

Programmed Cell Death Ligand 1 Expression in Untreated *EGFR* Mutated Advanced NSCLC and Response to Osimertinib Versus Comparator in FLAURA



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ABSTRACT

Introduction: EGFR mutated (EGFRm) NSCLC tumors occasionally express programmed cell death ligand 1 (PD-L1), although frequency and clinical relevance are not fully characterized. We report PD-L1 expression in patients with EGFRm advanced NSCLC and association with clinical outcomes following treatment with osimertinib or comparator EGFR tyrosine kinase inhibitors in the FLAURA trial (phase III, NCT02296125).

Methods: Of 231 tissue blocks available from the screened population (including EGFRm-positive and -negative samples), 197 had sufficient tissue for PD-L1 testing using the SP263 (Ventana, Tucson, Arizona) immunohistochemical assay. Tumor cell (TC) staining thresholds of PD-L1 TC greater than or equal to 1%, TC greater than or equal to 25%, and TC greater than or equal to 50% were applied. Progression-free survival (PFS) was investigator-assessed, per Response Evaluation Criteria in Solid Tumor, version 1.1, according to PD-L1 expressors (TC \geq 1%) or negatives (TC < 1%) in randomized patients.

Results: PD-L1 staining was successful in 193 of 197 patient formalin-fixed paraffin-embedded blocks; of these, 128 of 193 were EGFRm-positive and 106 of 128 patients were randomized to treatment (osimertinib: 54; comparator: 52).

At the PD-L1 TC greater than or equal to 25% threshold, 8% (10 of 128) of EGFRm-positive tumors expressed PD-L1 versus 35% (23 of 65) of EGFRm-negative tumors. With the TC greater than or equal to 1% threshold, 51% (65 of 128) versus 68% (44 of 65) were mutation-positive and -negative, respectively, and with the TC greater than or equal to 50% threshold, 5% (7 of 128) versus 28% (18 of 65), were mutation-positive and -negative, respectively. For PD-L1 expressors (TC \geq 1%), median PFS was 18.4 months

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with osimertinib and 6.9 months with comparator (hazard ratio = 0.30; 95% confidence interval: 0.15–0.60). For PD-L1-negative patients (TC < 1%), median PFS was 18.9 months with osimertinib and 10.9 months with comparator (hazard ratio = 0.37; 95% confidence interval: 0.17–0.74).

Conclusions: Clinical benefit with osimertinib was unaffected by PD-L1 expression status.

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Keywords: Osimertinib; Programmed death ligand 1; FLAURA; EGFR mutated; NSCLC

Introduction

Osimertinib is a third-generation, irreversible, oral EGFR tyrosine kinase inhibitor (TKI) that potently and selectively inhibits both EGFR TKI-sensitizing mutations and EGFR T790M and has shown efficacy in NSCLC central nervous system metastases.^{1–5} In FLAURA (NCT02296125, phase III), patients with newly diagnosed EGFR mutation-positive (EGFRm) NSCLC treated with osimertinib had significantly improved progression-free survival (PFS) versus comparator EGFR TKIs (erlotinib/gefitinib).² Median PFS was 18.9 months with osimertinib versus 10.2 months with comparator EGFR TKIs (hazard ratio [HR] = 0.46; 95% confidence interval [CI]: 0.37–0.57; $p < 0.001$).²

Separately from EGFR TKIs, the development of inhibitors targeting programmed cell death 1 (PD-1) receptor or programmed cell death ligand 1 (PD-L1) protein has resulted in a major advance in the treatment of NSCLC, with studies showing superior outcomes with PD-1/PD-L1 immunotherapies versus chemotherapy.^{6,7} However, the majority of the first-line immunotherapy trials have excluded EGFRm patients.^{8,9} Additionally, in a phase II trial, pembrolizumab (a PD-1 inhibitor) failed to show efficacy (with no reported objective responses) in

EGFR TKI-naïve patients with EGFRm and PD-L1-positive advanced NSCLC.¹⁰ The biological basis for the lack of efficacy with immune checkpoint inhibitors in EGFRm patients is not known.¹¹ With PD-L1 emerging as an important biomarker for immune checkpoint inhibition, interrogation of the interplay between PD-L1 expression and EGFRm is important. The relationship between PD-L1 expression level and sensitivity to EGFR TKIs in patients with EGFRm NSCLC is not firmly established.¹¹

We report the prevalence and impact of PD-L1 expression in patients with EGFRm advanced NSCLC from the phase III FLAURA trial.

Materials and Methods

Patients, Trial Design, and Treatments

Patients from the FLAURA study were included in this analysis; full details of this study, its patients, trial design, and treatments have been previously published.² Briefly, patients had locally advanced or metastatic NSCLC and were treatment-naïve for advanced disease. Local or central confirmation of EGFR Ex19del or L858R by tissue testing was required for enrollment. Patients were randomized to treatment with osimertinib 80 mg once daily or EGFR TKI comparator (gefitinib 250 mg once daily or erlotinib 150 mg once daily). Endpoints included PFS, objective response rate, and duration of response.²

Tumor Sample Analysis

Tissue samples from 994 patients were screened in the FLAURA study for EGFR Ex19del or L858R as part of the enrollment criteria using central testing (cobas EGFR Mutation Test, Roche Molecular Systems, Pleasanton, California) or a local EGFR test in an accredited laboratory. Five hundred fifty-six patients were randomized to treatment. For tumor cell (TC) scoring, evaluable tissue blocks were tested for PD-L1 status/expression using the VENTANA PD-L1 (SP263) immunohistochemical assay (Ventana Medical Systems, Tucson, Arizona). Positive tumor cell staining thresholds for PD-L1 of 1%

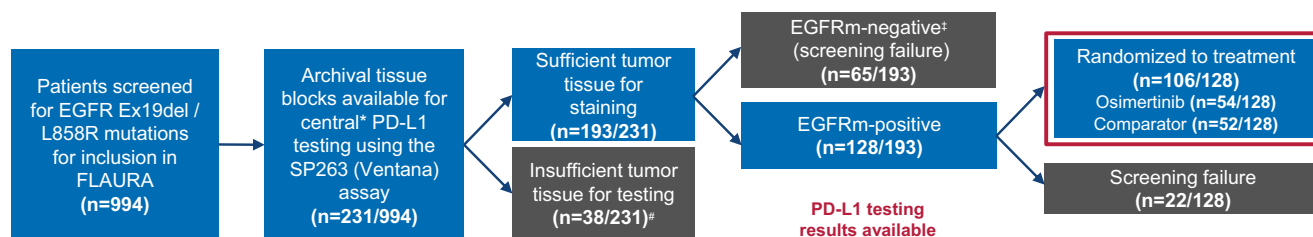


Figure 1. Flow chart of FLAURA study patients and PD-L1 testing in this analysis. *Central PD-L1 testing was carried out by Hematogenix (United Kingdom). #On inspection of the samples, 34 formalin-fixed paraffin-embedded blocks contained insufficient/no tissue for PD-L1 testing, an additional four samples were found to be insufficient for testing upon hematoxylin and eosin assessment. †Tumors were required to harbor one or both of the Ex19del and L858R EGFR-TKI sensitizing mutations for randomization into the study. EGFRm, EGFR mutated; Ex19del, exon 19 deletion; PD-L1, programmed cell death ligand 1.

(TC \geq 1%), 25% (TC \geq 25%), and 50% (TC \geq 50%) were applied. Clinical outcomes were available for those patients with PD-L1 TC staining data who were randomized to treatment.

Immune cell (IC) scoring does not form part of the PD-L1 determination in NSCLC using the Ventana SP263 assay, but is incorporated into the VENTANA PD-L1 (SP142) Assay. Therefore, a purely exploratory IC analysis was conducted whereby the proportion of tumor area occupied by PD-L1 staining ICs was determined and thresholds of 1% (IC \geq 1%) and 25% (IC \geq 25%) were applied.

Trial Oversight

The trial was conducted in accordance with the provisions of the Declaration of Helsinki, Good Clinical Practice guidelines (as defined by the International Conference on Harmonisation), applicable regulatory requirements, and the policy on bioethics and human biologic samples of the trial sponsor, AstraZeneca. This analysis was funded by the sponsor and was designed by the principal investigators and sponsor. Data underlying the findings described in this manuscript may be

obtained in accordance with AstraZeneca's data sharing policy described at <https://astrazenecagrouptrials.pharmacm.com/ST/Submission/Disclosure>.

Results

Patients

Of 994 patients screened for EGFR mutations for inclusion in FLAURA, 231 tissue blocks were available from the screened population (including both mutation-positive and -negative samples), of which 197 had sufficient tumor content for PD-L1 testing (results were available from 193 of 197 samples as four failed post-staining) (Fig. 1). Among these, 128 of 193 were EGFRm-positive; of these 106 of 128 were randomized in FLAURA (54, osimertinib; 52, comparator), the remaining 22 of 128 failed screening and were not enrolled. In total, 450 randomized patients had unknown PD-L1 status (no tissue available). Baseline demographics are shown in Table 1.

PD-L1 Expression and Efficacy

Expression of PD-L1 was less frequent in EGFRm-positive samples versus EGFRm-negative samples (51%

Table 1. Baseline Demographics and Disease Characteristics of Patients with EGFRm Advanced NSCLC Randomized in FLAURA, According to PD-L1 Expression Status

Characteristic	PD-L1 Expressers (n = 52)	PD-L1 Non-Expressers (n = 54)	PD-L1 Unknown (n = 450)
Median age (range), y	64 (35-85)	65 (40-85)	64 (26-93)
Sex			
Female	36 (69)	40 (74)	274 (61)
Male	16 (31)	14 (26)	176 (39)
Ethnicity			
White	33 (63)	43 (80)	125 (28)
Asian ^a	19 (37)	11 (20)	317 (70)
Other ^b	0	0	8 (2)
EGFR mutation status			
L858R	22 (42)	18 (33)	147 (33)
Exon 19 deletion	29 (56)	34 (63)	250 (56)
EGFRm not detected	1 (2)	1 (2)	4 (1)
Invalid test	0	1 (2)	8 (2)
No/inadequate sample	0	0	41 (9)
Smoking status			
Never	38 (73)	38 (70)	281 (62)
Current	2 (4)	1 (2)	14 (3)
Former	12 (23)	15 (28)	155 (34)
CNS metastases			
Present	12 (23)	9 (17)	95 (21)
Not present	40 (77)	45 (83)	355 (79)
Visceral metastases			
Present	18 (35)	15 (28)	164 (36)
Not present	34 (65)	39 (72)	286 (64)

Values are n (%) unless otherwise stated.

^aMost patients with PD-L1 status unknown were Asian (70%), due to several sites in Asia not providing tissue blocks for testing.

^bOther consisted of patients who were Black/African American, Native Hawaiian or other Pacific Islander, American Indian or Alaska Native, missing CNS, central nervous system; EGFRm, EGFR mutated; PD-L1, programmed cell death ligand 1.

Table 2. Tumor PD-L1 Expression According to EGFR Mutation Status

Mutation Status	PD-L1	PD-L1	PD-L1 TC	PD-L1 TC
	TC < 1%	TC ≥ 1%	≥ 25%	≥ 50%
EGFRm negative (n = 65)				
Screened population	21/65 (32)	44/65 (68)	23/65 (35)	18/65 (28)
EGFRm positive (n = 128)				
Screened population	63/128 (49)	65/128 (51)	10/128 (8)	7/128 (5)
Exon 19 deletions	40/79 (51)	39/79 (49)	6/79 (8)	5/79 (6)
L858R	23/49 (47)	26/49 (53)	4/49 (8)	2/49 (4)
Randomized to treatment (n = 106)	54/106 (51)	52/106 (49)	8/106 (8)	5/106 (5)
Randomized to osimertinib	26/54 (48)	28/54 (52)	3/54 (6)	2/54 (4)
Randomized to comparator EGFR TKI	28/52 (54)	24/52 (46)	5/52 (10)	3/52 (6)

Values are n (%). EGFRm, EGFR mutated; PD-L1, programmed cell death ligand 1; TC, tumor cell; TKI, tyrosine kinase inhibitor.

versus 68% at a TC greater than or equal to 1% threshold) (Table 2). This difference was more pronounced at higher PD-L1 TC thresholds: 8% versus 35% at TC greater than or equal to 25% and 5% versus 28% at TC greater than or equal to 50%. Of these EGFRm-positive samples, prevalence of PD-L1 expression (TC ≥ 1%) for Ex19del and L858R was 49% and 53% of samples, respectively (Table 2).

In PD-L1 expressors (TC ≥ 1%), median PFS was 18.4 months (95% CI: 10.9–noncalculable [NC]) with osimertinib and 6.9 months (95% CI: 5.3–12.5) with comparator (HR = 0.30; 95% CI: 0.15–0.60) (Fig. 2A). For PD-L1–negative patients (TC < 1%), median PFS was 18.9 months (95% CI: 12.4–NC) with osimertinib and 10.9 months (95% CI: 8.3–12.4) with comparator (HR = 0.37; 95% CI: 0.17–0.74) (Fig. 2B). In the PD-L1 status-unknown group, median PFS was 19.1 months (95% CI: 14.9–23.4) with osimertinib and 10.8 months (95% CI: 9.6–12.3) with comparator (HR = 0.49; 95% CI: 0.39–0.62) (Fig. 2C). For objective response rate and median duration of response results, see Table 3.

PD-L1 Immune Cell Scoring Outcomes

Most randomized patients (98 of 106 [92%]) had some PD-L1 IC expression (IC ≥ 1%). Outcomes of the exploratory IC analysis are in the Supplementary Data.

Discussion

In this analysis of PD-L1 expression among screened patients in the FLAURA trial, tumor EGFR mutations and PD-L1 expression were not mutually exclusive; however,

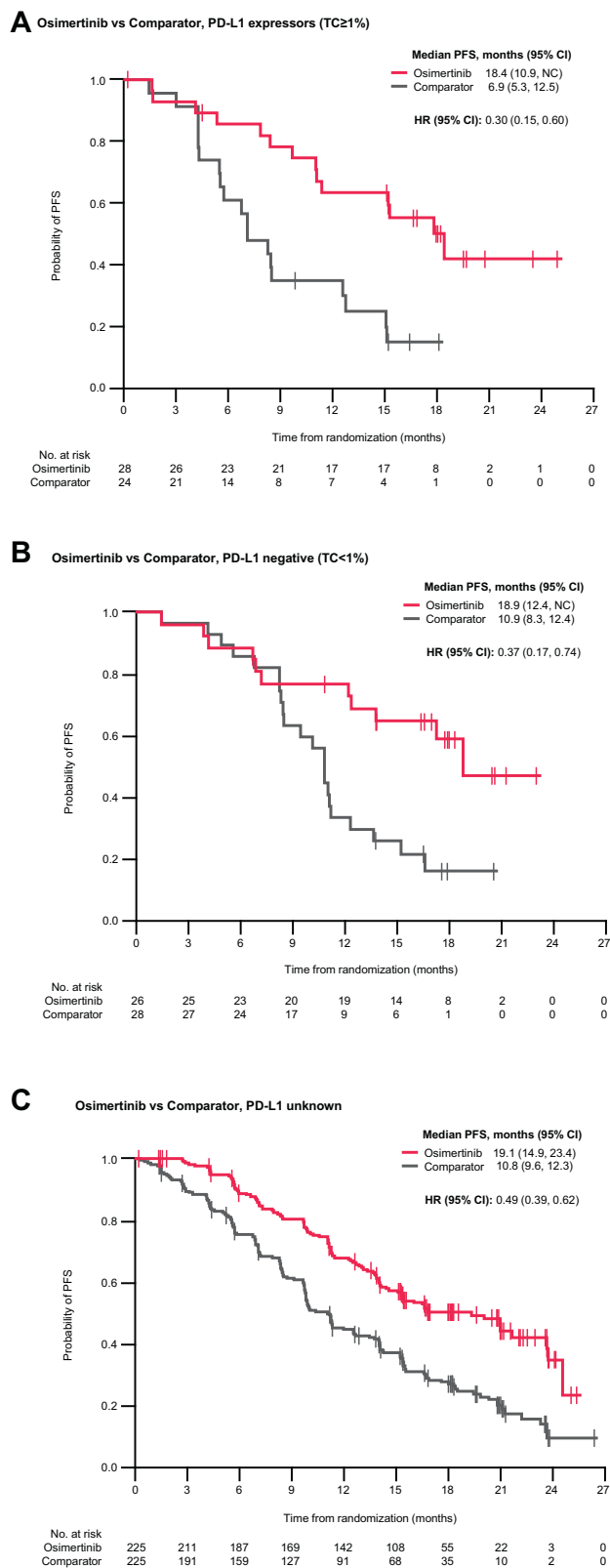


Figure 2. PFS by treatment group according to tumor cell PD-L1 expression (TC 1% cutoff) in (A) PD-L1 expressors (TC ≥ 1%), (B) PD-L1 negatives (TC < 1%), and (C) PD-L1 unknown. CI, confidence interval; HR, hazard ratio; PD-L1, programmed cell death ligand 1; PFS, progression-free survival; TC, tumor cell; NC, noncalculable.

Table 3. Response Rates Across Treatments and PD-L1 Status

	Osimertinib (n = 54)			Comparator (n = 52)		
	PD-L1 Expressers	PD-L1 Non-Expressers	PD-L1 Status Unknown	PD-L1 Expressers	PD-L1 Non-Expressers	PD-L1 Status Unknown
Response, n (%)	n=28 22 (79)	n=26 22 (85)	n=225 179 (80)	n=24 17 (71)	n=28 23 (82)	n=225 170 (76)
Median DoR, mo (95% CI)	n=22 17.2 (10.0-NC)	n=22 NC (11.0-NC)	n=179 17.6 (12.5-21.9)	n=17 6.9 (2.9-13.8)	n=23 8.8 (6.9-11.1)	n=170 8.5 (7.2-11.0)

CI, confidence interval; DoR, duration of response; NC, noncalculable; PD-L1, programmed cell death ligand 1.

a lower proportion of patients with EGFRm-positive tumors expressed PD-L1 versus patients with EGFRm-negative tumors. This difference was more pronounced above the threshold of PD-L1 TC greater than or equal to 25%. These data are in agreement with a pooled analysis of 18 studies that found a 41% lower likelihood of EGFRm-positive tumors expressing PD-L1 versus EGFRm-negative tumors (odds ratio = 0.59; 95% CI: 0.38–0.92; $p < 0.02$).¹¹

PFS analysis showed benefit with osimertinib regardless of PD-L1 expression with both the SP263 assay TC classification (TC \geq 1%) and an exploratory IC classification (IC \geq 1%), suggesting that the PD-L1 status is not prognostic of outcomes with osimertinib therapy. Furthermore, median PFS with osimertinib was closely aligned across the PD-L1 subgroups, as well as to the median PFS of osimertinib for the overall FLAURA study population (18.9 months).² Regarding response, results were similar to that of the overall FLAURA population for all PD-L1 subgroups, suggesting that expression of PD-L1 does not affect response, although this group size was small. Here, the scoring algorithm of the SP142 assay was chosen for the exploratory IC analysis as another study; using this assay had indicated that in patients with EGFRm advanced NSCLC and treated with an EGFR TKI, PD-L1 expression was associated with poor response and a significantly shorter PFS than in patients with no/low PD-L1 expression.¹² As shown previously, this is not the case in the present study.

The numerical difference in PFS by PD-L1 expression in the EGFR TKI comparator group correlates with findings from retrospective analyses of studies in patients with EGFRm advanced NSCLC treated with EGFR TKIs, in which a shorter median PFS was observed in patients with PD-L1 expression versus no expression.^{12,13} However, the literature is inconsistent, with other studies showing high PD-L1 expression results and improved outcomes in patients with NSCLC treated with gefitinib or erlotinib.^{13,14} This inconsistency may be due to a number of factors, including differences in PD-L1 threshold cutoffs, and differences among patients, including previous treatments received.¹¹

Because data on efficacy of EGFR TKIs in patients with EGFRm NSCLC and PD-L1 expressing tumors are inconsistent, and many trials in the first-line setting exclude patients with EGFRm NSCLC altogether, treating these patients with an immune checkpoint inhibitor as first-line therapy, instead of an EGFR TKI, may be a suboptimal option.^{8,9,11} This has been shown in a phase II study investigating pembrolizumab monotherapy as first-line treatment for patients with EGFRm- and PD-L1-positive advanced NSCLC that closed enrollment early due to futility.¹⁰ Clinical practice guidelines recommend that patients who are EGFRm-positive should be treated with an EGFR TKI regardless of PD-L1 status.¹⁵

Strengths of this study include that all data were from a single prospective global trial, and central testing was used for PD-L1 expression. As this was an exploratory endpoint of the FLAURA trial, a limitation was that viable tissue samples were only available in a subset of the randomized patients. Further limitations were that no data were captured on post-study immunotherapy for these patients and tumor mutational burden was not recorded; explorations of both would have provided further insight. Tumor mutational burden and immunotherapy response are now being investigated with samples from the FLAURA trial.

In conclusion, our analysis of tissue samples from patients enrolled in the FLAURA study supports the use of osimertinib as first-line treatment for patients with EGFRm advanced NSCLC regardless of PD-L1 expression.

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Supplementary Data

Note: To access the supplementary material accompanying this article, visit the online version of the *Journal of Thoracic Oncology* at www.jto.org and at <https://doi.org/10.1016/j.jtho.2019.09.009>.

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