Antitumor Activity of Amivantamab (JNJ-61186372), an EGFR–MET Bispecific Antibody, in Diverse Models of EGFR Exon 20 Insertion–Driven NSCLC

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ABSTRACT

EGFR exon 20 insertion driver mutations (Exon20ins) in non–small cell lung cancer (NSCLC) are insensitive to EGFR tyrosine kinase inhibitors (TKI). Amivantamab (JNJ-61186372), a bispecific antibody targeting EGFR–MET, has shown preclinical activity in TKI-sensitive EGFR-mutated NSCLC models and in an ongoing first-in-human study in patients with advanced NSCLC. However, the activity of amivantamab in Exon20ins-driven tumors has not yet been described. Ba/F3 cells and patient-derived cells/organoids/xenograft models harboring diverse Exon20ins were used to characterize the antitumor mechanism of amivantamab. Amivantamab inhibited proliferation by effectively downmodulating EGFR–MET levels and inducing immune-directed antitumor activity with increased IFNγ secretion in various models. Importantly, in vivo efficacy of amivantamab was superior to cetuximab or poziotinib, an experimental Exon20ins-targeted TKI. Amivantamab produced robust tumor responses in two Exon20ins patients, highlighting the important translational nature of this preclinical work. These findings provide mechanistic insight into the activity of amivantamab and support its continued clinical development in Exon20ins patients, an area of high unmet medical need.

SIGNIFICANCE: Currently, there are no approved targeted therapies for EGFR Exon20ins-driven NSCLC. Preclinical data shown here, together with promising clinical activity in an ongoing phase I study, strongly support further clinical investigation of amivantamab in EGFR Exon20ins-driven NSCLC.

INTRODUCTION

Molecular segmentation of advanced non–small cell lung cancer (NSCLC) based on oncogenic driver mutations has improved the overall survival and quality of life for patients with actionable driver mutations, and solidified solid-tumor targeted therapy. Mutations in the EGFR (1, 2) gene constitutively activate downstream growth and survival signaling pathways leading to dependency on the EGFR pathway for tumor growth. Nearly 20% of Caucasians and up to 50% of Asians with lung adenocarcinomas harbor mutations in EGFR (3, 4).

EGFR activating mutations have been reported in the first four exons (18 through 21) of its tyrosine kinase domain. NSCLCs that harbor “classic” EGFR mutations in exons 18, 19, and 21, for example, exon 19 deletions or L858R, are sensitive to treatment with first-, second-, and third-generation EGFR tyrosine kinase inhibitors (TKI) such as erlotinib, afatinib, and osimertinib (5, 6). In contrast, the EGFR exon 20 mutations encompass nucleotides that translate into amino acids at position 762–823, and include a C-helix (762–766) followed by a loop (position 767–775; ref. 7). The insertion mutations of one to seven amino acids in exon 20 form a wedge at the end of the C-helix in EGFR that promotes active kinase conformation. EGFR exon 20 insertion driver mutations (Exon20ins), a distinct and highly heterogeneous subset of NSCLCs, represent 4% to 12% of all EGFR mutations (7, 8). These Exon20ins mutations are generally insensitive to approved EGFR TKIs and are associated with poor prognosis, thus representing an area of high unmet medical needs (6, 9).

Recently, poziotinib and TAK-788 have been undergoing clinical evaluation in patients whose tumors carry EGFR Exon20ins mutations (10, 11). Despite initial promising efficacy, the Zenith 20 trial demonstrated that poziotinib had a low response rate (RR; ~14%) in patients with NSCLC with the EGFR Exon20ins mutation. Furthermore, both poziotinib and TAK-788 have high rates of EGFR wild-type (WT)-driven toxicity due to the lack of selectivity upon Exon20ins compared as with EGFR WT and other kinases, limiting their clinical utility (6).

Amivantamab (JNJ-61186372; Fig. 1A) is an EGFR–MET bispecific antibody with immune cell–directing activity that targets activating and resistant EGFR mutations and MET mutations and amplifications. Ongoing first-in-human studies in patients with advanced, refractory EGFR-mutant NSCLC have demonstrated preliminary clinical activity of amivantamab in patients with diverse EGFR mutations (12, 13). Of note, amivantamab showed promising efficacy (30% RR) with a manageable safety profile in patients with heavily pretreated EGFR Exon20ins NSCLC. Although amivantamab has been reported to harbor activity in preclinical tumor models driven by EGFR mutations sensitive to approved TKIs (e.g., L858R and Exon 19 deletions; ref. 14), its activity has not yet been explored in the context of EGFR Exon20ins.
Here, we comprehensively evaluated the antitumor activity and mechanisms of action (MOA) of amivantamab in multiple EGFR Exon20ins models, including engineered cell lines, patient-derived cells (PDC), and patient-derived xenografts (PDX). We also present evidence of clinical activity in two case studies of patients with EGFR Exon20ins NSCLC treated with amivantamab from an ongoing phase I clinical trial, highlighting the important translational nature of this work.

RESULTS

Amivantamab Inhibits Proliferation of Ba/F3 Cells Harboring Diverse EGFR Exon20ins Mutations

To demonstrate the antitumor activity of amivantamab in the context of Exon20ins, multiple Exon20ins were stably expressed in Ba/F3 cells. Five distinct Exon20ins were introduced (Fig. 1B), all of which have been observed in patients with NSCLC (V769_D770insASV, D770delinsGY, H773_V774insNPH, H773_V774insH, and D770_N771insSVD; refs. 15, 16). In Ba/F3 cells treated with amivantamab ranging from 0.05 to 1 mg/mL, a significant and dose-dependent decrease in Ba/F3 cell viability (P < 0.0001) was observed in all five EGFR Exon20ins mutations (Fig. 1C). In contrast, treatment with the first- and third-generation irreversible EGFR TKIs gefitinib and osimertinib reduced pEGFR in Ba/F3 cells overexpressing the V769_D770insASV, D770delinsGY, and H773_V774insH EGFR Exon20ins mutations. The total EGFR levels were reduced following treatment with amivantamab, compared with those of untreated cells (Fig. 1D, Supplementary Fig. S1A) or cells treated with the IgG1 control antibody (Supplementary Fig. S1C). Consistent with the reduction in EGFR expression levels, the EGFR downstream signaling pathways phospho-ERF (pERF), phospho-AKT (pAKT), phospho-ERK (pERK), and phospho-S6 (pS6) were also significantly reduced following amivantamab treatment (Fig. 1D), suggesting that amivantamab targeted EGFR and inhibited EGFR-related downstream signaling cascades. Similar results were observed in Ba/F3 cells expressing the V769insASV, Y764 insH, and D770_N771insSVD Exon20ins mutations (Supplementary Fig. S1B). Although 100 nmol/L of gefitinib and osimertinib reduced pEGFR in Ba/F3 cells overexpressing D770delinsGY and H773_V774insH, downstream EGFR signaling pathway components were not significantly inhibited, which correlated with the lack of TKI effects on cell viability (Fig. 1E; Supplementary Fig. S1D). In recent studies, poziotinib has shown antitumor activity in EGFR Exon20ins NSCLC (18, 19). We further assessed the cell viability test for poziotinib in Ba/F3 cells overexpressing EGFR Exon20ins (Supplementary Table S1). Consistent with a previous report (18), poziotinib strongly inhibited the cell viability in the mutant EGFR Exon20ins cells (IC50 ranging from 0.8 to 10.9 nmol/L). As reported in a previous study (20), poziotinib also potentially suppressed proliferation of Ba/F3 cells harboring WT EGFR (IC50 = 0.8 nmol/L). To present the selectivity for Exon20ins mutation in a more balanced manner, we compared antiproliferative potency between amivantamab and poziotinib in EGFR Exon20ins mutants over WT EGFR. Poziotinib exhibited lower EGFR Exon20ins–mutant selectivity over WT EGFR compared with amivantamab, suggesting that poziotinib may adversely affect normal tissues, thereby producing substantial toxicities such skin rash and diarrhea (21).

To better understand the mechanisms involved in amivantamab-mediated cellular cytotoxicity, we assessed the effect of amivantamab treatment on cell-cycle progression and programmed cell death. In Ba/F3 cells expressing the EGFR D770delinsGY and H773_V774insH EGFR Exon20ins mutations, an accumulation of cells in G1 phase was observed in amivantamab-treated cells compared with vehicle-treated cells (Fig. 1F). As EGFR TKIs have been reported to drive apoptosis in NSCLC cells harboring sensitizing EGFR mutations (22, 23), we investigated whether treatment with amivantamab resulted in engagement of the apoptotic machinery. Amivantamab treatment resulted in the induction of proapoptotic proteins, including BIM and cleaved caspase-3 (Fig. 1G), suggesting that amivantamab, in addition to inhibiting downstream EGFR signaling cascade, also induced apoptosis in a BIM- and caspase-dependent manner.

Amivantamab Displays Antitumor Activity in PDCs and Organoids

To extend our findings from Ba/F3 cells engineered to express the exogenous EGFR Exon20ins mutations, we evaluated the activity of amivantamab in several PDCs harboring the Exon20ins. The antitumor activity of amivantamab and associated mechanistic endpoints were evaluated in PDCs generated from patients harboring P772ins_H773insPNNP (DFCI-127), H773_V774insNPH (DFCI-58), and S768_D770dup (YU-1163) EGFR Exon20ins mutations (Supplementary Fig. S2A–S2C; Supplementary Table S2). In both DFCI-127 and DFCI-58 cells, amivantamab treatment resulted in decreased expression of total EGFR and MET levels, as well as inhibition of pEGFR, pMET, pAKT, pERK, and pS6 (Fig. 2A), consistent with the results observed in Ba/F3 cell lines harboring EGFR Exon20ins mutations. Analysis of cell viability and colony formation revealed that amivantamab dose-dependently inhibited the cell growth and proliferation of PDCs, compared with IgG1 controls (Fig. 2B and C). In contrast to the significant reduction in EGFR, MET, pEGFR, pMET, pAKT, pERK, and pS6 in DFCI-127 and DFCI-58 cells, YU-1163 treated with amivantamab unexpectedly revealed an induction of pERK (Fig. 2A). Consistent with this result, the growth of YU-1163 was not inhibited after amivantamab treatment for 72 hours or following long-term treatment (Fig. 2B and C). From the whole-exome sequencing data of YU-1163, we observed a co-occurring mutation in the TP53 gene (R280T; 96% of mutant allele frequency; Supplementary Fig. S2C and S2D). According to recent studies, mutations...
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**Figure 1.** Amivantamab shows antitumor activity and suppresses EGFR and MET signaling pathways in Ba/F3 cells with EGFR Exon20ins mutations. **A**, Schematic of the structure of amivantamab, an EGFR and MET bispecific antibody. **B**, Schematic of EGFR Exon20 insertions in stable Ba/F3 cells. **C**, Schematic of the structure of amivantamab, an EGFR and MET bispecific antibody. EGFR-dependent apoptosis were induced during amivantamab treatment. The expression of BIM and cleaved caspase-3 were detected by Western blotting. **D**, Ba/F3 cells overexpressing the indicated Exon20ins mutations were treated with osimertinib or gefitinib for 6 hours at the indicated concentrations. Immunoblot analysis was performed for EGFR, MET, AKT, ERK, and S6 expression after amivantamab treatment. Amivantamab inhibited the cell cycle and induced synergistic apoptosis in Ba/F3 cells overexpressing the EGFR Exon20ins mutations. **E**, Ba/F3 cells overexpressing the indicated Exon20ins mutations were treated with amivantamab for 72 hours at the indicated concentrations. Immunoblot analysis was performed for EGFR, MET, AKT, ERK, and S6 expression after amivantamab treatment. Amivantamab inhibited the cell cycle and induced synergistic apoptosis in Ba/F3 cells overexpressing the EGFR Exon20ins mutations. Cell cycles were analyzed using propidium iodide staining and FACS analysis. Data are presented as averages ± SD of triplicate independent experiments. *P < 0.0001; **P < 0.001; Student t test. **G**, Ba/F3 cells overexpressing the indicated Exon20ins mutations were treated with amivantamab for 72 hours. Data are presented as averages ± SD of triplicate independent experiments. *P < 0.0001; Student t test. **G**, Ba/F3 cells overexpressing the indicated Exon20ins mutations were treated with amivantamab for 72 hours. Data are presented as averages ± SD of triplicate independent experiments. *P < 0.0001; Student t test.
Figure 2. Amivantamab has antitumoral activity and suppresses EGFR and MET signaling pathways in PDCs and PDOs harboring EGFR Exon20ins mutations. **A**, PDCs with the indicated EGFR Exon20ins mutations were treated with amivantamab for 72 hours at the indicated concentrations. Immunoblot analysis was performed for EGFR, MET, AKT, ERK, and S6 expression after amivantamab treatment. **B**, The viability of PDCs was determined via CellTiter-Glo. Amivantamab was treated for 72 hours. Data are presented as averages ± SD of triplicate independent experiments. *, P < 0.0001; **, P < 0.001; Student t test. **C**, Effects of amivantamab on the colony formation and cell proliferation of PDCs. Representative images and quantitative analysis of the colony formation assay. Data are presented as averages ± SD of triplicate independent experiments. *, P < 0.0001; Student t test. O.D., optical density. **D and E**, Dose–response curves of **D** YUO-036 (A767_V769dup) and **E** YUO-029 (S768_D770dup) PDOs treated with IgG1 control or amivantamab. Cell viability was measured using CellTiter-Glo 3D cell viability reagent 72 hours after drug treatment. Representative images of PDOs treated with amivantamab for 72 hours at the indicated concentrations. Data are presented as averages ± SD of triplicate independent experiments. *, P < 0.0001; Student t test; NS, not significant.
in TP53 commonly occurred with EGFR mutations in NSCLC. Particularly, TP53 mutations in exon 8 in patients with NSCLC with EGFR mutations show lower responsiveness to EGFR TKIs and worse prognosis than patients with WT TP53 (24, 25). Indeed, accumulated studies have revealed that the R280T mutation in TP53 plays crucial roles in the proliferation and survival of cancer cells, and knockdown of the mutant TP53 causes G1-phase arrest and apoptosis in bladder cancer cells (26, 27). As shown in Supplementary Fig. S2E, depletion of mutant TP53 by three different TP53-directed siRNAs significantly inhibited cell proliferation with a reduction in activated ERK in YU-1163 pretreated with 1 mg/mL amivantamab. Given that mutant TP53 is associated with EGFR-TKI resistance (28) and the depleted mutant TP53 restored the sensitivity of amivantamab by downregulation of pERK, induction of pERK following amivantamab treatment in YU-1163 cells might be a key regulator of cell survival, potentially through the cross-talk between mutant TP53 and ERK signaling cascade (29, 30). In addition, we generated two patient-derived organoid (PDO) models from plural effusion of patients who had A767_V769dup (YUO-036) and S768_D770dup (YUO-029) to recapitulate the phenotypic and molecular landscape of the original NSCLC with EGFR Exon20ins (Supplementary Fig. S2F and S2G; Supplementary Table S2). YUO-029 was derived from the same patient from whom YU-1163 PDC (S768_D770dup) was derived. As shown in Fig. 2D, YUO-036 was sensitive to amivantamab in a dose-dependent manner, whereas YUO-029 derived from the same patient with YU-1163 showed no significant decrease in cell viability following amivantamab treatment compared with IgG1 control (Fig. 2E). Taken together, these results indicate that amivantamab has potent antitumor activity in NSCLC patient-derived cancer cells with EGFR Exon20ins mutations by downmodulation of EGFR and MET signaling pathways.

**EGFR and MET Are Internalized in Response to Amivantamab**

Treatment with amivantamab results in downmodulation of EGFR and MET, as observed in Ba/F3 cells (Fig. 1) and PDCs (Fig. 2). According to many studies, anti-EGFR mAb induces internalization of EGFR, leading to downregulation of its expression on the cell surface (31, 32). To investigate whether amivantamab directly binds to EGFR on cells with EGFR Exon20ins mutation, Ba/F3 cells overexpressing D770delinsGY or H773_V774insH were incubated with 0.1 mg/mL IgG1 control and 0.1 mg/mL amivantamab. FACS was used to measure the level of plasma membrane-bound EGFR. EGFR expression on the plasma membrane began to dwindle by almost 2-fold 30 minutes after amivantamab treatment. The percentage changes in median fluorescence intensity (MFI) of EGFR relative to IgG1 control–treated cells at 30 minutes were 56% and 68% in D770delinsGY and H773_V774insH, respectively, and subsequently remained at 40% EGFR expression relative to IgG1 control–treated cells 72 hours after amivantamab treatment (Fig. 3A). To explore the internalization of MET as well as EGFR on PDCs harboring EGFR Exon20ins, DFCI-127 and DFCI-58 PDCs were treated with 0.1 mg/mL amivantamab and the plasma membrane–bound MET and EGFR were measured 72 hours after amivantamab treatment (Fig. 3B and C). The results showed that amivantamab reduced EGFR and MET on PDCs compared with IgG1 control. Immunofluorescence (IF) staining was used to visualize the internalization of EGFR and MET following amivantamab treatment. Treatment with 0.1 mg/mL amivantamab for 72 hours led to the redistribution of EGFR and MET receptors into internal compartments, whereas IgG-treated cells showed no change in the staining pattern for EGFR or MET (Fig. 3D; Supplementary Fig. S3). Internalization and subsequent downregulation of EGFR and MET receptors by lysosomes could account for the decreased EGFR and MET protein levels observed in the immunoblot, FACS, and IF assays following amivantamab treatment. To determine whether lysosomal degradation was involved in downregulating EGFR protein levels, Ba/F3 cells overexpressing D770delinsGY and H773_V774insH were treated with amivantamab in the absence and presence of the autophagy inhibitor bafilomycin. Bafilomycin treatment inhibited the degradation of EGFR (Fig. 3E), suggesting that downmodulation of the total EGFR protein level following amivantamab treatment may involve lysosomal degradation of internalized cell-surface receptors. Taken together, these results suggest that treatment with amivantamab induces receptor internalization and may contribute to the observed antiproliferative effects of amivantamab by inhibiting EGFR- and MET-mediated signaling.

**Amivantamab Inhibits EGFR Exon20ins Mutation-Driven Growth of Ba/F3 and PDC Models In Vivo**

To determine whether amivantamab is active against EGFR Exon20ins–derived tumors in vivo, xenograft models were generated using Ba/F3 cells overexpressing EGFR D770delinsGY and H773_V774insH Exon20ins mutations and PDCs (DFCI-127 and YU-1163) harboring P772insPNP and S768_D770dup EGFR Exon20ins mutations, respectively. Mice were treated with amivantamab, IgG1 control, or vehicle at 30 mg/kg twice per week intraperitoneally. Amivantamab–treated mice showed reduced tumor volumes compared with vehicle– or IgG1 control–treated mice in the Ba/F3 cells bearing NOD.Cg-Prkdcscid I2rgtm1Sug/Jic (NOG) mice models (Fig. 4A and B). Inhibition of tumor growth occurred early and was sustained 15 days following treatment. As shown in Ba/F3 cells and PDCs in vitro (Figs. 1D and 2A), protein expression of EGFR, MET, pEGFR, and pMET were significantly reduced following amivantamab treatment in the Ba/F3-bearing NOG mice models (Fig. 4C; Supplementary Fig. S4C). Similarly, in the PDC xenograft models, amivantamab–treated mice showed a reduction in tumor volume compared with vehicle–treated mice (Fig. 4D–G), as well as a reduction in EGFR, MET, pEGFR, and pMET protein levels (Fig. 4H and I). Intriguingly, although amivantamab could not inhibit the proliferation of YU-1163 PDC in vitro (Fig. 2), a dramatic tumor regression was observed in YU-1163-bearing BALB/c nude mice after amivantamab treatment (Fig. 4F), suggesting that additional factors might contribute to the in vivo antitumor effect of amivantamab. As mentioned above, poziotinib is a targeted agent that has shown preliminary clinical activity in EGFR Exon20ins disease (18, 19). We compared the antitumor activity and safety of poziotinib with those of amivantamab in YU-1163 (S768_D770dup)–bearing BALB/c nude mice and Ba/F3 cells overexpressing D770_N771insSVD-bearing...
Figure 3. Amivantamab strongly promotes internalization of EGFR and MET in Ba/F3 and PDC cells expressing EGFR Exon20ins mutations. The expression of EGFR and MET on the cell surface were determined by FACS analysis. A, EGFR expression on the plasma membrane was detected in Ba/F3 cells overexpressing D770delinsGY and H773_V774insH at the indicated time. After 0.1 mg/mL IgG1 control (cont) or 0.1 mg/mL amivantamab treatment for 72 hours, PE-EGFR and FITC-MET expression on the plasma membrane was detected in (B) DFCI-127 (P772_H773insPNP) and (C) DFCI-58 (H773_V774insNPH) cells. D, Amivantamab induced redistribution of EGFR and MET in DFCI-127 PDCs. IF staining for EGFR (green) and MET (red) in a panel of DFCI-127 treated with 0.1 mg/mL IgG1 control or 0.1 mg/mL amivantamab for 72 hours. E, Pretreatment with the autophagy inhibitor bafilomycin (100 nmol/L) for 30 minutes rescued the decreased EGFR expression in 1 mg/mL amivantamab-treated Ba/F3 cell lines overexpressing D770delinsGY or H773_V774insH.

NOG mice (Supplementary Fig. S4D and S4E). Using the previously reported dosing regimen of 5 mg/kg poziotinib, once a day (18), sudden death occurred within 6 days of treatment. Skin toxicity analyses with poziotinib and amivantamab revealed that poziotinib-treated mice showed severe skin toxicities on the face, abdomen, and back at dose of 5 and 10 mg/kg, whereas 30 mg/kg amivantamab showed only minimal keratosis on the face (Supplementary Fig. S4F and S4G). In addition to skin toxicity, a dramatic loss of body weight was observed in poziotinib-treated mice compared with amivantamab-treated mice (Supplementary Fig. S4H). The favorable toxicity profiles with amivantamab were consistent with those shown in an ongoing phase I study (13).

Amivantamab Induces Antibody-Dependent Cell-Mediated Cytotoxicity in Exon20ins Models

The process of antibody-dependent cell-mediated cytotoxicity (ADCC) is known to be initiated when both the target
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Amivantamab in NSCLC with EGFR Exon20Ins mutations cocultured with peripheral blood mononuclear cells (PBMCs) as effector cells [effector:target (E:T) = 50:1]. Treatment with amivantamab resulted in cytotoxicity in both PDCs in a dose-dependent manner and to a greater extent than cetuximab, a mAb targeting EGFR (Fig. 5A–C). By extension, cetuximab treatment led to a less-pronounced reduction in tumor volume in YU-1163–bearing BALB/c nude mice relative to that observed with amivantamab (Supplementary Fig. S5A). Amivantamab-mediated cellular cytotoxicity shown in Fig. 5A was significantly impaired by incubation with an Fc receptor (FcR) blocker in DFCI-127 and YU-1163 PDCs (Fig. 5D), suggesting that the amivantamab-mediated ADCC effect requires the interaction with FcRs on PBMCs. Similarly, the antitumor effect of amivantamab was abrogated in vivo when amivantamab was cotreated with anti-mouse CD16/CD32 antibodies to block FcγRII/FcγRI on monocytes/macrophages and NK cells in YU-1163–bearing BALB/c nude mice (Supplementary Fig. S5A). It is known that inflammatory cytokines such as IFNγ and TNFα are secreted from infected monocytes and activated NK cells during ADCC, encouraging antigen presentation and adaptive immune responses (34, 35). To explore the correlation between amivantamab-dependent ADCC and secreted IFNγ levels, we measured the level of IFNγ in a medium cocultured with PDCs and PBMC after amivantamab treatment. Consistent with the degree of the ADCC effect, IFNγ levels were significantly increased with amivantamab treatment compared with cetuximab treatment (Fig. 5E). Treatment with a FcR blocker reduced IFNγ secretion, indicating that IFNγ secretion was dependent on the interaction between the Fc domain of amivantamab and the FcR on immune cells (Fig. 5F). Induced inflammatory cytokines including IFNγ secreted from NK cells activated by amivantamab bound to EGFR and MET on EGFR Exon20Ins-driven tumors may lead to the recruitment and activation of adjacent immune cells to tumor cells in vivo. To explore this, we analyzed the infiltration of macrophages and NK cells into the tumor in a PDX model (YHIM-1029), which was generated from a patient-derived tumor harboring the D770_N771insG Exon20Ins mutation (Supplementary Table S2), and YU-1163–bearing BALB/c nude mice models treated with amivantamab at 10 and 30 mg/kg dose, respectively. mF4/80 and mNKp46, markers of macrophages and NK cells in BALB/c nude mice, respectively, were elevated in tumors following treatment with amivantamab, suggesting that the mechanistic components of ADCC observed in vitro may translate to recruitment of key effector cells in tumors in vivo.

Figure 4. Amivantamab reduces tumor burden in Ba/F3 cells and PDCs with EGFR Exon20Ins xenograft models. Antitumor effects of amivantamab in (A–C) Ba/F3 cells overexpressing D770delinsGY- or H773_V774insH-bearing NOG mice and (D–I) DFCI-127- or YU-1163–bearing NOG or BALB/c nude mice models relative to that of vehicle treatment. (A, B) Tumor volume was measured and the % change in tumor volume is depicted over the treatment period (D770delinsGY). (C, D) H773_V774insH xenograft models. Antitumor effects of amivantamab in (A–C) Ba/F3 cells and PDCs with EGFR Exon20Ins mutations cocultured with peripheral blood mononuclear cells (PBMCs) as effector cells (effector:target (E:T) = 50:1). (D, E) Treatment with amivantamab at 10 and 30 mg/kg dose, respectively. (G, H) Vehicle Amivantamab (30 mg/kg). (I) Western blot analysis showing pEGFR, pMET, MET, and GAPDH expression in D770delinsGY- or H773_V774insH xenograft models. (J) Cytotoxicity was measured by MTT assay in Ba/F3 cells overexpressing D770delinsGY- or H773_V774insH-bearing NOG mice and (D–I) DFCI-127- or YU-1163–bearing NOG or BALB/c nude mice models relative to that of vehicle treatment. (A, B) Tumor volume was measured and the % change in tumor volume is depicted over the treatment period (D770delinsGY). (C, D) H773_V774insH xenograft models. Antitumor effects of amivantamab in (A–C) Ba/F3 cells and PDCs with EGFR Exon20Ins mutations cocultured with peripheral blood mononuclear cells (PBMCs) as effector cells (effector:target (E:T) = 50:1). (D, E) Treatment with amivantamab at 10 and 30 mg/kg dose, respectively. (G, H) Vehicle Amivantamab (30 mg/kg). (I) Western blot analysis showing pEGFR, pMET, MET, and GAPDH expression in D770delinsGY- or H773_V774insH xenograft models. (J) Cytotoxicity was measured by MTT assay in Ba/F3 cells overexpressing D770delinsGY- or H773_V774insH-bearing NOG mice and (D–I) DFCI-127- or YU-1163–bearing NOG or BALB/c nude mice models relative to that of vehicle treatment.
In addition, these results suggest that amivantamab has greater ADCC and antitumor activity than cetuximab in the context of \( \text{EGFR} \) \text{Exon20ins} and that ADCC is an important mechanism in mediating the cytotoxic effects of amivantamab.

**Amivantamab Demonstrates Antitumor Activity in a PDX Model Harboring the D770_N771insG Exon20ins Mutation**

Treatment with amivantamab in the YHIM-1029 PDX model with D770_N771insG (Fig. 6A) resulted in a robust decrease in tumor volume, indicating that the antitumor activity observed in Ba/F3 and PDC models was preserved in a PDX model (Fig. 6B). In contrast, treatment with cetuximab (10 mg/kg) or poziotinib (1 mg/kg) only modestly reduced tumor volume. The dose of poziotinib was reduced to 1 mg/kg for this experiment due to the toxicity of poziotinib described above (Supplementary Fig. S4D–S4H). Pharmacodynamic analysis showed that amivantamab treatment resulted in EGFR and MET downmodulation, inhibition of the downstream signaling pathways pAKT, pERK, and pS6, and increased markers of apoptosis (Fig. 6C). In contrast, tumors from mice treated with cetuximab or poziotinib maintained the EGFR downstream signaling components pERK and pS6 (Fig. 6D), which was consistent with the modest effects observed on tumor growth. Histopathologic examination of tumor sections obtained following amivantamab or vehicle treatment using hematoxylin and eosin (H&E) staining, and IHC staining for EGFR, MET, and Ki-67, and terminal deoxynucleotidyl transferase–mediated dUTP nick end labeling (TUNEL) staining, further confirmed receptor inhibition and engagement of apoptotic machinery in \( \text{EGFR} \) \text{Exon20ins}–driven tumors in vivo (Fig. 6E). To verify whether the antitumor effect of amivantamab was affected by innate immunity in the in vivo models, we blocked the mouse CD16/CD32 via administration of anti-CD16/CD32 antibodies. The antitumor effect of amivantamab shown in Fig. 6B was abrogated when the amivantamab-treated PDX-bearing BALB/c nude mice were cotreated with anti-CD16/CD32 antibodies, indicating that the antitumor effects of amivantamab were partially mediated by immune cells in this condition (Supplementary Fig. S6).

**Antitumor Activity of Amivantamab in Patients with \( \text{EGFR} \) Exon20ins Disease**

In an ongoing first-in-human study of amivantamab in patients with advanced NSCLC (NCT02609776), promising clinical activity has been observed in patients with \( \text{EGFR} \) Exon20ins disease (13). A 58-year-old patient harboring the \( \text{EGFR} \) H773delinsNPY Exon20ins mutation achieved a partial response with a 65% tumor reduction (Fig. 7A), and a 48-year-old
Figure 5. Amivantamab has superior ADCC activity compared with cetuximab. A, Amivantamab-mediated ADCC activity against NSCLC PDCs expressing EGFR Exon20ins mutations using PBMC (E:T ratio = 50:1). ADCC assays were performed using DFCI-127 and YU-1163 PDCs as targets in the presence of IgG1 control, amivantamab, or cetuximab at various concentrations. PBMCs were cocultured for 4 hours with PDCs. *, P < 0.0001. Data are presented as averages ± SD of triplicate independent experiments. B, Amivantamab-mediated cytotoxicity against DFCI-127 and YU-1163 PDCs. PDCs were treated with IgG1, amivantamab (10 μg/mL), or cetuximab (10 μg/mL) for 24 hours in the presence or absence of PBMC (E:T ratio = 5:1). C, Quantitative analysis of the cells shown in the representative images (n = 3). *, P < 0.0001; **, P < 0.001. D, Pretreatment with FcR blocker with PBMC (E:T ratio = 50:1) reduced the amivantamab (10 μg/mL)-mediated ADCC effects. *, P < 0.0001. Data are presented as averages ± SD of triplicate independent experiments. E, IFNγ (pg/mL) levels in the cell culture media were detected by ELISA. The PDCs were cocultured with PBMCs in the presence of IgG1, amivantamab, or cetuximab at 1 μg/mL for 4 hours, and the culture medium was used for detection of IFNγ. *, P < 0.0001 versus cetuximab at the same concentration. F, PBMCs pretreated with FcR blocker reduced the IFNγ level in the culture medium in the presence of amivantamab (10 μg/mL). *, P < 0.0001; **, P < 0.001. Data are presented as averages ± SD of triplicate independent experiments. G, IHC staining for mF4/80 (macrophages) and mNKp46 (NK cells) of tumor sections in YHIM-1029 PDX-bearing BALB/c nude mice following 10 mg/kg IgG1 or 10 mg/kg amivantamab treatment.
Figure 6. Amivantamab reduces tumors in a PDX model with D770_N771insG EGFR mutation. A, Sanger sequencing data depicting the D770_N771insG mutations of the EGFR gene in a PDX model. B, Patient-derived tumors implanted in BALB/c nude mice were treated with vehicle, amivantamab (10 mg/kg), cetuximab (10 mg/kg), twice per week, intraperitoneal injections, or poziotinib (1 mg/kg), once a day. Data represent the mean ± SEM (n = 7/group). *, P < 0.0001. Western blot analysis for the downstream signaling pathways of EGFR, MET, and apoptosis markers in tumors obtained from YHIM-1029 PDX models treated with (C) 10 mg/kg amivantamab, (D) 10 mg/kg cetuximab, or 1 mg/kg poziotinib. E, Histopathologic examination of tumor sections obtained from the PDX models following 10 mg/kg amivantamab or vehicle treatment. H&E staining and IHC staining for EGFR, pEGFR, MET, pMET, Ki-67, and TUNEL.
**Figure 7.** Amivantamab reduces tumors in patients with NSCLC with EGFR Exon20ins mutations. Radiologic response following amivantamab 1,050 mg treatment in (A) a 58-year-old patient with the EGFR H773delinsNPY mutation and (B) a 48-year-old patient with the EGFR S768_D770dup mutation. C, A proposed model of diverse antitumor mechanisms of amivantamab in NSCLC with EGFR Exon20ins.

**CASE #1**

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<th>Pre</th>
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**CASE #2**

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**DISCUSSION**

In our study, we characterized the antitumor activity of amivantamab, a novel EGFR–MET bispecific antibody, in multiple preclinical models harboring *EGFR* Exon20ins mutations. In several Ba/F3 and PDC models expressing diverse *EGFR* Exon20ins mutations, amivantamab treatment resulted in EGFR and MET internalization, inhibition of downstream signaling cascades, engagement of apoptotic machinery, and subsequent inhibition of tumor cell proliferation. Importantly, these diverse action mechanisms of amivantamab were preserved *in vivo* as evidenced by pharmacodynamic analyses of tumors from cell line xenografts and PDX models treated with amivantamab (Fig. 7C). Furthermore, to the best of our knowledge, we first presented...
evidence of clinical activity of amivantamab in two case studies of patients with EGFR Exon20ins NSCLC from an ongoing phase I trial, highlighting the important translational nature of our preclinical work. Currently, there are no targeted therapies approved for EGFR Exon20ins–positive advanced NSCLC. Owing to its small size and flexibility, poziotinib, an oral pan-HER inhibitor, has demonstrated greater activity than approved EGFR TKIs in vitro and in PDX models of EGFR Exon20ins–mutant NSCLC (18). In a single-center phase II trial, poziotinib showed a 43% confirmed RR in heavily pretreated advanced EGFR Exon20ins–mutant NSCLC (21). However, in a subsequent pivotal phase II trial (NCT03318939), poziotinib yielded only 14.8% RR in a similar patient population (https://www.precisiononcologynews.com/drug-discovery-development/spectrums-poziotinib-failed-meet-primary-phase-ii-trial-endpoint#XjfkWgzaUk). TAK-788 has also shown preclinical activity against activating EGFR and HER2 mutations including EGFR Exon20ins. Although preliminary, in a phase I/II study (NCT02716116), TAK-788 produced clinical activity in a small subset of EGFR Exon20ins–mutant NSCLC (11). Importantly, treatment with poziotinib or TAK-788 was associated with a high incidence of EGFR WT–driven toxicity such as diarrhea and rashes, further limiting their clinical utility. Therefore, there is a substantial clinical need to identify new therapies for patients with EGFR Exon20ins.

In our study, amivantamab was clearly superior to poziotinib or cetuximab in terms of efficacy and tolerability in xenografts. Multiple facets of the MOA of amivantamab may contribute to the superior antitumor activity of amivantamab in the context of Exon20ins. In addition to the different MOA described in the various Exon20ins models described above, the ability of amivantamab to simultaneously bind two distinct epitopes of EGFR and MET may result in the concurrent interference with highly interconnected signaling pathways. To this end, amivantamab has been shown to have higher efficacy in decreasing tumor growth in the H1975-HGF model compared with the combination of anti-EGFR and anti-MET monovalent antibodies (36). This finding can be partly explained by an “avidity effect” whereby tumor cells expressing both targets, that is, EGFR and MET, bind to both arms of amivantamab with higher affinity than do cells that express only one target or engage a single Fab arm. A previous study has described correlations between binding affinity, receptor density, and receptor phosphorylation with amivantamab (37). Overall, bispecific antibodies show greater antitumor efficacy compared with a combination of monospecific mAbs via potential synergistic effects, and they increase selectivity by simultaneously targeting both receptors, favoring overexpressing cells as a consequence of avidity effects (38).

Amivantamab, produced by an engineered cell line defective for protein fucosylation, has a low-level core fucosylation. The human FcγRIIIα, critical for ADCC, binds antibodies with low core fucosylation with higher affinity and consequently mediates more potent and effective NK cell–mediated killing of cancer cells (12). In this study, amivantamab demonstrated more robust ADCC in EGFR Exon20ins mutation models than cetuximab, an EGFR-directed antibody that has not shown robust utility in NSCLC (39, 40). This ADCC activity was correlated with secreted IFNγ levels. It is possible that the IFNγ secreted from amivantamab mediated active immune cells, including NK cells and macrophages, may restimulate and recruit surrounding immune cells to the tumor, although additional studies are required to investigate this hypothesis. In addition, immunocytokines secreted from NK cells lead to upregulation of ICAM1 on target cells, rendering them more susceptible to target cell cytolyis (41). Indeed, pharmacodynamic analyses of tumors from mice treated with amivantamab revealed increased tumor levels of NK cells and macrophages. In contrast to many other therapeutic antibodies used in the clinical setting, amivantamab was designed and engineered with a low fucose backbone, which enhances its binding to FcγRIIa (17), which is present on NK cells, monocytes, and macrophages. The human FcγRIIIa, critical for ADCC, binds antibodies with low-level core fucosylation more tightly and consequently mediates more potent and effective ADCC killing of cancer cells (42). Thus, the enhanced binding of amivantamab to FcγRIIIa may lead to increased induction of Fc effector functions in comparison with other (normal fucose) hlgG1 antibodies such as cetuximab.

In this study, we used two different xenograft models, NOG mice and BALB/c nude mice for in vivo study. As demonstrated in many studies, NOG mice have impaired innate immunity and extremely low NK-cell activity, whereas BALB/c nude mice have intact innate immunity and active NK cells (43, 44). For this reason, more potent ADCC activity was expected in BALB/c nude mice than NOG mice. Therefore, a minimal amivantamab-mediated ADCC activity resulted in the modest efficacy with amivantamab in Ba/F3- and DFCI-127–bearing NOG mice (Fig. 4A and D; Supplementary Fig. S4A). On the other hand, the significant tumor regression was observed with amivantamab in YU-1163–bearing and YHIM-1029–bearing BALB/c nude mice (Figs. 4F and 6B), which resulted from multiple MOA of amivantamab to block the EGFR and MET downstream signaling pathway and elicit ADCC.

It has been observed that EGFR and tumor suppressor TP53 genes are commonly mutated in patients with NSCLC with independent prognostic implications. Furthermore, in patients with concomitant mutations in EGFR and TP53, there have been reports of decreased responsiveness to EGFR TKIs (45). A similar effect was observed in our study in the YU-1163 PDC and YUO-029 PDO following treatment with amivantamab. On the other hand, amivantamab exhibited a potent in vitro activity in YU-1163–bearing BALB/c nude mice, suggesting that ADCC activity of amivantamab, shown in Fig. 5A–G, was believed to be involved in the in vivo antitumor activity. These results suggest that the combination of effector cell–dependent and -independent MOAs elicited by amivantamab (Fig. 7C) may result in antitumor activity in tumors harboring a coalescence of intractable mutations, for example in EGFR Exon20ins disease, and concomitant deleterious mutations such as the TP53 mutation present in our preclinical model and reported in the broader patient population. In our study, amivantamab demonstrated less skin toxicity than poziotinib. Amivantamab-treated BALB/c nude mice appeared phenotypically normal with only minimal signs of keratosis on the face, indicating that amivantamab was well tolerated in this preclinical model. In contrast, treatment with poziotinib resulted in severe keratosis,
significant weight loss, and even sudden death (Supplementary Fig. S4D–S4H).

In conclusion, our data demonstrated that amivantamab functions through multiple MOA to elicit antitumor activity in multiple preclinical models of EGFR Exon20ins disease. Consequently, amivantamab warrants further clinical investigation, as evidenced by clinical results from 2 patients with EGFR Exon20ins NSCLC who have been treated with amivantamab in the clinical setting. This represents important progress toward the identification of an effective therapeutic option for patients with NSCLC with EGFR Exon20ins, an area of high unmet medical need.

**METHODS**

**Bo/F3 Cell Lines and Drug Compounds**

All mutant Ba/F3 cell lines were purchased from the German Collection of Microorganisms and Cell Cultures and were obtained from the Dana-Farber Cancer Institute, Harvard University (Boston, MA). All cells were maintained in RPMI1640 medium supplemented with 10% FBS and puromycin in a humidified incubator with 5% CO2. Amivantamab and IgG1 controls were provided by Janssen. Gefitinib, osimertinib, cetuximab, and pemetrexed were purchased from Cell Signaling Technology, Inc.; and GAPDH (PAB13195) was purchased from Santa Cruz Biotechnology, Inc.

**Antibodies**

Primary antibodies specific for pEGFR (2234), EGFR (4267), pMET (3077), MET (8198), pERK (4370), ERK (9107), pAKT (9271), AKT (9272), pS6 (4858), S6 (2217), p27 (2252), cleaved PARP (5625S), cleaved caspase-3 (9661), and BIM (2933) were purchased from Cell Signaling Technology; p121(28-181) and p53 (28-125) were purchased from Santa Cruz Biotechnology, Inc.; and GAPDH (PAB13195) was purchased from Abnova. For the IHC assay, mF4/80 (#70076) and mNKp46 (AF2225) were purchased from Cell Signaling Technology and R&D Systems, respectively.

**PDCs**

YU-1163 (S768,D770dup) cell lines were derived from malignant effusions from patients with NSCLC and cultured on collagen-coated plates in ACL-4 medium supplemented with 5% FBS. The cells maintained the driver oncogenes that were observed in the patients. Cells were enriched in an epithelial cell adhesion molecule (EPCAM)–positive cell population with a purity of over 95% before they were subjected to further assays. DFCI-58 (H773_V774insNPH) and DFCI-127 (P772_H773insPNP) cell lines were obtained from the Dana-Farber Cancer Institute and were cultured in ACL-4 medium and RPMI medium with 10% FBS, respectively. All patient samples were collected after written informed consent from the patients was obtained. The study protocols were approved by the respective institutional review boards.

**PDC Culture**

PDOs (YUO-029 and YUO-036) were established as described previously (46). Briefly, malignant effusions from 2 patients with NSCLC were collected and centrifuged, and the cell pellets were mixed with growth factor–reduced Matrigel (Corning) and seeded into 48-well plates. Solidified gels were overlaid with advanced DMEM/F12 (Invitrogen) containing 1× Glutamax (Invitrogen), 10 mmol/L HEPES (Invitrogen), 1× Antibiotic-Antimycotic (Invitrogen), 1× B-27 (Invitrogen), 20% R-spondin conditioned medium, 5 mmol/L Nicotinamide (Sigma), 1.25 mmol/L N-acetylcysteine (Sigma), 500 mmol/L SB202190 (Sigma), 500 mmol/L AB3-01 (Tocris), 100 ng/mL Mouse Noggin (PeproTech), 100 ng/mL human FGF10 (PeproTech), 25 ng/mL human FGF7 (PeproTech), 50 μg/mL Primocin (InvivoGen), and 0.523 μmol/L Y-27632 (Enzo). R-spondin conditioned medium was produced from HA-R-Spondin-1-Fc 293T Cells (Ambio). For passaging, organoids were collected, mechanically sheared with a 25-gauge needle, and washed with cold PBS before the organoid pellets were resuspended in the Matrigel and seeded into 24-well plates at ratios of 1:2 to 1:4. The culture medium was replenished at least twice a week. Cell viability tests were performed as described previously (47).

Briefly, organoids were trypsinized into single cells and cultured for 5 to 10 days. Then, the organoids were collected, resuspended in the medium containing 5% Matrigel, and plated in a 96-well plate (Corning) at a concentration of 2,000 organoids/μL. The medium with the IgG1 control or amivantamab at diverse concentrations was added and incubated for 72 hours. Cell viability was measured using CellTiter-Glo 3D Culture Reagent (Promega) on a microplate luminometer according to the manufacturer’s instructions.

**PDX Models**

PDXs were created using 5- to 8-week-old female SCID (NOG) and nude (nu/nu) mice obtained from Orient Bio. All methods complied with the guidelines of our Institutional Animal Care and Use Committee (Yonsei University College of Medicine, Seoul, Republic of South Korea) and were approved by the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC). After removal of the necrotic and supporting tissues from core biopsy specimens, small specimens of the tumor tissue (3 mm × 3 mm × 3 mm) from each patient were implanted subcutaneously in 1 to 2 mice. After the tumor reached 1.5 cm in diameter, it was excised, dissected into small specimens (3 mm × 3 mm × 3 mm), and reimplanted into nude mice.

**In Vivo Xenograft Studies**

Female athymic BALB-c/nu mice were obtained from Orient Bio at 5 to 6 weeks of age. All mice were handled in accordance with the Animal Research Committee’s Guidelines at Yonsei University College of Medicine, and all facilities were approved by AAALAC. Ba/F3 cells and PDCs (1 × 10⁶ cells) were injected subcutaneously into the NOG and BALB-c/nu mice, respectively, and growth was measured twice weekly; after establishment of palpable lesions, mice were assigned to testing. Once the tumor volume reached approximately 150 to 200 mm³, mice were randomly allocated into groups of 5 animals to receive either vehicle, IgG1 control, or amivantamab. The tumor size was measured every 2 days using calipers. The average tumor volume in each group was expressed in mm³ and calculated according to the equation for a prolate spheroid: tumor volume = 0.523 × (large diameter) × (small diameter)².

**Antiproliferation Assay**

Ba/F3 cells or PDCs expressing EGFR Exon20ins mutations were seeded onto 96-well plates in 100 μL. After treatment with IgG1 control, amivantamab, gefitinib, or osimertinib for 72 hours, cell viability was measured by quantifying the total amount of ATP using the CellTiter-Glo 2.0 Assay Kit (Promega) according to the manufacturer’s instructions.

**Colony Formation Assay**

Cells were seeded onto 6-well culture plates and incubated for 12 days at 37°C with amivantamab (0, 0.1, or 1 mg/mL). Cells were washed with PBS, fixed, and stained with 4% paraformaldehyde in 5% crystal violet for 10 minutes. Colonies were eluted with 1% SDS, and the optical density value was determined using ELISA at 470 nm.

**ADCC Assays**

The ADCC assay was conducted using the Lactate Dehydrogenase (LDH) Cytotoxicity Detection Kit (Roche) in accordance with the manufacturer's instructions. Human PBMCs obtained from healthy donors were used to generate the ADCC assay. The assay was performed according to the manufacturer's instructions.
volunteers were used as the effector cells. ADCC was conducted using an E:T cell ratio ranging from 50:1 to 5:1 and incubated for 4 to 24 hours at 37°C in 5% CO₂. Amivantamab concentrations of 100 to 0.01 μg/mL were tested. The LDH activity of the cell culture supernatants was measured, and the percentage cytotoxicity was calculated as described in the manufacturer’s protocol.

**IF Analysis**

PDCs were seeded on 0.01% poly-L-lysine (Sigma-Aldrich) coated coverslips. The following day, cells were treated with IgG1 control or amivantamab at 0.1 mg/mL. After 72 hours, the coverslips were fixed in 4% formaldehyde for 15 minutes, permeabilized with 0.3% Triton X-100 for 5 minutes, and incubated with primary antibody for 1 hour at room temperature. The primary antibodies used in the study were rabbit monoclonal anti-EGFR and anti-MET (Santa Cruz Biotechnology) and ab992 (Millipore) at a dilution of 1:100. The coverslips were rinsed twice with PBS, followed by incubation with the appropriate fluorescein-conjugated secondary antibody (Invitrogen) for 1 hour at room temperature. The cells were counterstained with 4',6-diamidino-2-phenylindole (DAPI; 300 nmol/L; Invitrogen), and the coverslips were mounted on slides using Faramount Aqueous Mounting Medium (Dako).

**IHC**

IHC was performed using the Automated Staining System (BOND Rx, Leica Biosystems). Briefly, 4-mm paraffin-embedded tumor sections were deparaffinized and rehydrated. Slides then underwent heat-induced epitope retrieval with citrate buffer at 100°C for 20 minutes. Antibodies were used at 1:100 dilution and hematoxylin followed by the Dunnett test or Student test. Dose–response curves were prepared using GraphPad Prism (Ver. 5, GraphPad Software Inc.).

**Statistical Analysis**

Data were collected from three independent experiments and either presented descriptively or analyzed by one-way ANOVA, followed by the Dunnet test or Student t test. Dose–response curves were prepared using GraphPad Prism (Ver. 5, GraphPad Software Inc.).

**Disclosure of Potential Conflicts of Interest**

J.C. Curtin is an associate director, oncology translational research at Janssen Research and Development. R.E. Knoblauch is an executive medical director at Janssen. M.V. Lorenzi is vice president, oncology at Janssen, Pharmaceutical Companies of Johnson and Johnson and has ownership interest (including patents) in Johnson & Johnson. B.C. Cho is a professor at Novartis, AstraZeneca, Takoda, Janssen, Medpacto, Blueprint Medicines, MSD, Boehringer-Ingelheim, Roche, BMS, Ono, Yuhan, Pfizer, Eli Lilly, and Janssen, reports receiving commercial research grants from Novartis, Bayer, MSD, AbbVie, Medpacto, GilInnovation, Eli Lilly, Blueprint Medicines, AstraZeneca, MOGAM Institute, Dong-A ST, Champions Oncology, Janssen, Yuhan, Ono, and Dizal Pharma, reports receiving speakers bureau honoraria from Novartis, Bayer, MSD, AstraZeneca, MOGAM Institute, Dong-A ST, Champions Oncology, Janssen, Yuhan, Ono, and Dizal Pharma, has ownership interest (including patents) in TheraCanVac Inc, Gencurix Inc, Bridgemo Therapeutics, and KANAPH Therapeutic Inc, and has received other remuneration from Daan Biotherapeutics. No potential conflicts of interest were disclosed by the other authors.

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Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): J. Yun, J.-H. Kim, K.-H. Pyo, S.G. Heo, S. Lim, J.C. Curtin, R.E. Knoblauch, B.C. Cho

Writing, review, and/or revision of the manuscript: J. Yun, S.-H. Lee, M.R. Yun, M. Thayu, J.C. Curtin, R.E. Knoblauch, M.V. Lorenzi, A. Rosah, B.C. Cho

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Antitumor Activity of Amivantamab (JNJ-61186372), an EGFR–MET Bispecific Antibody, in Diverse Models of EGFR Exon 20 Insertion–Driven NSCLC

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