



Efficacy of Atezolizumab in Patients With Advanced NSCLC Receiving Concomitant Antibiotic or Proton Pump Inhibitor Treatment: Pooled Analysis of Five Randomized Control Trials

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ABSTRACT

Introduction: Gut dysbiosis may reduce immune checkpoint inhibitor (ICI) efficacy. Antibiotics and proton pump inhibitors (PPIs) are commonly used drugs causing gut dysbiosis. There is limited randomized controlled trial (RCT) evidence on whether antibiotics or PPIs impact ICI benefit versus comparator treatments.

Methods: This study pooled five RCTs (IMpower130, IMpower131, IMpower150, OAK, and POPLAR) evaluating atezolizumab in advanced NSCLC. Atezolizumab efficacy (hazard ratio with 95% confidence intervals) was assessed for subgroups on the basis of antibiotic and PPI use at randomization. The association between antibiotic and PPI use with pretreatment peripheral blood immunophenotype was also explored.

Results: Of 4458 participants, 285 received an antibiotic in the 30-day pretreatment and 1225 were using a PPI at treatment initiation. Overall survival efficacy of atezolizumab was similar (p [interaction] = 0.35) for antibiotic users (hazard ratio 95% confidence interval: 0.73 [0.53–0.99]) and antibiotic nonusers (0.82 [0.74–0.91]). Nevertheless, efficacy was reduced (p [interaction] = 0.003) for PPI users (1.00 [0.85–1.17]) compared with PPI nonusers (0.76 [0.69–0.83]). Findings were consistent across RCTs and for progression-free survival. PPI use was associated with 9%, 18%, and 9% lower counts of lymphocytes, CD19+, and CD16+CD56+ immune cells, respectively ($p < 0.01$).

Conclusions: Reassuringly, atezolizumab efficacy did not differ for antibiotic users. Oppositely, PPI use was consistently associated with decreased atezolizumab efficacy and lower pretreatment counts of lymphocytes, CD19+, and

CD16+CD56+ immune cells. Given that approximately 30% of patients with cancer use PPIs, there is an urgent need for evidence on the impacts of PPIs on other ICIs and for the development of guidelines on nonessential PPI use with ICIs.

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Keywords: Atezolizumab; Non-small cell lung cancer; Antibiotics; Proton pump inhibitors; Gut dysbiosis; Survival

Introduction

The gut microbiota plays a profound role in regulating homeostasis and immune function, and a disrupted gut bacteria ecosystem (gut dysbiosis) can negatively impact adaptive immunity (e.g., T cell, B cell,

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Dr. Hopkins and Ms. Badaoui contributed equally to this work.

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and natural killer cell expressions) and the efficacy of immune checkpoint inhibitors (ICIs).¹⁻⁴ Although ICIs are an important step forward in the treatment of advanced NSCLC, their use is associated with considerable heterogeneity in survival benefit between patients.⁵ Coincidentally, much research is being conducted to identify factors associated with ICI resistance, as current markers do not reliably predict treatment benefit. One hypothesis is that commonly used concomitant non-cancer medicines, such as antibiotics and proton pump inhibitors (PPIs), which significantly disrupt the gut microbiota, may negatively influence the efficacy of ICIs.⁶⁻¹¹

Antibiotics cause rapid and ubiquitous decreases in the diversity of gut bacteria, which can persist for months after completion. PPIs likewise cause profound gut dysbiosis, occurring through both direct compound effects and altered stomach acidity.^{12,13} PPIs are also often used as long-term medications in patients with cancer, leading to the hypotheses that their impacts on ICIs could be profound and long lasting. Much recent research has revealed that antibiotic and PPI use may be associated with poor prognosis in patients treated with ICI therapies.^{7-11,14-17} Nevertheless, the research has largely been based on small single-arm studies, leading to conflicting results and an inability to validly infer whether antibiotics or PPIs specifically impact ICI efficacy (i.e., the ability of treatment to improve outcomes) or whether they just identify poor prognosis. In addition, the potential impact of antibiotics and PPIs on immune system functioning is poorly understood.

Conducting prospective randomized trials to evaluate the impact of antibiotics and PPIs on the efficacy of ICIs has limited feasibility owing to required sample size, cost of trials, and urgency of quality evidence. This highlights the importance of well-designed post hoc analyses to answer this question. Three recent post hoc analyses of randomized controlled trials (RCTs) have suggested that both antibiotics and PPIs may be associated with decreased ICI efficacy, but the studies were insufficiently powered to be conclusive.⁸⁻¹¹ Pooled analysis of multiple RCTs enables superior statistical power.¹⁸ An additional limitation of the studies was the inclusion of antibiotic and PPI use in the 30 days after anticancer treatment commencement,⁸⁻¹¹ which compromises randomized design and the validity of evaluating treatment efficacy.¹⁹ Guidelines outline that when evaluating the heterogeneity of anticancer treatment efficacy, subgroup discrimination should be based on information available before treatment commencement (pretreatment) to avoid the introduction of multiple biases.¹⁸⁻²⁰

In a pooled analysis of individual-participant data from five RCTs, this study aimed to (1) evaluate whether patients with advanced NSCLC using antibiotics or PPIs

have reduced atezolizumab efficacy and (2) evaluate the association of antibiotic and PPI use with differences in peripheral blood immunophenotypes.

Material and Methods

Population

The study was a pooled post hoc analysis of individual-participant data from RCTs IMpower130 (NCT02367781, March 15, 2018, data cutoff),²¹ IMpower131 (NCT02367794, April 20, 2018, data cutoff),²² IMpower150 (NCT02366143, September 15, 2017, data cutoff),²³ OAK (NCT02008227, July 7, 2016, data cutoff),²⁴ and POPLAR (NCT01903993, December 1, 2015, data cutoff).²⁴ The RCTs were conducted in accordance with International Conference on Harmonisation Good Clinical Practice guidelines and the Declaration of Helsinki, with participants providing written informed consent.²¹⁻²⁴ The research was deemed negligible risk research and exempt from review by the Southern Adelaide Clinical Human Research Ethics Committee.

Briefly, IMpower130 was a phase 3 RCT evaluating (2:1) atezolizumab (1200 mg intravenous every 3 wk) plus carboplatin plus nab-paclitaxel (ACnP) versus carboplatin plus nab-paclitaxel (CnP) in patients with chemotherapy-naïve, stage IV nonsquamous NSCLC.²¹ IMpower131 was a phase 3 RCT evaluating (1:1:1) atezolizumab plus carboplatin plus paclitaxel (ACP) versus ACnP versus CnP in patients with chemotherapy-naïve, stage IV squamous NSCLC.²² IMpower150 was a phase 3 RCT evaluating (1:1:1) ACP versus bevacizumab plus carboplatin plus paclitaxel (BCP) versus atezolizumab plus BCP (ABCP) in patients with chemotherapy-naïve, metastatic nonsquamous NSCLC.²³ OAK was a phase 3 RCT evaluating (1:1) atezolizumab versus docetaxel in patients with NSCLC who had received one or more platinum-based combination therapies for stage IIIB or IV disease.²⁴ POPLAR was a phase 2 RCT evaluating (1:1) atezolizumab versus docetaxel in patients with advanced NSCLC whose disease had progressed on platinum-containing therapy.²⁴ Descriptions of treatment, eligibility, recruitment, randomization, and study protocols have been published previously.²¹⁻²⁴ Best practice guidelines informed that five RCTs pooled would provide superior statistical power than prior studies for evaluating treatment effect modification in antibiotic and PPI treatment subgroups.¹⁸⁻²⁰

Predictor and Outcome Data

The primary assessed outcome was overall survival (OS), with progression-free survival (PFS) assessed as a secondary outcome. Primary study definitions of PFS were used. In IMpower130, IMpower131, IMpower150,

OAK, and POPLAR, PFS was assessed by the investigator according to Response Evaluation Criteria in Solid Tumors version 1.1.²¹⁻²⁴ The primary assessed predictor variables were any antibiotic use occurring within a period of 30 days before treatment initiation and any PPI use documented as continuing the day of treatment initiation.

Distributions of pretreatment neutrophil, lymphocyte, and peripheral blood immune cell counts were explored according to antibiotic and PPI use. In consenting participants of the IMpower131, IMpower150, OAK, and POPLAR trials, collected whole blood specimens were analyzed according to standard flow cytometry procedures for quantifying CD3+, CD4+, CD8+, CD19+, and CD16+CD56+ immunophenotypes.²²⁻²⁴ Other available pretreatment variables included age, sex, race, Eastern Cooperative Oncology Group performance status (ECOG PS), smoking status, tumour histology, presence of liver metastases, programmed death-ligand 1 (PD-L1) expression, effector T-cell gene signature score, and blood-based tumor mutation burden.²¹⁻²⁴

Statistical Analysis

Adjusting for between-trial differences, this study used a two-stage individual-participant data meta-analysis approach,¹⁸ whereby hazard ratios (HRs) and 95% confidence intervals (CIs) were estimated individually for each trial using Cox proportional hazards regression. HRs and 95% CI were then pooled and presented in forest plot. *p* values less than 0.05 were considered statistically significant. On a trial-by-trial basis, the prognostic association of antibiotic and PPI use with survival outcomes in atezolizumab-treated participants was modeled. Analyses adjusted for age, sex, race, ECOG PS, smoking status, tumour histology, presence of liver metastases, and PD-L1 expression were conducted (complete case analyses). Heterogeneity in atezolizumab efficacy was assessed using an antibiotic or PPI-by-treatment interaction term. Evaluating the statistical interaction of antibiotic and PPI use on atezolizumab efficacy was based on the prospective randomized design of each trial and used their intent-to-treat [ITT] populations defined at the randomization date. Survival probabilities were estimated by means of Kaplan-Meier analysis. All analyses used the R statistical environment (version 3.6.2).

As an exploratory analysis, linear regression was used to evaluate the percentage difference in neutrophil, lymphocyte, CD3+, CD4+, CD8+, CD19+, and CD16+CD56+ peripheral blood immune cell counts according to antibiotic and PPI use status (i.e., evaluating the association of antibiotic and PPI use with biomarkers of the immune system).

As a sensitivity analysis, a 30-day landmark Cox proportional hazard analysis was used to evaluate the association between survival outcomes and antibiotics initiated early after atezolizumab commencement (i.e., evaluating the prognosis of participants requiring antibiotics within the 30-d post-atezolizumab commencement).

Results

Population

In a pooled cohort of 4458 participants, 2723 were randomized to treatment including atezolizumab and 1735 to treatment without atezolizumab (Supplementary Table 1). In the participants randomized to treatment including atezolizumab, 194 (7%) received an antibiotic within the 30-day pretreatment window and 762 (28%) were documented as actively using a PPI on the day of treatment initiation. In the participants randomized to treatment without atezolizumab, 91 (5%) received an antibiotic within the 30-day pretreatment window and 463 (27%) were documented as actively using a PPI on the day of treatment initiation. Median (95% CI) follow-up was 19 (18-19) months within the pooled cohort (Supplementary Table 2).

Supplementary Table 3 presents patient characteristics by antibiotic use status; of note, antibiotic use was associated with Asian race, higher ECOG PS, and higher PD-L1 expression (*p* < 0.05). Of the total 285 participants using an antibiotic, 77 (27%) used quinolones, 61 (21%) penicillins, 39 (14%) cephalosporins, 19 (7%) macrolides, 18 (6%) tetracyclines, 12 (4%) sulfonamides, 11 (4%) lincosamides, four (1%) nitrofurans, four (1%) aminoglycosides, two (>1%) glyco or polypeptides, one (>1%) carbapenem, and 37 (13%) other antibiotics. Conditions for antibiotic use included for prevention or prophylaxis (*n* = 76), respiratory tract infection (*n* = 69), urinary tract infection (*n* = 23), other infections (*n* = 76), and infection unspecified (*n* = 41).

Supplementary Table 4 presents patient characteristics by PPI use status; of note, PPI use was associated with older age, white race, higher ECOG PS, previously smoking, and a higher blood-based tumor mutation burden (*p* < 0.05). Of the total 1225 participants using a PPI, 559 (46%) were using omeprazole, 391 (32%) pantoprazole, 136 (11%) esomeprazole, 104 (8%) lansoprazole, 27 (2%) rabeprazole, five (>1%) dexlansoprazole, and three (>1%) vanoprazan. Of the PPI users, 96% (*n* = 1178) were using the PPI for either gastric protection (*n* = 568), gastroesophageal reflux disease (*n* = 431), gastritis (*n* = 55), ulcer (*n* = 51), dyspepsia (*n* = 21), epigastric pain or discomfort (*n* = 19), gastrointestinal disorder (*n* = 11), nausea (*n* = 11), and hernia (*n* = 10).

Antibiotic Use and Prognosis

In patients randomized to atezolizumab, no significant association between antibiotic use (within 30 d before atezolizumab initiation) and OS was identified on univariable (HR [95% CI] = 1.19 [0.98–1.45], $p = 0.07$) or adjusted (1.17 [0.96–1.42], $p = 0.13$) analysis (Supplementary Fig. 1). Similarly, no significant association between antibiotic use and PFS was identified on univariable (1.01 [0.85–1.19], $p = 0.93$) or adjusted (0.95 [0.80–1.12], $p = 0.53$) analysis (Supplementary Fig. 2).

In the comparator arms randomized to therapies without atezolizumab, antibiotic use (within 30 d before treatment initiation) was identified as prognostically associated with worse OS (HR [95% CI] = 1.45 [1.13–1.86], $p = 0.003$) and PFS (1.50 [1.21–1.87], $p < 0.001$) (Supplementary Fig. 3).

On exploratory analysis, it was observed that the frequency of postrandomization antibiotic use (within the 30-d post-initiation of atezolizumab) was 215% greater than the frequency of antibiotic use within the 30 days before treatment initiation ($n = 612$ versus 194, respectively)—an indication that early after atezolizumab plus or minus chemotherapy initiation, there is an increase in the frequency of antibiotic use. Antibiotic use within the 30-day post-atezolizumab commencement was prognostically associated with worse OS and PFS on landmark univariable and adjusted analysis (Table 1).

Antibiotic Use and Atezolizumab Efficacy

In the pooled ITT population, atezolizumab OS efficacy (HR 95% CI of atezolizumab versus comparator arms) was 0.73 (0.53–0.99) for antibiotic users (within 30 d before atezolizumab initiation), compared with 0.82 (0.74–0.91) for antibiotic nonusers (p [interaction] = 0.35; Fig. 1A and B)—revealing patients who had used an antibiotic did not have reduced atezolizumab efficacy. The pooled atezolizumab PFS efficacy (HR 95% CI) was

0.65 (0.49–0.85) for antibiotic users, compared with 0.81 (0.70–0.93) for antibiotic nonusers (p [interaction] = 0.02; Supplementary Fig. 4)—indicative that antibiotic use was statistically associated with an increase in the magnitude of atezolizumab PFS benefit.

Matching the methodology of prior analyses,^{8,10} a sensitivity analysis of antibiotic use occurring within the period of 30 days before 30-day post-treatment initiation was conducted and similarly revealed no association with altered atezolizumab OS or PFS efficacy (Supplementary Figs. 5 and 6, respectively).

PPI Use and Prognosis

In patients randomized to atezolizumab, PPI use was associated with worse OS on univariable (HR [95% CI] = 1.30 [1.17–1.46], $p < 0.001$) and adjusted (1.23 [1.09–1.37], $p < 0.001$) analyses (Fig. 2A and B). In addition, PPI use was associated with worse OS consistently across the IMpower130, IMpower131, IMpower150, OAK, and POPLAR atezolizumab-treated cohorts (Fig. 2A and B). Similarly, PPI use was associated with worse PFS on univariable (1.18 [1.07–1.29], $p < 0.001$) and adjusted (1.15 [1.03–1.28], $p = 0.01$) analyses (Supplementary Fig. 7). Supplementary Figures 8 and 9 present Kaplan-Meier estimates of OS and PFS for PPI users versus nonusers by study and treatment arm, respectively.

In the comparator arms randomized to therapies without atezolizumab, no association between PPI use and altered OS (HR [95% CI] = 1.01 [0.88–1.16], $p = 0.87$) or PFS (0.95 [0.81–1.12], $p = 0.55$) was observed (Supplementary Fig. 10).

PPI Use and Atezolizumab Efficacy

In the pooled ITT population, atezolizumab OS efficacy (HR 95% CI of atezolizumab versus comparator arms) was 1.00 (0.85–1.17) for PPI users, compared with 0.76 (0.69–0.83) for PPI nonusers (p [interaction] = 0.003, Fig. 3A and B)—revealing PPI

Table 1. Association Between Antibiotic Use and Survival Outcomes in a 30-Day Landmark Cox Proportional Hazard Analysis

Evaluation	n	Univariable		Adjusted ^a	
		HR [95% CI]	p	HR [95% CI]	p
Overall survival					
No antibiotic	1899	1	0.01	1	<0.001
Antibiotic in the 30 days before treatment initiation	184	1.22 [0.99-1.49]		1.23 [1.00-1.51]	
Antibiotic in the 30 days post-treatment initiation	518	1.20 [1.05-1.37]		1.32 [1.15-1.51]	
Progression-free survival					
No antibiotic	1885	1	0.007	1	<0.001
Antibiotic in the 30 days before treatment initiation	176	0.98 [0.82-1.17]		0.95 [0.79-1.14]	
Antibiotic in the 30 days post-treatment initiation	499	1.19 [1.07-1.33]		1.23 [1.10-1.37]	

Note: All analyses stratified by study and arm.

^aAnalysis adjusted for age, sex, race, ECOG PS, smoking status, tumour histology, presence of liver metastases, and PD-L1 expression.

CI, confidence interval; ECOG PS, Eastern Cooperative Oncology Group performance status; HR, hazard ratio; PD-L1, programmed death-ligand 1.

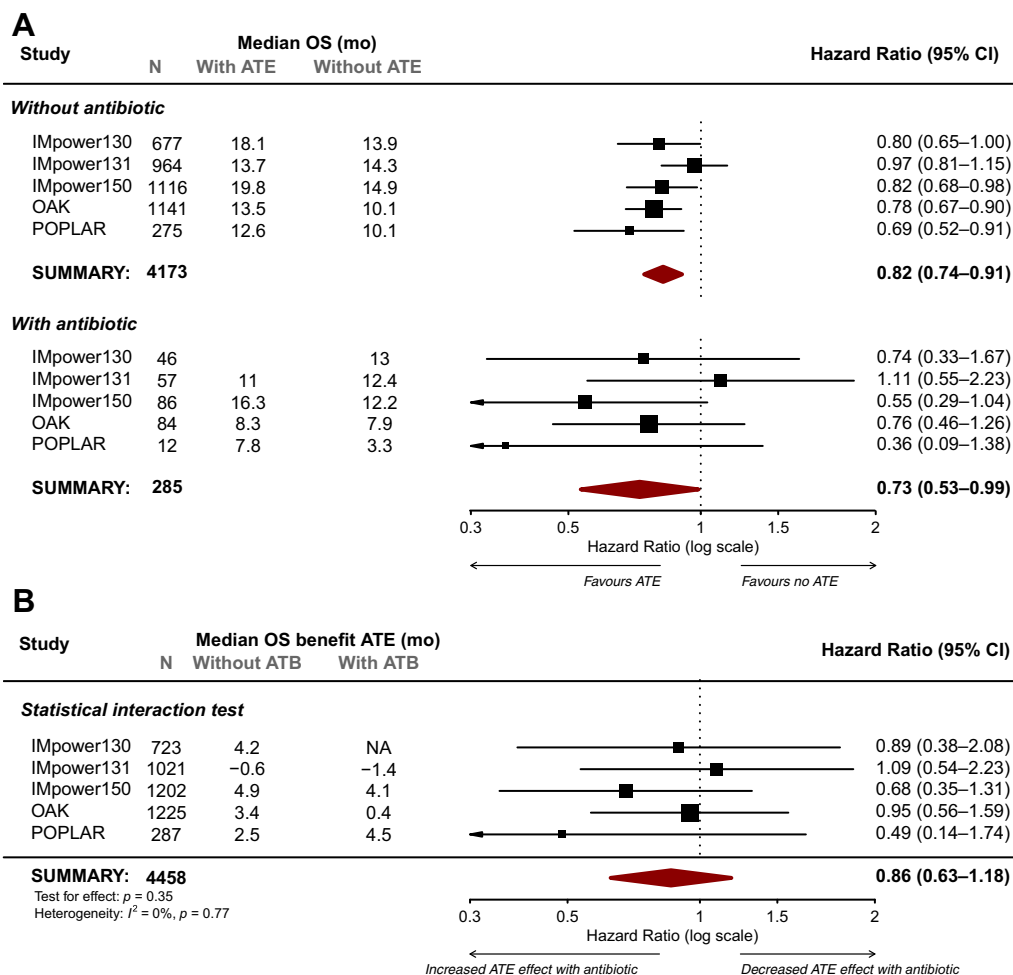


Figure 1. (A) Forest plot of atezolizumab OS efficacy (versus therapies without atezolizumab) according to antibiotic use status by study. (B) Forest plot of antibiotics modification of atezolizumab OS efficacy by study (statistical interaction test). ATB, antibiotic; ATE, atezolizumab; CI, confidence interval; OS, overall survival.

use was statistically associated with a decrease in the magnitude of atezolizumab OS benefit. Figure 3A and B present forest plots indicative of PPI use being associated with worse atezolizumab OS efficacy consistently across the IMpower130, IMpower131, IMpower150, OAK, and POPLAR trials—indicating consistent findings across first-line chemoimmunotherapy and later-line ICI monotherapy cohorts. Similarly, the pooled atezolizumab PFS efficacy (HR 95% CI) was 0.93 (0.76–1.13) for PPI users, compared with 0.75 (0.65–0.88) for PPI nonusers ($p[\text{interaction}] = 0.03$; Supplementary Fig. 11). Supplementary Figures 12 and 13 present Kaplan-Meier estimates of OS and PFS, respectively, in the randomized arms of IMpower130, IMpower131, IMpower150, OAK, and POPLAR, subgrouped by PPI use status. Supplementary Figures 14 and 15 present forest plots indicative of PPI use being associated with a reduced magnitude of atezolizumab OS and PFS benefit consistently across PD-L1 expression levels in a pooled interaction analysis, respectively.

Peripheral Blood Immunophenotypes

PPI use was associated with 18% and 9% lower pretreatment counts of CD19+ and CD16+CD56+ peripheral blood immune cells, respectively ($p < 0.01$; Fig. 4). PPI use was also associated with a 9% lower lymphocyte count ($p < 0.01$; Fig. 4). No significant difference in neutrophils, CD3+, CD4+, or CD8+ immune cells was observed according to PPI use (Supplementary Fig. 16).

No significant difference in pretreatment counts of lymphocytes, CD3+, CD4+, CD8+, CD19+, or CD16+CD56+ immune cells was observed according to antibiotic use (Supplementary Fig. 17). Antibiotic use was associated with a 7% higher neutrophil count ($p = 0.04$; Supplementary Fig. 17).

Discussion

In a pooled cohort of 4458 participants with advanced NSCLC, PPI use was associated with inferior atezolizumab OS and PFS efficacy. Nevertheless, there

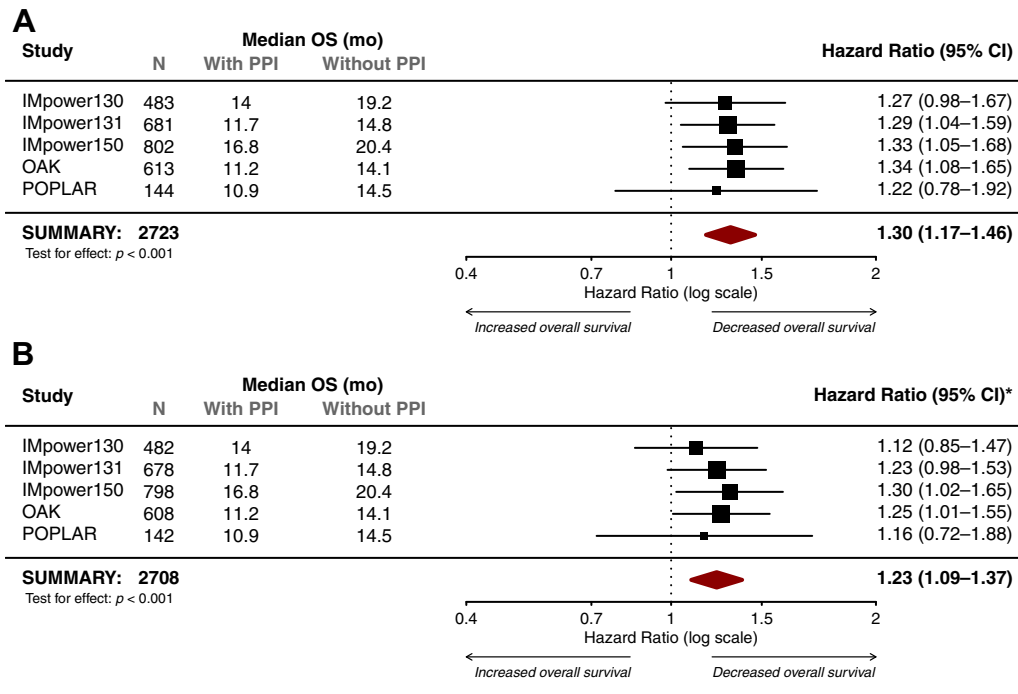


Figure 2. Forest plot of the association between PPI use and OS by study in the cohort-randomized therapies with atezolizumab. (A) Univariable. (B) Adjusted analysis. CI, confidence interval; ECOG PS, Eastern Cooperative Oncology Group performance status; OS, overall survival; PD-L1, programmed death-ligand 1; PPI, proton pump inhibitor.

was no evidence that participants who recently used an antibiotic had any reduction in atezolizumab efficacy. PPI use was identified as associated with lower pretreatment counts of lymphocytes and CD19+ and CD16+CD56+ peripheral blood immune cells. Importantly, our analysis is (1) the largest analysis of antibiotic or PPI use on the basis of RCTs of ICI therapy, (2) did not confound randomized design by including antibiotic or PPI use occurring after treatment initiation, and (3) may provide biological insights on PPI effects on ICI efficacy in advanced NSCLC.

Hypotheses are that antibiotic-induced gut dysbiosis may negatively impact ICI efficacy. Observational studies have revealed that antibiotic use is associated with poor prognosis in patients treated with ICIs.^{7,14,15} Importantly, such studies do not include a matched cohort treated with non-ICI therapies, and thus cannot distinguish whether antibiotic use is a prognostic or predictive (i.e., ICI treatment efficacy) marker. Identification of treatment efficacy modifiers (predictive markers) can only be validly achieved through randomized data.

Recently, Chalabi et al.¹⁰ conducted a post hoc analysis of the OAK and POPLAR RCTs demonstrating a trend toward decreased atezolizumab monotherapy efficacy (versus docetaxel) when antibiotics were used in the period of 30 days before 30-day post-treatment

initiation. The current study includes data from the complete OAK and POPLAR trials and three additional first-line RCTs (an extra 2946 participants). In addition, the current study focuses on antibiotic and PPI use at the time of treatment randomization, thereby avoiding well-known biases resulting from defining subgroups from post-treatment data. Importantly, our study demonstrates that antibiotic use within the 30-day post-initiation of atezolizumab-containing therapy was more than twice as common as antibiotic use within the 30 days before treatment initiation. In addition, antibiotic use within the 30-day post-atezolizumab commencement was prognostically associated with worse survival. This highlights that results of prior studies are likely dominated by post-treatment antibiotic use (commenced after starting ICI therapy) and that post-treatment antibiotic use—which may be influenced by the ICI therapy—has a different prognostic association than pretreatment antibiotic use.

High diversity in gut bacteria and high abundances of *Ruminococcaceae* and *Faecalibacterium* species have been associated with enhanced antitumor immune activity.^{1–4} PPIs significantly lower gut bacteria diversity and are associated with increases in *Actinomycetales*, *Micrococcaceae*, *Enterobacteriaceae*, and *Streptococcaceae*.^{12,13} Evidence further indicates that gut dysbiosis is prevalent in patients with gastrointestinal diseases,

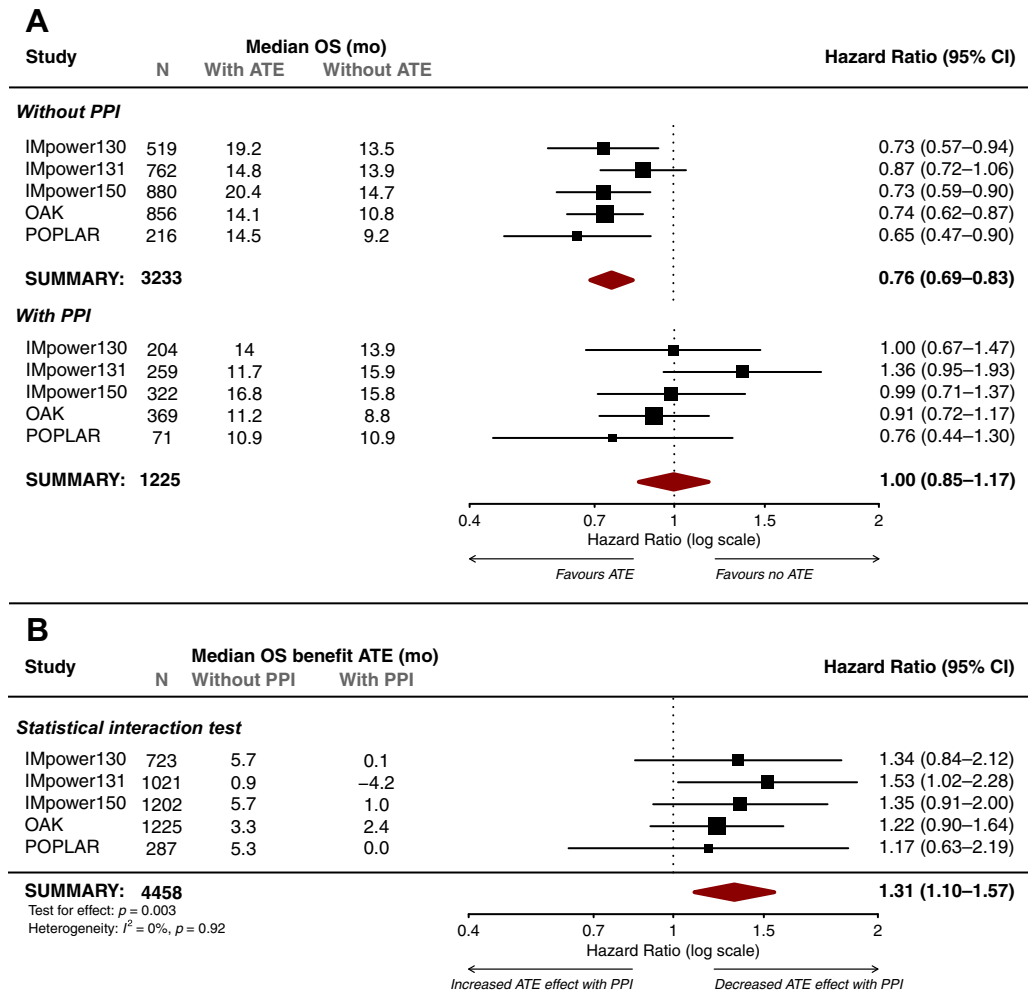
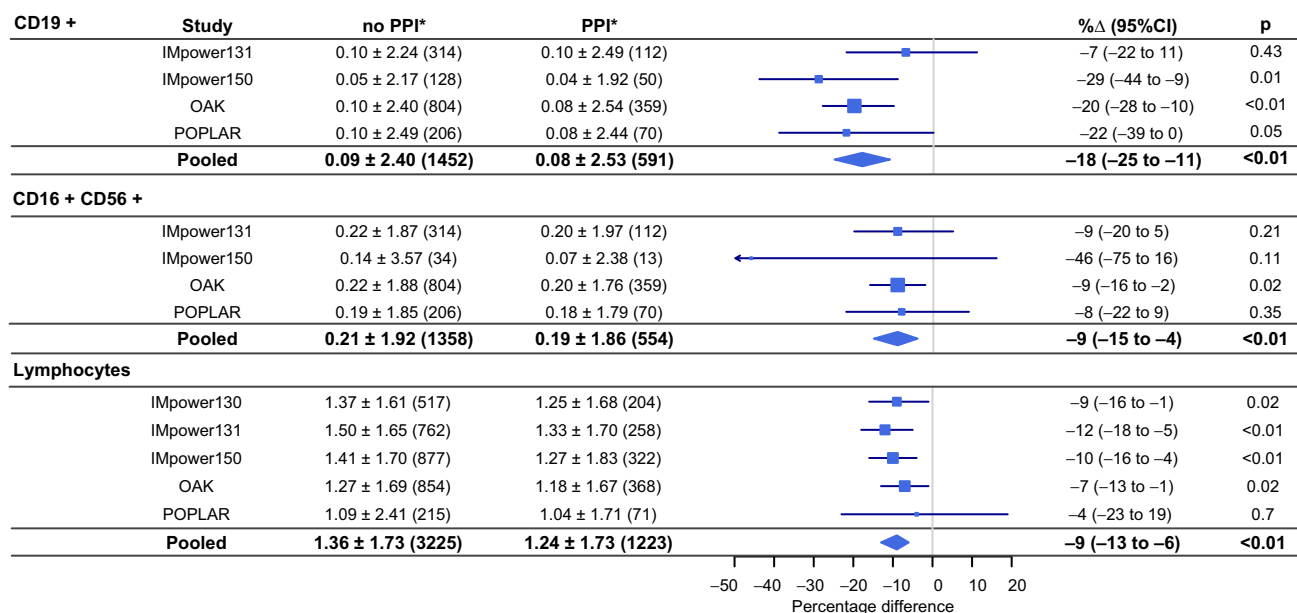


Figure 3. (A) Forest plot of atezolizumab OS efficacy (versus therapies without atezolizumab) according to PPI use status by study. (B) Forest plot of PPI modification of atezolizumab OS efficacy by study (statistical interaction test). ATB, antibiotic; ATE, atezolizumab; CI, confidence interval; OS, overall survival; PPI, proton pump inhibitor.

including conditions such as gastroesophageal reflux disease.^{25,26} These underlying gastrointestinal diseases are also often associated with poor prognostic factors, such as smoking, older age, and higher ECOG PS. Coincidentally, it is hypothesized that PPIs could be a multifaceted clinical marker associated with altered ICI efficacy; and with PPIs so often used in patients with cancer (often for long time periods), the association with ICI efficacy could be profound and long lasting. Nevertheless, most studies on PPIs with ICIs have been small and lacking an appropriate comparator arm,^{16,17} and resultantly findings have been conflicting and inform only prognosis. Chalabi et al.¹⁰ conducted a post hoc analysis of the OAK and POPLAR RCTs, reporting PPIs were trending toward a decrease in the magnitude of atezolizumab benefit compared with docetaxel in the platinum-resistant advanced NSCLC cohort. But the interaction test did not reach statistical significance. This was followed by independent analyses of the

IMvigor211⁹ (later-line advanced urothelial cancer cohort) and IMpower150¹¹ (chemotherapy-naive advanced NSCLC cohort) RCTs. These analyses revealed that PPI use was associated toward a decrease in the magnitude of atezolizumab benefit versus comparator chemotherapies. The key limitations of the above-mentioned studies have been that they were subgroup analyses of single RCTs (resulting in insufficient power and the potential for false positives) and they used a PPI window of 30 days before 30 days after anticancer treatment initiation (compromising randomized design).^{18–20}

The present study found that pretreatment PPI use was prognostically associated with worse OS and PFS in the cohort who initiated therapies containing atezolizumab, whereas no association with altered prognosis was identified in the cohort who initiated comparator chemotherapies. Furthermore, PPI use consistently predicted reduced atezolizumab benefit in patients with



* Denotes geometric mean cell count (10⁹/liter) ± SD (number of participants)

Figure 4. Summary of the percentage difference in lymphocyte, CD19+, and CD16+CD56+ peripheral blood immune cell counts according to PPI use status, by study in the total pooled cohort. PPI, proton pump inhibitor.

advanced NSCLC, including across lines of therapy, for both ICI monotherapy and chemoimmunotherapy, and across PD-L1 expression levels. Future research should aim to evaluate and provide clinical insights to the association of PPI use with the efficacy of other ICIs and with the use of ICIs in other cancer types. Basic research aiming to confirm causality is also warranted, as compensating for confounding through multivariable analysis of post hoc data has limitations.

The mechanism of action of ICIs has yet to be fully elucidated. Nonetheless, antibiotics and PPIs are proposed to indirectly impact ICI actions, which aim to remove inhibitory signals on antitumor T-cell cascades and thus boost and restore the antitumor effects of innate immunity (i.e., in part, ICIs aim to boost the antitumor effects of CD8+ and CD4+ T cells, B cells [CD19+ immune cells], and natural killer cells [CD16+CD56+ immune cells]).^{3,5,27,28} In the present study, it was observed that PPI use was associated with significantly lower expressions of lymphocytes, CD19+, and CD16+CD56+ peripheral blood immune cells. To the best of our knowledge, this study presents the first exploratory analysis of the association between PPI use and peripheral blood immune cell subgroups within an advanced NSCLC cohort and is the first study to provide biological insights on key immune markers for the actions of ICIs within PPI users. Interestingly, no changes in peripheral blood immune cell expressions were observed within antibiotic users. Future research should aim to confirm the impact of PPI and antibiotics on peripheral blood immunophenotypes.

A study limitation was an inability to assess the dose, duration, type, or compliance to antibiotic or PPI therapy. Nonetheless, more than 90% of participants were using PPIs for an indication that likely constituted a need for extended use. Future research should aim to investigate whether H₂-receptor antagonists and other antacids have similar associations with survival outcomes in ICI-treated cohorts, including broader analyses of participants by gastrointestinal disease subtypes, some of which may be associated with greater or lesser gut dysbiosis. Such analyses will require larger data sets than this study. While highlighting the clinical significance of the identified PPI associations, it is also acknowledged that the number of participants administered antibiotics was smaller than the PPI subgroup. Confirming the identified associations for antibiotic use (or lack thereof) with the efficacy of other ICIs is warranted. Finally, whether the findings of this study are validated, prospective studies will be required to determine clinical strategies to manage patients treated with ICIs who develop conditions requiring PPI treatment.

In a pooled analysis of five RCTs, this study concludes that PPI use is a clinical marker identifying reduced atezolizumab efficacy. Reassuringly, there is no evidence that antibiotic use reduces atezolizumab efficacy. On exploratory analysis, PPI use was identified as associated with lower pretreatment counts of lymphocytes, CD19+, and CD16+CD56+ peripheral blood immune cells, and these changes were not observed in antibiotic users. Given that approximately 30% of patients with cancer use PPIs, there is an urgent need for evidence on the

impacts of PPIs on the efficacy of other ICIs and for guidelines on the use of PPIs in patients considering ICIs. Specifically, guidelines may consider calling for a review of PPI use before ICI initiation, and if no appropriate indication for PPI use is identified, consideration to ceasing the PPI may be warranted—this recommendation is based on research indicating PPIs are overprescribed by up to 70%, seemingly from a perspective that they will do no harm.^{29,30}

CRediT Authorship Contribution Statement

Ashley M. Hopkins, Sarah Badaoui, Andrew Rowland, Michael J. Sorich: Conception and design.

Ashley M. Hopkins, Sarah Badaoui, Ganessan Kichenadasse, Christos S. Karapetis, Andrew Rowland, Michael J. Sorich: Development of methodology.

Ashley M. Hopkins, Andrew Rowland, Michael J. Sorich: Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.).

Ashley M. Hopkins, Sarah Badaoui, Ganessan Kichenadasse, Ross A. McKinnon, Christos S. Karapetis, Andrew Rowland, Michael J. Sorich: Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis); Writing, review, or revision of the manuscript.

Ashley M. Hopkins, Ross A. McKinnon, Andrew Rowland, Michael J. Sorich: Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases); Study supervision.

Ethics Approval and Consent to Participate

Secondary analysis of anonymized clinical trial data was confirmed negligible risk research by the Southern Adelaide Local Health Network, Office for Research and Ethics, and was exempt from review.

Data Statement

Data were accessed according to Roche's policy and process for clinical study data sharing and are available on request at vivli.org.

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approval of the manuscript; and decision to submit. This publication is based on research using data from Roche that has been made available through Vivli, Inc. Vivli has not contributed to or approved and is not in any way responsible for the contents of this publication.

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.2022.02.003>.

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