ORIGINAL ARTICLE

RELAY, Erlotinib Plus Ramucirumab in Untreated, *EGFR*-Mutated, Metastatic NSCLC: Outcomes by *EGFR* Exon 19 Deletion Variants

Kazumi Nishino, MD, PhD,[a,*] Jin-Yuan Shih, MD, PhD,[b] Kazuhiko Nakagawa, MD, PhD,[c] Martin Reck, MD, PhD,[d] Edward B. Garon, MD,[e] Michelle Carlsen, MS,[f] Tomoko Matsui,[g] Carla Visseren-Grul, MD,[h] Ernest Nadal, MD, PhD[i]

[a]*Department of Thoracic Oncology, Osaka International Cancer Institute, Osaka, Japan*
[b]*Department of Internal Medicine, National Taiwan University Hospital, Taipei City, Taiwan*
[c]*Department of Medical Oncology, Kindai University Faculty of Medicine, Osaka, Japan*
[d]*Lung Clinic, Airway Research Center North, German Center of Lung Research, Grosshansdorf, Germany*
[e]*Department of Medicine, David Geffen School of Medicine at UCLA/TRIO-US Network, Los Angeles, California*
[f]*Eli Lilly and Company, Indianapolis, Indiana*
[g]*Eli Lilly Japan K.K., Kobe, Japan*
[h]*Eli Lilly Netherlands, Utrecht, The Netherlands*
[i]*Department of Medical Oncology, Catalan Institute of Oncology (ICO), and Preclinical and Experimental Research Group in Thoracic Tumors, Oncobell, l'Institut d'Investigació Biomèdica de Bellvitge (IDIBELL), L'Hospitalet, Barcelona, Spain*

Received 8 September 2023; revised 4 December 2023; accepted 15 December 2023
Available online - 19 December 2023

ABSTRACT

Introduction: *EGFR* gene mutations are drivers of NSCLC. The RELAY double-blind, placebo (PBO)-controlled phase 3 study revealed superior progression-free survival (PFS) for ramucirumab plus erlotinib (RAM + ERL) versus PBO (PBO + ERL) in patients with untreated advanced NSCLC and an *EGFR*-activating mutation. This exploratory analysis evaluated potential associations between *EGFR* exon 19 deletion (ex19del) variants and clinical outcomes.

Methods: Patients (N = 449) were randomized (1:1) to RAM plus ERL or PBO plus ERL. Plasma samples were collected at baseline, on treatment, and at 30-day post-study treatment discontinuation follow-up. Baseline and treatment-emergent gene alterations were investigated by Guardant360 next-generation sequencing. Patients with a valid baseline plasma sample and ex19del were included (RAM + ERL, n = 62; PBO + ERL, n = 72).

Results: The most common ex19del variant was E746_A750del (67.2%); *EGFR* E746 deletions (E746del) occurred more frequently than L747 deletions (74.6% versus 25.4%, respectively). *TP53* mutations were the most frequently co-occurring baseline gene alterations. With treatment arms combined, median PFS was 18.0 months versus 12.5 months for patients with uncommon (non-E746_A750del, n = 44) versus common (E746_A750del, n = 90) ex19del variants (hazard ratio [HR] = 1.657 [95% confidence interval or CI:1.044–2.630]). Median PFS was longer with RAM plus ERL versus PBO plus ERL for patients with the common (15.2 versus 9.9 mo; HR = 0.564 [95% CI: 0.344–0.926]) and E746del (15.4 versus 9.9 mo; HR = 0.587 [95% CI: 0.363–0.951]) variants. Treatment-emergent post-progression *EGFR* T790M rates were higher in the common versus uncommon and E746del versus L747 deletion subgroups.

Conclusions: RAM plus ERL provides benefit and improves treatment outcomes for patients with metastatic NSCLC with *EGFR* ex19del variants.

***Corresponding author.**

Address for correspondence: Kazumi Nishino, MD, PhD, Department of Thoracic Oncology, Osaka International Cancer Institute, 3-1-69 Otemae Chuo-ku, Osaka, 541-8567, Japan. E-mail: kazumi.nishino@oici.jp

Cite this article as: Nishino K, Shih JY, Nakagawa K, et al. RELAY, erlotinib plus ramucirumab in untreated, *EGFR*-mutated, metastatic NSCLC: outcomes by *EGFR* exon 19 deletion variants. *JTO Clin Res Rep*. 2024;5:100624.

ISSN: 2666-3643

https://doi.org/10.1016/j.jtocrr.2023.100624

Keywords: Carcinoma; Non–small cell lung; EGFR exon 19 deletion variant; Ramucirumab; VEGFR inhibition; Targeted therapy

Introduction

Mutations in the *EGFR* gene occur in approximately 10% to 20% of White and 40% to 60% of Asian patients with NSCLC.[1] The most common *EGFR*-activating mutations are in-frame deletions (dels) of exon 19 (approximately 50%) and an L858R point mutation in exon 21 (approximately 40%).[1,2] The first-line standard of care for *EGFR* mutation-positive NSCLC is EGFR tyrosine kinase inhibitor (TKI) therapy,[3,4] but eventually all patients will develop treatment resistance.[5]

EGFR exon 19 del (ex19del) mutations consist of distinct molecular variants and represent a heterogeneous disease entity.[6,7] The most common ex19del variant is E746_A750del, detected in 63% to 72% of patients with *EGFR* ex19del.[6,8,9] E746_A750del results from the deletion of five amino acids between the third β-strand of the EGFR tyrosine kinase domain and its key regulatory αC-helix in the EGFR protein (Fig. 1*A* and *B*).[7] Other *EGFR* exon 19 variants, referred to as uncommon variants, include dels and or insertions (delins), with the most frequent variants encompassing the amino acids from codons E746 to L747.[6] Distinct *EGFR* ex19del variants have different in vitro sensitivities to different EGFR TKIs.[7,10,11] Furthermore, different clinical outcomes of EGFR TKIs have been associated with distinct *EGFR* ex19del variants[8,9,12–19]; however, conflicting results have been reported. Some studies with EGFR TKIs have revealed improved or comparable efficacy for patients with the common variant (E746_A750del),[13,14,16] whereas other studies have revealed improved or comparable efficacy for patients with uncommon (non-E746_A750del) ex19del variants.[12,15,18] After first-line EGFR TKI therapy, acquired T790M resistance is reported to occur more frequently in patients with the common (E746_A750del) ex19del variant versus uncommon variants.[9,20–22] Osimertinib, as subsequent therapy after first-line EGFR TKI treatment for T790M-positive patients,[23] has been reported to improve progression-free survival (PFS) and overall survival outcomes to a greater extent for T790M-positive patients with the common ex19del variant versus uncommon ex19del variants,[15] and E746del variants versus non-E746del variants.[24] Nevertheless, information on the impact of dual inhibition on the EGFR and vascular endothelial growth factor (VEGF) pathways on outcomes in patients with ex19del variants is lacking. Furthermore, no prospective randomized study evaluating the association of ex19del variants with clinical outcomes has been reported.

The RELAY global, double-blind, placebo (PBO)-controlled phase 3 study revealed superior PFS for ramucirumab (RAM), a human IgG1 VEGF receptor 2 antagonist, plus erlotinib (ERL; RAM + ERL) compared with PBO plus ERL (PBO + ERL) in the first-line treatment of patients with *EGFR* mutation-positive (ex19del or exon 21 L858R), metastatic NSCLC.[25] Median PFS was 19.4 months (95% confidence interval [CI]: 15.4–21.6) with RAM plus ERL versus 12.4 months (95% CI: 11.0–13.5) with PBO plus ERL (hazard ratio [HR] = 0.59, 95% CI: 0.46–0.76, $p < 0.0001$). Median PFS among patients with ex19del was 19.6 months (95% CI: 15.1–22.2) with RAM plus ERL versus 12.5 months (11.1–15.3) with PBO plus ERL; median PFS among patients with an exon 21 L858R mutation was 19.4 months (95% CI: 14.1–21.9) with RAM plus ERL versus 11.2 months (9.6–13.8) with PBO plus ERL.[26] The objective of this post hoc exploratory analysis was to evaluate the potential association between *EGFR* ex19del variants and clinical outcomes using prospective data from the RELAY global phase 3 study.

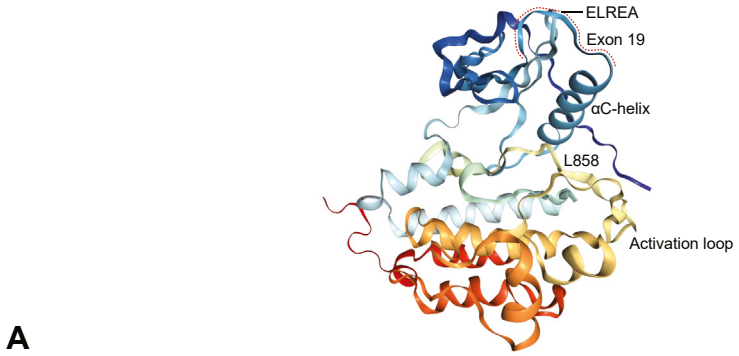
Materials and Methods

Study Design

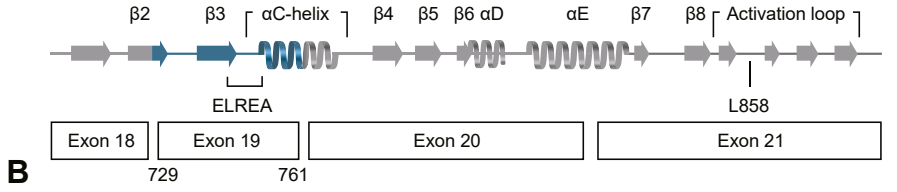
Study design and patient eligibility have been previously published.[25] Briefly, RELAY is a global, double-blind, PBO-controlled phase 3 study of patients with untreated *EGFR*-mutated metastatic NSCLC (ClinicalTrials.gov: NCT02411448). Randomized patients (1:1) received either intravenous RAM (10 mg/kg) or matching PBO every 2 weeks with oral ERL (150 mg) daily.[25] Patients continued therapy until disease progression, unacceptable toxicity, withdrawal of consent, noncompliance, or investigator decision. An exploratory liquid biopsy biomarker study was conducted in the global intent-to-treat (ITT) RELAY population. The study was conducted in accordance with the Declaration of Helsinki, the Council for International Organizations of Medical Sciences International Ethical Guidelines, Good Clinical Practice guidelines, and local guidelines. Local institutional review boards approved the protocol and addendum for each site; all patients provided written informed consent.

Study Population

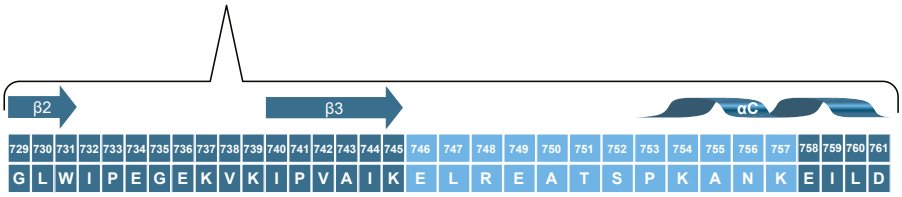
For the RELAY randomized study,[25] eligible patients met the following criteria: stage IV NSCLC; eligible for first-line treatment with ERL on the basis of *EGFR* ex19del or exon 21 L858R mutation; Eastern Cooperative Oncology Group performance status of 0 or 1; measurable disease according to Response Evaluation



A



B



C

	Exon 19 variants, n (%)	Total	RAM + ERL	PBO + ERL
E746del variants		**100 (74.6)**	**43 (69.4)**	**57 (79.2)**
E746_A750del	— — — — — T S P K A N K	90 (67.2)	39 (62.9)	51 (70.8)
E746_P753delinsVS	— — — — — — — V S A N K	1 (0.7)	1 (1.6)	0 (0)
E746_S752delinsA	— — — — — — A P K A N K	1 (0.7)	0 (0)	1 (1.4)
E746_S752delinsV	— — — — — — V P K A N K	5 (3.7)	1 (1.6)	4 (5.6)
E746_T751delinsI	— — — — — I S P K A N K	1 (0.7)	1 (1.6)	0 (0)
E746_T751delinsV	— — — — — V S P K A N K	1 (0.7)	1 (1.6)	0 (0)
E746_T751delinsVA	— — — — — V A P K A N K	1 (0.7)	0 (0)	1 (1.4)
L747del variants		**34 (25.4)**	**19 (30.6)**	**15 (20.8)**
L747_A750delinsP	E — — — P T S P K A N K	9 (6.7)	3 (4.8)	6 (8.3)
L747_A755delins PRD	E — — — — — — — P R D	1 (0.7)	1 (1.6)	0 (0)
L747_P753delinsS	E — — — — — — S K A N K	13 (9.7)	8 (12.9)	5 (6.9)
L747_S752del	E — — — — — — P K A N K	2 (1.5)	1 (1.6)	1 (1.4)
L747_S752delinsQ	E — — — — — Q P K A N K	1 (0.7)	0 (0)	1 (1.4)
L747_T751del	E — — — — — S P K A N K	5 (3.7)	4 (6.5)	1 (1.4)
L747_T751delinsP	E — — — — P S P K A N K	3 (2.2)	2 (3.2)	1 (1.4)

Figure 1. Structure of the EGFR protein and baseline *EGFR* ex19del variants in the RELAY TR population. (*A*) EGFR protein structure. (*B*) Schematic representation of the β2-activation loop encoded by exons 18, 19, 20, and 21. (*C*) Multiple sequence alignment of *EGFR* wild-type and *EGFR* ex19del variants identified in the RELAY TR population. TR population comprised patients in the RELAY ITT population with a valid baseline plasma sample with an ex19del variant detected. ATP, adenosine triphosphate; del, deletion; delins, deletion and insertion; E746del, E746 deletion; ERL, erlotinib; ex19del, exon 19 deletion; ITT, intent-to-treat; L747del, L747 deletion; PBO, placebo; RAM, ramucirumab; TR, translational research.EGFR protein image (part A) created with the Protein Data Bank (https://www.rcsb.org/3d-view/ngl/1m17) ID: IM17 and associated publication, NGL Viewer (Rose AS, et al. NGL Viewer: web-based molecular graphics for large complexes. *Bioinformatics*. 2018;34:3755-3758. https://doi.org/10.1093/bioinformatics/bty419), and RCSB PDB.

Criteria in Solid Tumours version 1.1; and adequate hematologic and organ function with a urinary protein less than or equal to 1+ on dipstick or routine urinalysis. Patients were excluded if they had an *EGFR* T790M mutation, central nervous system (CNS) metastases, uncontrolled hypertension, or a history of substantial bleeding.

For this post hoc analysis, the translational research (TR) population comprised patients in the RELAY ITT population with a valid baseline plasma sample (i.e., a

plasma sample that passed quality control) with an *EGFR* ex19del variant detected by Guardant360 next-generation sequencing (NGS) (Guardant Health, Redwood City, CA).

Outcome Measures

The primary end point of the RELAY randomized portion was PFS according to Response Evaluation Criteria in Solid Tumours version 1.1. PFS was defined as the time from randomization to disease progression or death from any cause. Secondary end points included objective response rate (ORR), disease control rate (DCR), and duration of response (DoR) as previously reported.[25] Exploratory end points of this post hoc analysis included the association of ex19del variants with patient and disease characteristics, co-occurring gene alterations at baseline, clinical outcomes (PFS, ORR, DCR, and DoR), and treatment-emergent gene alterations at disease progression. Adverse events (AEs) were graded using the National Cancer Institute Common Terminology Criteria for Adverse Events version 4.0.

EGFR Mutation and Co-Occurring Gene Alteration Evaluation

For RELAY, patients were included if they had documented evidence of a tumor that had an ex19del or an exon 21 L858R substitution mutation by local testing of tumor tissue and preplanned confirmatory central *EGFR* testing had been conducted. Plasma samples were collected before the first dose of the study drug, on day 1 of cycle 4, and at the 30-day treatment discontinuation follow-up. Gene alterations co-occurring with an *EGFR* ex19del variant were assessed in plasma by Guardant360 NGS.

EGFR Ex19del Subgroups

For this post hoc analysis, patients were divided into two pairs of subgroups according to their *EGFR* ex19del variant: (1) common (E746_A750del variant) versus uncommon (non-E746_A750del ex19del variants); and (2) E746del (ex19del mutations starting from codon E746) versus L747del (ex19del mutations starting from codon L747).

Statistical Analysis

Data cutoff was January 23, 2019. The TR population comprised all patients who received at least one dose of the study treatment and had a valid baseline plasma sample with an *EGFR* ex19del variant. Baseline gene alterations are reported for patients in the TR population with a valid baseline plasma sample with an ex19del variant detected. Treatment-emergent gene alterations are reported for patients with a valid plasma sample with an ex19del variant detected at baseline and a post-progression 30-day follow-up plasma sample with any alteration detected. The Kaplan-Meier method was used to generate curves and estimate median PFS and DoR. A Cox proportional hazards regression model was used to estimate HRs with 95% CIs for PFS and DoR. The Wilson score interval method was used to calculate 95% CIs for ORR and DCR. p values for comparison between patient subgroups were calculated using the likelihood-ratio chi-square test. All analyses were conducted for each patient subgroup (common versus uncommon; E746del versus L747del) and by treatment arm (RAM + ERL; PBO + ERL). Number and percentage of patients in each subgroup who received post-treatment discontinuation osimertinib treatment as any subsequent therapy are summarized. No statistical comparisons were made between the ex19del variants within the treatment arms. Safety was summarized as the number and percentage of patients reporting each AE by subgroup and treatment arm. Analyses were conducted using SAS 9.4 (SAS Institute Inc., Cary, NC) and R 4.1.2 (R Core Team, R Foundation for Statistical Computing, Vienna, Austria).

Results

Demographic and Baseline Clinical Characteristics

The RELAY ITT population comprised 449 patients (RAM + ERL, n = 224; PBO + ERL, n = 225); 243 patients (54.1%) had an ex19del mutation on the basis of local testing.[25] Of the 449 patients, 390 (86.9%) had assessable baseline plasma Guardant360 NGS results. The TR population for this post hoc analysis comprised 134 patients with a valid baseline plasma sample positive for an *EGFR* ex19del variant (common, n = 90; uncommon, n = 44; E746del, n = 100; L747del, n = 34; Supplementary Fig. 1).

In the TR population, the most common ex19del variant was E746_A750del occurring in 90 patients (67.2%); the second most common variant was L747_P753delinsS occurring in 13 patients (9.7%; Fig. 1*C*). E746_A750del occurred at a lower frequency (62.9% versus 70.8%, respectively) and L747_P753delinsS occurred at a higher frequency (12.9% versus 6.9%, respectively) in the RAM plus ERL arm compared with the PBO plus ERL arm (Fig. 1*C*). In the TR population, dels starting with E746 occurred in 100 patients (74.6%) and dels starting from L747 occurred in 34 patients (25.4%). E746del variants occurred at a lower frequency (69.4% versus 79.2%, respectively) and

L747del variants occurred at a higher frequency (30.6% versus 20.8%, respectively) in the RAM plus ERL arm compared with the PBO plus ERL arm (Fig. 1*C*).

Patient and disease characteristics at baseline were comparable between the common and uncommon subgroups and between the E746del and L747del subgroups, with the exception of smoking history. Smoking history (ever smokers, never smokers, and unknown) was statistically significantly different between the common versus uncommon subgroups ($p = 0.0101$; Supplementary Table 1) and numerically different between E746del versus L747del subgroups ($p = 0.0761$; Supplementary Table 1), with the proportion of never smokers being greater in the uncommon versus common subgroup (75.0% versus 52.2%, respectively) and in the L747del versus E746del subgroup (73.5% versus 55.0%, respectively; Supplementary Table 1).

Within the RAM plus ERL arm, patient and disease characteristics at baseline were comparable and no statistically significant differences were observed between the common and uncommon subgroups and between the E746del and L747del subgroups (Table 1). Within the PBO plus ERL arm, smoking history was statistically significantly different between the common versus uncommon subgroups ($p = 0.0007$) and E746del versus L747del subgroups ($p = 0.0039$), and the distribution of sex was significantly different between the common versus uncommon subgroups ($p = 0.0148$) and E746del versus L747del subgroups ($p = 0.0112$; Table 1). No other statistically significant differences were observed between the subgroups.

Baseline Co-Occurring Gene Alterations

TP53 was the most common co-occurring gene alteration at baseline in all ex19del patient subgroups (Table 2 and Supplementary Fig. 2): common subgroup: RAM plus ERL, 21 of 39 patients, PBO plus ERL, 37 of 51 patients; uncommon subgroup: RAM plus ERL, 14 of 23 patients, PBO plus ERL, 12 of 21 patients; E746del subgroup: RAM plus ERL, 23 of 43 patients, PBO plus ERL, 38 of 57 patients; and L747del subgroup: RAM plus ERL, 12 of 19 patients, PBO plus ERL, 11 of 15 patients. Apparent differences in co-occurring gene alterations at baseline in the common subgroup were *BRCA2* (RAM + ERL, three of 39 patients; PBO + ERL, zero of 51 patients) and *ESR1* (RAM + ERL, zero of 39 patients; PBO + ERL, four of 51 patients), and in the uncommon subgroup was *NF1* (RAM + ERL, three of 23 patients; PBO + ERL, zero of 21 patients); in the E746del subgroup were *AR* (RAM + ERL, three of 43 patients; PBO + ERL, zero of 57 patients), *BRCA2* (RAM + ERL, three of 43 patients; PBO + ERL, zero of 57 patients), *DDR2* (RAM + ERL, three of 43 patients; PBO + ERL, zero of 57 patients), and *ESR1* (RAM + ERL, zero of 43 patients; PBO + ERL, four of 57 patients); and in the L747del subgroup was *CDK6* (RAM + ERL, four of 19 patients; PBO + ERL, zero of 15 patients).

Efficacy

Overall median PFS (RAM + ERL and PBO + ERL arms combined) was 18.0 months versus 12.5 months for patients in the uncommon subgroup versus the common subgroup (HR = 1.657, 95% CI: 1.044–2.630; Fig. 2*A*) and 15.1 months versus 12.5 months for patients in the L747del subgroup versus the E746del subgroup (HR = 1.348, 95% CI: 0.825–2.202; Fig. 2*B*). By treatment arm, median PFS was longer in the common and E746del subgroups in the RAM plus ERL arm versus the PBO plus ERL arm (Fig. 2*C* and *D*). Median PFS (RAM + ERL versus PBO + ERL, respectively) was 15.2 months versus 9.9 months (HR = 0.564, 95% CI: 0.344–0.926) for the common subgroup and 19.4 months versus 13.9 months (HR = 0.654, 95% CI: 0.282–1.515) for the uncommon subgroup (Fig. 2*C*); 15.4 months versus 9.9 months (HR = 0.587, 95% CI: 0.363–0.951) for the E746del subgroup and 18.0 months versus 12.5 months (HR = 0.605, 95% CI: 0.246–1.489) for the L747del subgroup (Fig. 2*D*).

ORR and DCR were greater than 80% in each treatment arm for each ex19del variant subgroup (Table 3). Median DoR (RAM + ERL versus PBO + ERL, respectively) was 14.1 months versus 8.4 months (HR = 0.618, 95% CI: 0.369–1.035) in the common subgroup and 13.8 months versus 11.3 months (HR = 0.693, 95% CI: 0.305–1.574) in the uncommon subgroup (Fig. 2*E*); and 14.1 months versus 9.6 months (HR = 0.695, 95% CI: 0.424–1.141) in the E746del subgroup and 13.8 months versus 11.0 months (HR = 0.515, 95% CI: 0.208–1.273) in the L747del subgroup (Fig. 2*F*).

Treatment-Emergent Gene Alterations After Disease Progression

On disease progression, treatment-emergent *EGFR* T790M mutations were more frequent in the common subgroup (43.8%; 21 of 48 patients [RAM + ERL, six of 15 patients; PBO + ERL, 15 of 33 patients]) versus the uncommon subgroup (21.4%; three of 14 patients [RAM + ERL, one of five patients; PBO + ERL, two of nine patients]), and in the E746del subgroup (42.0%; 21 of 50 patients [RAM + ERL, six of 15 patients; PBO + ERL, 15 of 35 patients]) versus the L747del subgroup (25.0%; three of 12 patients [RAM + ERL, one of five patients; PBO + ERL, two of seven patients]), regardless of treatment arm (Table 4). *TP53* mutations were observed in the common subgroup but not in the

Table 1. Baseline Patient Demographics and Characteristics by *EGFR* Ex19del Subgroup and Treatment Arm (TR Population)

	Common (n = 90)		Uncommon (n = 44)		E746del (n = 100)		L747del (n = 34)	
Characteristics, n (%)	RAM + ERL (n = 39)	PBO + ERL (n = 51)	RAM + ERL (n = 23)	PBO + ERL (n = 21)	RAM + ERL (n = 43)	PBO + ERL (n = 57)	RAM + ERL (n = 19)	PBO + ERL (n = 15)
Sex[a]								
Female	24 (61.5)	26 (51.0)	10 (43.5)	17 (81.0)	26 (60.5)	30 (52.6)	8 (42.1)	13 (86.7)
Male	15 (38.5)	25 (49.0)	13 (56.5)	4 (19.0)	17 (39.5)	27 (47.4)	11 (57.9)	2 (13.3)
Age, y								
Median (range)	59 (27-83)	62 (23-82)	60 (41-73)	63 (35-73)	62 (27-83)	62 (23-82)	60 (41-73)	63 (35-73)
<65	24 (61.5)	31 (60.8)	16 (69.6)	13 (61.9)	26 (60.5)	35 (61.4)	14 (73.7)	9 (60.0)
≤65	15 (38.5)	20 (39.2)	7 (30.4)	8 (38.1)	17 (39.5)	22 (38.6)	5 (26.3)	6 (40.0)
Race[b]								
Asian	28 (71.8)	38 (74.5)	18 (78.3)	16 (76.2)	31 (72.1)	41 (71.9)	15 (78.9)	13 (86.7)
White	11 (28.2)	13 (25.5)	5 (21.7)	4 (19.0)	12 (27.9)	16 (28.1)	4 (21.1)	1 (6.7)
Missing	0	0	0	1 (4.8)	0	0	0	1 (6.7)
Smoking history[c]								
Ever	14 (35.9)	23 (45.1)	6 (26.1)	1 (4.8)	15 (34.9)	23 (40.4)	5 (26.3)	1 (6.7)
Never	23 (59.0)	24 (47.1)	14 (60.9)	19 (90.5)	26 (60.5)	29 (50.9)	11 (57.9)	14 (93.3)
Unknown	2 (5.1)	4 (7.8)	3 (13.0)	1 (4.8)	2 (4.7)	5 (8.8)	3 (15.8)	0
ECOG Performance status								
0	20 (51.3)	30 (58.8)	14 (60.9)	9 (42.9)	21 (48.8)	31 (54.4)	13 (68.4)	8 (53.3)
1	19 (48.7)	21 (41.2)	9 (39.1)	12 (57.1)	22 (51.2)	26 (45.6)	6 (31.6)	7 (46.7)

Note: TR population comprised patients in the RELAY ITT population with a valid baseline plasma sample with an ex19del variant detected.
[a]Distribution of sex was statistically significantly different between the common versus uncommon subgroups ($p = 0.0148$) and E746del versus L747del subgroups ($p = 0.0112$) within the PBO plus ERL arm.
[b]Race was statistically significantly different between the E746del versus L747del subgroups within the PBO plus ERL arm ($p = 0.0393$).
[c]Within the PBO plus ERL arm, smoking history was statistically significantly different between the common versus uncommon subgroups ($p = 0.0007$) and E746del versus L747del subgroups ($p = 0.0039$).
E746del, E746 deletion; ECOG, Eastern Cooperative Oncology Group; ERL, erlotinib; ex19del, exon 19 deletion; ITT, intent-to-treat; L747del, L747 deletion; PBO, placebo; RAM, ramucirumab; TR, translational research.

Table 2. Baseline Co-Occurring Gene Alterations by *EGFR* Ex19del Subgroup and Treatment Arm (TR Population)

	Common (n = 90)		Uncommon (n = 44)		E746del (n = 100)		L747del (n = 34)	
Genetic Region, n (%)	RAM + ERL (n = 39)	PBO + ERL (n = 51)	RAM + ERL (n = 23)	PBO + ERL (n = 21)	RAM + ERL (n = 43)	PBO + ERL (n = 57)	RAM + ERL (n = 19)	PBO + ERL (n = 15)
APC	2 (5.1)	4 (7.8)	3 (13.0)	2 (9.5)	2 (4.7)	5 (8.8)	3 (15.8)	1 (6.7)
AR	2 (5.1)	0	2 (8.7)	0	3 (7.0)	0	1 (5.3)	0
ARID1A	3 (7.7)	1 (2.0)	0	1 (4.8)	3 (7.0)	1 (1.8)	0	1 (6.7)
BRAF	2 (5.1)	7 (13.7)	4 (17.4)	1 (4.8)	2 (4.7)	7 (12.3)	4 (21.1)	1 (6.7)
BRCA1	1 (2.6)	6 (11.8)	2 (8.7)	1 (4.8)	1 (2.3)	6 (10.5)	2 (10.5)	1 (6.7)
BRCA2	3 (7.7)	0	0	0	3 (7.0)	0	0	0
CDK4	1 (2.6)	4 (7.8)	0	0	1 (2.3)	4 (7.0)	0	0
CDK6	4 (10.3)	3 (5.9)	4 (17.4)	1 (4.8)	4 (9.3)	4 (7.0)	4 (21.1)	0
CTNNB1	6 (15.4)	2 (3.9)	1 (4.3)	2 (9.5)	6 (14.0)	2 (3.5)	1 (5.3)	2 (13.3)
DDR2	2 (5.1)	0	2 (8.7)	0	3 (7.0)	0	1 (5.3)	0
ESR1	0	4 (7.8)	0	0	0	4 (7.0)	0	0
FGFR2	1 (2.6)	3 (5.9)	0	0	1 (2.3)	3 (5.3)	0	0
GNAS	1 (2.6)	1 (2.0)	0	2 (9.5)	1 (2.3)	1 (1.8)	0	2 (13.3)
KIT	0	2 (3.9)	2 (8.7)	0	0	2 (3.5)	2 (10.5)	0
MET	2 (5.1)	2 (3.9)	4 (17.4)	2 (9.5)	2 (4.7)	3 (5.3)	4 (21.1)	1 (6.7)
MTOR	2 (5.1)	1 (2.0)	0	0	2 (4.7)	1 (1.8)	0	0
MYC	2 (5.1)	2 (3.9)	0	2 (9.5)	2 (4.7)	3 (5.3)	0	1 (6.7)
NF1	1 (2.6)	6 (11.8)	3 (13.0)	0	1 (2.3)	6 (10.5)	3 (15.8)	0
NRAS	1 (2.6)	0	0	2 (9.5)	1 (2.3)	1 (1.8)	0	1 (6.7)
PDGFRA	1 (2.6)	3 (5.9)	1 (4.3)	0	1 (2.3)	3 (5.3)	1 (5.3)	0
PIK3CA	8 (20.5)	11 (21.6)	3 (13.0)	2 (9.5)	9 (20.9)	11 (19.3)	2 (10.5)	2 (13.3)
RB1	0	2 (3.9)	2 (8.7)	2 (9.5)	0	3 (5.3)	2 (10.5)	1 (6.7)
SMAD4	2 (5.1)	3 (5.9)	2 (8.7)	2 (9.5)	2 (4.7)	3 (5.3)	2 (10.5)	2 (13.3)
SMO	2 (5.1)	0	0	0	2 (4.7)	0	0	0
TP53	21 (53.8)	37 (72.5)	14 (60.9)	12 (57.1)	23 (53.5)	38 (66.7)	12 (63.2)	11 (73.3)

Note: Occurring in at least 5% of patients with either common or uncommon variants or E746del variants. TR population comprised patients in the RELAY ITT population with a plasma sample with an ex19del variant detected at baseline.

E746del, E746 deletion; ERL, erlotinib; ex19del, exon 19 deletion; ITT, intent-to-treat; L747del, L747 deletion; PBO, placebo; RAM, ramucirumab; TR, translational research.

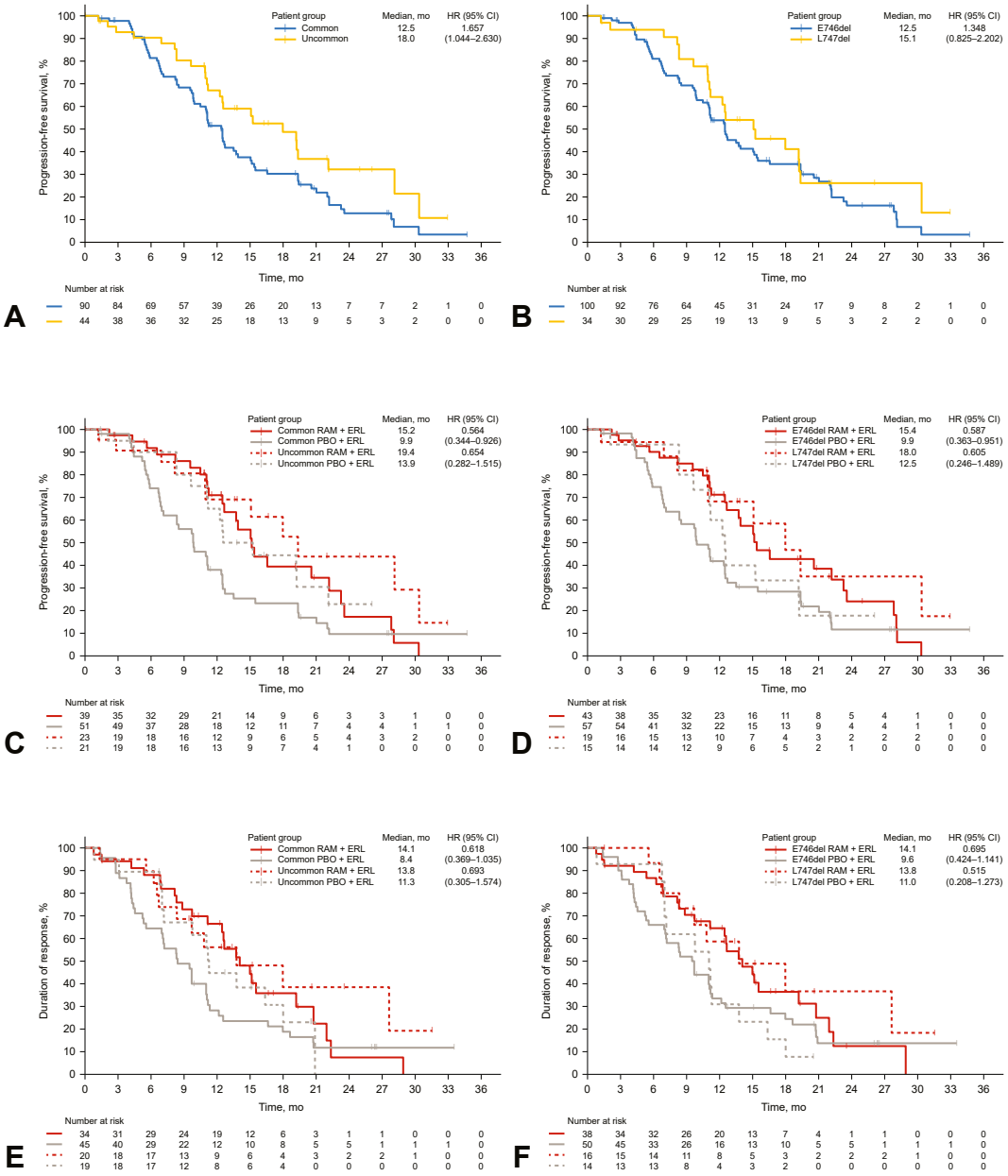


Figure 2. Kaplan-Meier curves of PFS and DoR by ex19del subgroups and by treatment arm. (*A*) PFS, common versus uncommon ex19del variants. (*B*) PFS, E746del versus L747del variants. (*C*) PFS by treatment arm, common versus uncommon ex19del variants. (*D*) PFS by treatment arm, E746del versus L747del variants. (*E*) DoR by treatment arm, common versus uncommon ex19del variants. (*F*) DoR by treatment arm, E746del versus L747del variants. Population comprised RELAY ITT patients with a valid plasma sample with an ex19del variant detected at baseline. CI, confidence interval; DoR, duration of response; E746del, E746 deletion; ex19del, exon 19 deletion; HR, hazard ratio; ITT, intent-to-treat; L747del, L747 deletion; PFS, progression-free survival.

uncommon subgroup, and in the E746del subgroup but not the L747del subgroup (Table 4). Within the common and E746del patient subgroups, treatment-emergent *TP53* mutations were statistically significantly more frequent in the RAM plus ERL arm versus the PBO plus ERL arm (common: RAM + ERL, 40.0% [six of 15 patients] versus PBO + ERL, 9.1% [three of 33 patients], $p = 0.0141$; E746del: RAM + ERL, 40.0% [six of 15 patients] versus PBO + ERL, 8.6% [three of 35 patients]; $p = 0.0110$). Overall, patients with uncommon variants or L747del variants more frequently had no treatment-emergent mutations compared with patients with the common or E746del variants.

Metastases at Disease Progression

No CNS metastases were observed at disease progression in the RAM plus ERL arm in any ex19del subgroup (Supplementary Table 2). In the PBO plus

Table 3. ORR and DCR by *EGFR* Ex19del Subgroup and Treatment Arm (TR Population)

	Common (n = 90)		Uncommon (n = 44)		E746del (n = 100)		L747del (n = 34)	
Response	RAM + ERL (n = 39)	PBO + ERL (n = 51)	RAM + ERL (n = 23)	PBO + ERL (n = 21)	RAM + ERL (n = 43)	PBO + ERL (n = 57)	RAM + ERL (n = 19)	PBO + ERL (n = 15)
ORR (CR + PR), n	34	45	20	19	38	50	16	14
% (95% CI)	87.2 (73.3-94.4)	88.2 (76.6-94.5)	87.0 (67.9-95.5)	90.5 (71.1-97.4)	88.4 (75.5-94.9)	87.7 (76.8-93.9)	84.2 (62.4-94.5)	93.3 (70.2-98.8)
DCR (CR + PR + SD), n	39	49	21	20	43	54	17	15
% (95% CI)	100.0 (91.0-100.0)	96.1 (86.8-98.9)	91.3 (73.2-97.6)	95.2 (77.3-99.2)	100.0 (91.8-100.0)	94.7 (85.6-98.2)	89.5 (68.6-97.1)	100.0 (79.6-100.0)

Note: TR population comprised patients in the RELAY ITT population with a valid plasma sample with an ex19del variant detected at baseline.
CI, confidence interval; CR, complete response; DCR, disease control rate; ERL, erlotinib; ex19del, exon 19 deletion; ITT, intent-to-treat; ORR, objective response rate; PBO, placebo; PR, partial response; RAM, ramucirumab; SD, stable disease; TR, translational research.

ERL arm, CNS metastases were observed in one patient each in the common and uncommon subgroups and two patients in the E746del subgroup (Supplementary Table 2). The presence of new liver metastases at disease progression was observed in one patient in the RAM plus ERL arm of each ex19del subgroup. In the PBO plus ERL arm, new liver metastases were observed in one patient each in the uncommon and L747del subgroups and in two patients each in the common and E746del subgroups (Supplementary Table 2).

Post-Treatment Discontinuation Osimertinib Treatment

In patients who discontinued the study treatment, more than 50% of the patients in each ex19del subgroup received osimertinib treatment as any subsequent therapy: common subgroup: 37 of 71 patients (52.1%); the uncommon subgroup: 14 of 24 patients (58.3%); E746del subgroup: 39 of 74 patients (52.7%); and L747del subgroup: 12 of 21 patients (57.1%).

Safety

In the TR safety population, all patients in all subgroups had greater than or equal to 1 treatment-emergent AEs (TEAEs; Supplementary Table 3). The proportion of patients with treatment-related grade greater than or equal to 3 AEs was slightly higher in the RAM plus ERL arm compared with the PBO plus ERL arm in all patient subgroups. Between patient subgroups, treatment-related grade greater than or equal to 3 AEs generally occurred at a higher frequency in the uncommon versus common subgroups and in the L747del versus E746del subgroups, respectively. No patient in the TR safety population died due to an AE related to the study treatment.

Discussion

This is the first report to use prospective data from a global phase 3 study to evaluate potential associations between *EGFR* ex19del variants and clinical outcomes for combination treatment with an EGFR TKI and an angiogenesis inhibitor. In this study of patients from the RELAY phase 3 trial,[25] the distribution of common (E746_A750del variant) versus uncommon (non-E746_A750del variants) ex19del variants at baseline was similar to that reported in previous studies.[6,8,9] In all ex19del subgroups, RAM plus ERL had consistent improvements in PFS compared with PBO plus ERL. After disease progression, a higher proportion of patients with the common ex19del variant was positive for treatment-emergent *EGFR* T790M, suggesting potentially different resistance mechanisms between ex19del variants.

Table 4. Treatment-Emergent Gene Alterations Occurring After Disease Progression by *EGFR* Ex19del Subgroup

	Common (n = 48)		Uncommon (n = 14)		E746del (n = 50)		L747del (n = 12)	
Genetic Region, n (%)	RAM + ERL (n = 15)	PBO + ERL (n = 33)	RAM + ERL (n = 5)	PBO + ERL (n = 9)	RAM + ERL (n = 15)	PBO + ERL (n = 35)	RAM + ERL (n = 5)	PBO + ERL (n = 7)
APC	1 (6.7)	0	1 (20.0)	1 (11.1)	1 (6.7)	0	1 (20.0)	1 (14.3)
ARID1A	1 (6.7)	1 (3.0)	0	0	1 (6.7)	1 (2.9)	0	0
BRAF	1 (6.7)	1 (3.0)	0	1 (11.1)	1 (6.7)	1 (2.9)	0	1 (14.3)
BRCA2	0	1 (3.0)	0	0	0	1 (2.9)	0	0
CCNE1	1 (6.7)	0	0	0	1 (6.7)	0	0	0
CDK4	0	1 (3.0)	1 (20.0)	0	0	1 (2.9)	1 (20.0)	0
CDK6	0	0	1 (20.0)	0	0	0	1 (20.0)	0
EGFR T790M	6 (40.0)	15 (45.5)	1 (20.0)	2 (22.2)	6 (40.0)	15 (42.9)	1 (20.0)	2 (28.6)
EGFR other	1 (6.7)	3 (9.1)	1 (20.0)	1 (11.1)	1 (6.7)	3 (8.6)	1 (20.0)	1 (14.3)
ERBB2	0	1 (3.0)	1 (20.0)	0	0	1 (2.9)	1 (20.0)	0
FGFR1	1 (6.7)	0	0	0	1 (6.7)	0	0	0
FGFR2	1 (6.7)	2 (6.1)	1 (20.0)	0	1 (6.7)	2 (5.7)	1 (20.0)	0
GNAS	1 (6.7)	0	0	0	1 (6.7)	0	0	0
KIT	1 (6.7)	0	0	0	1 (6.7)	0	0	0
KRAS	2 (13.3)	1 (3.0)	1 (20.0)	0	2 (13.3)	1 (2.9)	1 (20.0)	0
MAP2K2	0	1 (3.0)	0	0	0	1 (2.9)	0	0
MAPK3	1 (6.7)	0	0	0	1 (6.7)	0	0	0
MET	1 (6.7)	1 (3.0)	0	0	1 (6.7)	1 (2.9)	0	0
MTOR	0	1 (3.0)	0	0	0	1 (2.9)	0	0
NF1	2 (13.3)	1 (3.0)	0	0	2 (13.3)	1 (2.9)	0	0
PDGFRA	1 (6.7)	0	0	0	1 (6.7)	0	0	0
PIK3CA	1 (6.7)	0	0	1 (11.1)	1 (6.7)	0	0	1 (14.3)
RB1	1 (6.7)	0	0	0	1 (6.7)	0	0	0
SMAD4	1 (6.7)	0	0	0	1 (6.7)	0	0	0
STK11	0	0	0	1 (11.1)	0	0	0	1 (14.3)
TP53[a]	6 (40.0)	3 (9.1)	0	0	6 (40.0)	3 (8.6)	0	0
TSC1	1 (6.7)	0	0	0	1 (6.7)	0	0	0
No treatment-emergent gene alterations	3 (20.0)	14 (42.4)	2 (40.0)	5 (55.6)	3 (20.0)	16 (45.7)	2 (40.0)	3 (42.9)

Note: Population consists of patients in the TR population with a valid plasma sample with an ex19del variant detected at baseline and a post-study treatment discontinuation 30-day follow-up sample with any gene alteration detected.

[a]*TP53* mutations were statistically significantly more frequent in the RAM plus ERL arm versus the PBO plus ERL arm in the common subgroup ($p = 0.0141$) and E746del subgroup ($p = 0.0110$); no other statistically significant differences were observed across the treatment arms.

E746del, E746 deletion; ERL, erlotinib; ex19del, exon 19 deletion; L747del, L747 deletion; PBO, placebo; RAM, ramucirumab; TR, translational research.

Furthermore, patients with uncommon ex19del variants more frequently had no treatment-emergent gene alterations compared with those with the common variant. When patients were classified into subgroups according to the *EGFR* exon 19 deleted codon (E746del variants versus L747del variants), results for the E746del subgroup were similar to those for the common variant, which is a subset of the E746del subgroup, and L747del results were similar to the uncommon subgroup. These results suggest that the type of *EGFR* ex19del variant does not affect RAM plus ERL treatment outcomes. Nevertheless, the results of these post hoc analyses should be interpreted with caution due to the small sample sizes.

Baseline patient and disease characteristics did not reveal any phenotypic difference between ex19del variants, except for patients with the uncommon and L747del variants more frequently being never smokers. This differs from what was observed in a retrospective study of Chinese patients with NSCLC, where more patients with the common/E746del variants were never smokers compared with patients with uncommon/L747del variants.[9] In a second retrospective study of Chinese patients with NSCLC, most patients were never smokers but no difference was observed between the E746del and L747del subgroups.[18] Similarly, in an Italian study of patients with metastatic NSCLC, more than half the patients were never smokers but the proportion of never smokers was similar between the ex19del variants.[16] In the current study, the higher frequency of never smokers among patients with uncommon and L747del variants may be related to the better clinical outcomes. *TP53* mutations are regarded to be more prevalent in patients with a smoking history than in never smokers.[27] The most frequent concurrent baseline alteration in all ex19del variants in the current study was a *TP53* mutation, and its frequency varied among the treatment arms (53.8%–73.2%), as noted previously.[15]

In the current study, ex19del variants were determined in cell-free circulating tumor DNA, which only captures "shedding tumors,"[28] and not from tumor tissue. Circulating tumor DNA–positive tumors are generally associated with worse treatment outcomes,[29] and this may, in part, explain why the median PFS for the different ex19del variants in this study was generally lower than that reported for patients with an ex19del mutation in the RELAY ITT population.[26] When evaluating combined treatment arms in the current study, PFS was statistically significantly longer for patients with uncommon (non-E746_A750del) variants versus those with the common (E746_A750del) variant (18.0 mo versus 12.5 mo, respectively). When analyzed by treatment arm, RAM plus ERL had statistically significant improvements in PFS compared with PBO plus ERL in patients with the common variant (15.2 mo versus 9.9 mo, respectively) and E746del variants (15.4 mo versus 9.9 mo, respectively) and marginal improvements in patients with uncommon variants and those with L747del variants, but sample size was small; therefore, interpretation of these outcomes should be considered with caution. A similar PFS benefit for patients with uncommon (non-E746_A750del) variants or L747del variants has been reported. In a retrospective case-control comparative study, a significantly longer PFS was reported for patients with uncommon variants who received first-line EGFR TKI therapy versus those with the common variant (19 mo versus 13 mo, $p = 0.0016$).[15] In two retrospective studies of patients who received first-generation EGFR TKIs as initial therapy, median PFS was longer for L747del versus E746del subgroups but was not statistically significantly different.[12,18]

Nevertheless, other studies have revealed the converse, with longer PFS being reported for patients with the common variant and E746del variants. In two observational studies of patients who received first-line EGFR TKI therapy (ERL or gefitinib), significantly longer PFS was observed for patients with E746del variants versus L747del variants.[13,14] In addition, among patients treated with first-line EGFR TKIs (gefitinib or afatinib), longer PFS was observed for those with the common variant versus uncommon variants, and with E746del versus L747del variants (14.4 mo versus 11.9 mo for both analyses), but the differences were not statistically significant.[16] In a prospective study of patients treated with either first-, second-, or third-generation EGFR TKIs, PFS was longer for patients with E746 insertions and deletions than for those with L747 insertions and deletions.[30] In a retrospective study of patients treated with the third-generation EGFR TKI osimertinib as initial therapy, the common ex19del variant E746_A750del was associated with improved PFS compared with the ex19del variant L747_A750>P.[19]

Other studies have reported no difference in PFS for patients with E746del variants compared with L747del variants, including a prospective study of patients treated with afatinib monotherapy.[8] In a retrospective study of patients with ex19del variants treated with first-line EGFR TKIs, no difference was observed between E746del and L747del subgroups, but multivariate analysis revealed that ex19del subtypes had a marginal effect on PFS ($p = 0.051$).[9] The conflicting differences observed for ex19del variants may be explained in part by the drug type; however, Zhao et al.[9] found no difference in PFS between ex19del subgroups treated with different EGFR TKIs (gefitinib, ERL, icotinib), and, therefore, other biological mechanisms might have

potentially contributed to the conflicting clinical outcomes reported.

Although patients with the common ex19del variant were more likely to have a treatment-emergent gene alteration on disease progression than patients with uncommon ex19del variants, not all treatment-emergent gene alterations are involved in acquisition of treatment resistance. A higher proportion of patients with the common ex19del variant was positive for a treatment-emergent *TP53* gene alteration compared with patients with uncommon ex19del variants. The analyses conducted in the current study were based on plasma, and the presence of a *TP53* gene alteration on disease progression could be related to the tumor burden, tumor distribution, shedding ability of that tumor, or the relatively small sample sizes. This could explain why a *TP53* gene alteration emerged in some patients who did not have a *TP53* mutation at baseline. Higher rates of treatment-emergent *EGFR* T790M in patients with the common ex19del variant than the uncommon variants were observed in this study, as reported previously,[9,20–22] and in patients with E746del than in those with other variants.[31] These results further support the idea that different resistance mechanisms exist between ex19del variants. Because the *EGFR* T790M emergent rate is higher in patients with the common ex19del variant than in those with uncommon ex19del variants,[9,20–22] and because osimertinib was found to have better efficacy for T790M-positive patients with the common ex19del variant,[15,24] sequential RAM plus ERL and osimertinib treatment could be considered as a treatment option for patients with the common ex19del variant.

The strength of this post hoc analysis was that data from a large randomized, PBO-controlled, global study were used to evaluate clinical outcomes of patients with ex19del variants. Previous studies reporting on outcomes of different treatments in patients with different ex19del variants have been based largely on retrospective analyses from chart reviews with small sample sizes. Furthermore, this study is the first global phase 3 study to evaluate the impact of dual EGFR and VEGF pathway inhibition on the outcomes of ex19del variants.

This post hoc exploratory analysis had several limitations. Although randomization did not stratify patients by ex19del variant subtype, patient and disease characteristics, with the exception of smoking history, were comparable between the ex19del subgroups at baseline. Only patients with assessable baseline plasma Guardant360 NGS results positive for an *EGFR* ex19del variant were included in this analysis, which may have led to a selection bias. The number of patients for whom valid baseline plasma samples were available was small; results should be viewed with caution. Time-to-event data may also be biased toward patients who progressed and discontinued study treatment because patients who were still on treatment were not included.

These results support RAM plus ERL as a suitable first-line treatment to provide benefit and improve outcomes for patients with metastatic NSCLC with *EGFR* ex19del variants. Understanding the potential impact of different *EGFR* ex19del variants on treatment outcomes and their mechanism of resistance may help inform treatment decisions.

CRediT Authorship Contribution Statement

Kazumi Nishino: Conceptualization, Investigation, and Writing—review and editing.

Jin-Yuan Shih: Data curation, Formal analysis, Methodology, Project administration, and Writing—review and editing.

Kazuhiko Nakagawa, Martin Reck, Edward B. Garon, Ernest Nadal: Investigation and Writing—review and editing.

Michelle Carlsen, Tomoko Matsui, Carla Visseren-Grul: Writing—original draft.

Disclosure

Dr. Nishino reports grants or contracts to their institution from AbbVie, Amgen, Chugai Pharmaceutical Co., Ltd., Daiichi Sankyo Company, Limited, Eisai Co., Ltd., Eli Lilly Japan K.K., Janssen Pharmaceutical K.K., Merck Biopharma Co., Ltd., Merus N.V., Merck Sharp & Dohme, Novartis, Ono Pharmaceutical Co., Ltd., Pfizer, Sanofi K.K., Taiho Pharmaceutical Co., Ltd., and Takeda Pharmaceutical Company Limited; payments or honoraria from AstraZeneca, Bristol-Myers Squibb, Chugai Pharmaceutical Co., Ltd., Eli Lilly Japan K.K., Janssen Pharmaceutical K.K., Merck, Nippon Boehringer Ingelheim Co., Ltd., Nippon Kayaku Co., Ltd., Novartis, Pfizer, and Roche Diagnostics; and participation on safety/advisory boards with AstraZeneca, Eli Lilly Japan K.K., and Pfizer. Dr. Shih reports grants or contracts from Genconn Biotech and Roche; payments or honoraria from ACT Genomics, Amgen, AstraZeneca, Bayer Pharmaceuticals, Boehringer Ingelheim, Bristol-Myers Squibb, Chugai Pharmaceutical Co., Ltd., CStone Pharmaceuticals, Eli Lilly and Company, Genconn Biotech, GlaxoSmithKline, Janssen, Lotus, Merck Sharp & Dohme, Mundipharma, Novartis, Orient EuroPharma, Ono Pharmaceutical Co., Ltd., Pfizer, Roche, Takeda Pharmaceutical Company Limited, and TTY Biopharm; and support for attending meetings and/or travel from AstraZeneca, Chugai Pharmaceutical Co., Ltd., and Roche. Dr. Nakagawa reports grants or contracts to

their institution from Amgen, Ascent Development Services, Astellas Pharma Inc., AstraZeneca K.K., Bayer Yakuhin, Ltd., Bristol-Myers Squibb, Chugai Pharmaceutical Co., Ltd., CMIC Co., Ltd., Daiichi Sankyo Company, Limited, Eisai Co., Ltd., Eisai Inc., Eli Lilly Japan K.K., EP-CRSU Co., Ltd., EPS Corporation, GlaxoSmithKline K.K., Iqvia Services Japan K.K., Janssen Pharmaceutical K.K., Japan Clinical Research Operations, Kissei Pharmaceutical Co., Ltd., Kobayashi Pharmaceutical Co., Ltd., Labcorp Development Japan K.K. (Covance Japan Co., Ltd.), Mebix, Inc., Medical Research Support, Mochida Pharmaceutical Co., Ltd., Merck Sharp & Dohme K.K., Nippon Boehringer Ingelheim Co., Ltd., Nippon Kayaku Co., Ltd., Novartis Pharma K.K., Ono Pharmaceutical Co., Ltd., Otsuka Pharmaceutical Co., Ltd., Parexel International Corp., Pfizer Japan Inc., Pfizer R&D Japan G.K., PPD-SNBL K.K., PRA Health Sciences, Sanofi K.K., Shionogi & Co., Ltd., SRL, Inc., SymBio Pharmaceuticals Limited, Syneos Health Clinical K.K., Sysmex Corporation, Taiho Pharmaceutical Co., Ltd., and Takeda Pharmaceutical Company Limited; consulting fees from Eli Lilly Japan K.K. and Ono Pharmaceutical Co., Ltd.; payments or honoraria from 3H Clinical Trial Inc., Amgen, AstraZeneca K.K., Bayer Yakuhin, Ltd., Bristol-Myers Squibb K.K., CareNet, Inc., Chugai Pharmaceutical Co., Ltd., CMIC Co., Ltd., CMIC ShiftZero K.K., Daiichi Sankyo Company, Limited, Eli Lilly Japan K.K., Incyte Biosciences Japan, Janssen Pharmaceutical K.K., Japan Clinical Research Operations, Kyowa Kirin Co., Ltd., Life Technologies Japan, Medical Mobile Communications Co., Ltd., Medical Review Co., Ltd., Merck Biopharma Co., Ltd., Merck Sharp & Dohme K.K., Neo Communication A.G., Nikkei Business Publications, Inc., Nippon Boehringer Ingelheim Co., Ltd., Nippon Kayaku Co., Ltd., Novartis Pharma K.K., Ono Pharmaceutical Co., Ltd., Pfizer Japan Inc., Taiho Pharmaceutical Co., Ltd., Taiyo Pharma Co., Ltd., Takeda Pharmaceutical Company Limited, and Yodosha Company, Ltd.; and patents planned, issued, or pending with Daiichi Sankyo Company, Limited, and their institution. Dr. Reck reports consulting fees from Amgen, AstraZeneca, BeiGene, Bristol-Myers Squibb, Boehringer Ingelheim, Daiichi Sankyo Company, Limited, Eli Lilly and Company, GlaxoSmithKline, Merck, Merck Sharp & Dohme, Mirati Therapeutics, Inc., Novartis, Pfizer, Roche, Samsung Bioepis, and Sanofi; payments or honoraria from Amgen, AstraZeneca, BeiGene, Bristol-Myers Squibb, Boehringer Ingelheim, Daiichi Sankyo Company, Limited, Eli Lilly and Company, GlaxoSmithKline, Merck, Merck Sharp & Dohme, Mirati Therapeutics, Inc., Novartis, Pfizer, Roche, Sanofi, and Samsung Bioepis; support for attending meetings and/or travel from Amgen, AstraZeneca, BeiGene, Bristol-Myers Squibb, Boehringer Ingelheim, Daiichi Sankyo Company, Limited, Eli Lilly and Company, GlaxoSmithKline, Merck, Merck Sharp & Dohme, Mirati Therapeutics, Inc., Novartis, Pfizer, Roche, Sanofi, and Samsung Bioepis; and honoraria for participation on a data safety monitoring board or advisory board from Daiichi Sankyo Company, Limited, and Sanofi. Dr. Garon reports consultant and/or advisory fees from AbbVie, ABL Bio, AstraZeneca, Boehringer Ingelheim, Bristol-Myers Squibb, Dracen Pharmaceuticals, EMD Serono, Eisai Co., Ltd., Eli Lilly and Company, Gilead Sciences, GlaxoSmithKline, Merck, Natera, Novartis, Personalis, Regeneron, Sanofi, Shionogi, Xilio Therapeutics, and Zymeworks; grant/research support from ABL Bio, AstraZeneca, Bristol-Myers Squibb, Daiichi Sankyo Company, Limited, Dynavax Technologies, Eli Lilly and Company, EMD Serono, Genentech, Gilead Sciences, Iovance Biotherapeutics, Merck, Mirati Therapeutics, Neon, and Novartis; sponsored independent medical education from Daiichi Sankyo Company, Limited, and Ipsen; and travel support from A2 Biotherapeutics and Novartis. Ms. Carlsen and Dr. Visseren-Grul are employees and minor shareholders of Eli Lilly and Company. Ms. Matsui is an employee of Eli Lilly Japan K.K. and is a minor shareholder of Eli Lilly and Company. Dr. Nadal received research funding from Bristol-Myers Squibb, Merck Serono, Pfizer, and Roche; participated in advisory boards or received honoraria from Amgen, AstraZeneca, Bayer Pharmaceuticals, Boehringer Ingelheim, Bristol-Myers Squibb, Daiichi Sankyo Company, Limited, Eli Lilly and Company, Janssen, Merck Serono, Merck Sharp & Dohme, Pfizer, Pierre Fabre, Qiagen, Roche, Sanofi, and Takeda Pharmaceutical Company Limited; and received travel support from Merck Sharp & Dohme, Pfizer, Roche, and Takeda Pharmaceutical Company Limited.

Acknowledgments

This study was funded by Eli Lilly and Company, the manufacturer/licensee of ramucirumab. Medical writing assistance was provided by Prudence Stanford, PhD, CMPP, and Rebecca Lew, PhD, CMPP, of ProScribe – Envision Pharma Group, and was funded by Eli Lilly Japan K.K. ProScribe's services complied with international guidelines for Good Publication Practice. Eli Lilly and Company was involved in the study design, data collection, data analysis, and preparation of the manuscript. The authors are grateful to the patients, their families, and caregivers for participating in the RELAY trial and to the study investigators and site staff for their collaboration.

Data Availability

This manuscript reports the results of an exploratory biomarker study of the RELAY randomized, double-blind, placebo-controlled, phase 3 study; data are therefore of an exploratory nature only and will not be shared. Trial Protocol: Nakagawa et al.[25]

Supplementary Data

Note: To access the supplementary material accompanying this article, visit the online version of the *JTO Clinical and Research Reports* at www.jtocrr.org and at https://doi.org/10.1016/j.jtocrr.2023.100624.

References

1. Hsu WH, Yang JC, Mok TS, Loong HH. Overview of current systemic management of EGFR-mutant NSCLC. *Ann Oncol*. 2018;29(suppl 1):i3–i9.
2. Shigematsu H, Lin L, Takahashi T, et al. Clinical and biological features associated with epidermal growth factor receptor gene mutations in lung cancers. *J Natl Cancer Inst*. 2005;97:339–346.
3. Hendriks LE, Kerr KM, Menis J, et al. Oncogene-addicted metastatic non-small-cell lung cancer: ESMO Clinical Practice Guideline for diagnosis, treatment and follow-up. *Ann Oncol*. 2023;34:339–357.
4. Singh N, Jaiyesimi IA, Ismaila N, et al. Therapy for stage IV non-small-cell lung cancer with driver alterations: ASCO living guideline, version 2023.1. *J Clin Oncol*. 2023;41:e42–e50.
5. Passaro A, Jänne PA, Mok T, Peters S. Overcoming therapy resistance in EGFR-mutant lung cancer. *Nat Cancer*. 2021;2:377–391.
6. Gu W, Lu Z, Shi S, et al. Molecular characteristics of EGFR exon 19 deletion subtypes in NSCLC patients. *J Clin Oncol*. 2021;39:8530.
7. Brown BP, Zhang YK, Kim S, et al. Allele-specific activation, enzyme kinetics, and inhibitor sensitivities of EGFR exon 19 deletion mutations in lung cancer. *Proc Natl Acad Sci U S A*. 2022;119:e2206588119.
8. Tokudome N, Koh Y, Akamatsu H, et al. Differential significance of molecular subtypes which were classified into EGFR exon 19 deletion on the first line afatinib monotherapy. *BMC Cancer*. 2020;20:103.
9. Zhao C, Jiang T, Li J, et al. The impact of *EGFR* exon 19 deletion subtypes on clinical outcomes in non-small cell lung cancer. *Transl Lung Cancer Res*. 2020;9:1149–1158.
10. Kobayashi Y, Mitsudomi T. Not all epidermal growth factor receptor mutations in lung cancer are created equal: perspectives for individualized treatment strategy. *Cancer Sci*. 2016;107:1179–1186.
11. Kohsaka S, Nagano M, Ueno T, et al. A method of high-throughput functional evaluation of *EGFR* gene variants of unknown significance in cancer. *Sci Transl Med*. 2017;9:eaan6566.
12. Chung KP, Wu SG, Wu JY, et al. Clinical outcomes in non-small cell lung cancers harboring different exon 19 deletions in EGFR. *Clin Cancer Res*. 2012;18:3470–3477.
13. Kaneda T, Hata A, Tomioka H, et al. Possible differential EGFR-TKI efficacy among exon 19 deletional locations in EGFR-mutant non-small cell lung cancer. *Lung Cancer*. 2014;86:213–218.
14. Lee VH, Tin VP, Choy TS, et al. Association of exon 19 and 21 EGFR mutation patterns with treatment outcome after first-line tyrosine kinase inhibitor in metastatic non-small-cell lung cancer. *J Thorac Oncol*. 2013;8:1148–1155.
15. Peng X, Long X, Liu L, et al. Clinical impact of uncommon epidermal growth factor receptor exon 19 insertion-deletion variants on epidermal growth factor receptor-tyrosine kinase inhibitor efficacy in non-small-cell lung cancer. *Eur J Cancer*. 2020;141:199–208.
16. Rossi S, Toschi L, Finocchiaro G, et al. Impact of exon 19 deletion subtypes in EGFR-mutant metastatic non-small-cell lung cancer treated with first-line tyrosine kinase inhibitors. *Clin Lung Cancer*. 2019;20:82–87.
17. Sutiman N, Tan SW, Tan EH, et al. EGFR mutation subtypes influence survival outcomes following first-line gefitinib therapy in advanced Asian NSCLC patients. *J Thorac Oncol*. 2017;12:529–538.
18. Xu H, Li W, Yang G, et al. Heterogeneous response to first-generation tyrosine kinase inhibitors in non-small-cell lung cancers with different EGFR exon 19 mutations. *Target Oncol*. 2020;15:357–364.
19. Grant MJ, Aredo JV, Starrett JH, et al. Efficacy of osimertinib in patients with lung cancer positive for uncommon EGFR exon 19 deletion mutations. *Clin Cancer Res*. 2023;29:2123–2130.
20. Huang YH, Hsu KH, Tseng JS, et al. The association of acquired T790M mutation with clinical characteristics after resistance to first-line epidermal growth factor receptor tyrosine kinase inhibitor in lung adenocarcinoma. *Cancer Res Treat*. 2018;50:1294–1303.
21. Zou B, Lee VHF, Chen L, Ma L, Wang DD, Yan H. Deciphering mechanisms of acquired T790M mutation after EGFR inhibitors for NSCLC by computational simulations. *Sci Rep*. 2017;7:6595.
22. Huang LT, Zhang SL, Han CB, Ma JT. Impact of EGFR exon 19 deletion subtypes on clinical outcomes in EGFR-TKI-treated advanced non-small-cell lung cancer. *Lung Cancer*. 2022;166:9–16.
23. Mok TS, Wu YL, Ahn MJ, et al. Osimertinib or platinum-pemetrexed in EGFR T790M-positive lung cancer. *N Engl J Med*. 2017;376:629–640.
24. Zheng Q, Huang Y, Zhao H, et al. *EGFR* mutation genotypes affect efficacy and resistance mechanisms of osimertinib in T790M-positive NSCLC patients. *Transl Lung Cancer Res*. 2020;9:471–483.
25. Nakagawa K, Garon EB, Seto T, et al. Ramucirumab plus erlotinib in patients with untreated, EGFR-mutated, advanced non-small-cell lung cancer (RELAY): a randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet Oncol*. 2019;20:1655–1669.
26. Nakagawa K, Nadal E, Garon EB, et al. RELAY subgroup analyses by EGFR ex19del and ex21L858R mutations for ramucirumab plus erlotinib in metastatic non-small cell lung cancer. *Clin Cancer Res*. 2021;27:5258–5271.
27. Mogi A, Kuwano H. TP53 mutations in nonsmall cell lung cancer. *J Biomed Biotechnol*. 2011;2011:583929.
28. Sánchez-Herrero E, Serna-Blasco R, Robado de Lope L, González-Rumayor V, Romero A, Provencio M. Circulating tumor DNA as a cancer biomarker: an overview of biological features and factors that may impact on ctDNA analysis. *Front Oncol*. 2022;12:943253.

29. Nishio K, Sakai K, Nishio M, et al. Impact of ramucirumab plus erlotinib on circulating cell-free DNA from patients with untreated metastatic non-small cell lung cancer with *EGFR*-activating mutations (RELAY phase 3 randomized study). *Transl Lung Cancer Res*. 2023;12:1702–1716.
30. Wu SG, Gow CH, Chen YL, Liu YN, Tsai MF, Shih JY. Different treatment efficacies and T790M acquisition of EGFR-TKIs on NSCLC patients with variable Del-19 subtypes of EGFR. *Int J Cancer*. 2023;153:352–363.
31. Gu Y, Yu J, Hu H, Zhang H, Cao B, Liang L. Clinical outcomes of advanced NSCLC patients with different EGFR exon 19 deletion subtypes treated with first-line tyrosine kinase inhibitors: a single-center ambispective cohort study. *Thorac Cancer*. 2023;14:3147–3160.