-small cell lung cancer (NSCLC) with actionable genomic alterations. *(AGAs) Ann Onco*. 2022;34:S1671–S1672.

Please cite this article as: Antoine Bronstein et al, Rebiopsy Feasibility and Clinical Impact on Metastatic Non-Small-Cell Lung Cancer With EGFR/ALK/ROS Oncogenic Driver Progression After Optimal Targeted Therapy: A Multicenter Real-World Analysis, Clinical Lung Cancer, https://doi.org/10.1016/j.cllc.2025.09.009