American Society of Hematology
2021 L Street NW, Suite 900,
Washington, DC 20036
Phone: 202-776-0544 | Fax 202-776-0545
editorial@hematology.org

Birtamimab plus standard of care in light chain amyloidosis: the phase 3 randomized placebo-controlled VITAL trial

Tracking no: BLD-2022-019406R3

Morie Gertz (Division of Hematology, Department of Internal Medicine, Mayo Clinic, United States) Adam Cohen (University of Pennsylvania, United States) Raymond Comenzo (Tufts Medical Center, United States) Efstathios Kastritis (Department of Clinical Therapeutics, National and Kapodistrian University of Athens, School of Medicine, Greece) Heather Landau (Memorial Sloan-Kettering Cancer Center, United States) Edward Libby (Fred Hutchinson Cancer Center, United States) Michaela Liedtke (Stanford University, United States) Vaishali Sanchorawala (Boston University Chobanian & Avedisian School of Medicine and Boston Medical Center, United States) Stefan Schönland (University of Heidelberg, Germany) Ashutosh Wechalekar (University College London, United Kingdom) Jeffrey Zonder (Karmanos Cancer Institute, United States) Giovanni Palladini (University of Pavia, Italy) Jackie Walling (Prothena Biosciences Inc., United States) Spencer Guthrie (Prothena Biosciences, United States) Christie Nie (Prothena Biosciences Inc., United States) Carol Karp (Prothena Biosciences Inc., United States) Yuying Jin (Prothena Biosciences Inc., United States) Gene Kinney (Prothena Biosciences Inc., United States) Giampaolo Merlini (Amyloidosis Research and Treatment Center, Fondazione IRCCS, Policlinico San Matteo, Italy)

Abstract:
Amyloid light chain (AL) amyloidosis is a rare, typically fatal disease characterized by accumulation of misfolded immunoglobulin light chains (LCs). Birtamimab is an investigational humanized monoclonal antibody designed to neutralize toxic LC aggregates and deplete insoluble organ-deposited amyloid via macrophage-induced phagocytosis. VITAL was a phase 3 randomized, double-blind, placebo-controlled clinical trial assessing the efficacy and safety of birtamimab + standard of care (SOC) in 260 newly diagnosed, treatment-naïve patients with AL amyloidosis. Patients received 24 mg/kg intravenous birtamimab + SOC or placebo + SOC every 28 days. The primary composite endpoint was time to all-cause mortality (ACM) or centrally adjudicated cardiac hospitalization {greater than or equal to}91 days after first study drug infusion. The trial was terminated early after an interim futility analysis; there was no significant difference in the primary composite endpoint (hazard ratio [HR] = 0.826; 95% confidence interval [CI] 0.574-1.189; log-rank *P* = .303). A post hoc analysis in Mayo Stage IV patients, those at highest risk of early mortality, showed significant improvement in time to ACM with birtamimab at month 9 (HR = 0.413; 95% CI: 0.191-0.895; log-rank *P* = .021). At month 9, 74% of Mayo Stage IV patients treated with birtamimab and 49% of those given placebo survived. Overall, the rates of treatment-emergent adverse events (TEAEs) and serious TEAEs were generally similar between treatment arms. A confirmatory phase 3 randomized, double-blind, placebo-controlled clinical trial of birtamimab in patients with Mayo Stage IV AL amyloidosis (AFFIRM-AL; NCT04973137) is currently enrolling. The VITAL trial was registered at www.clinicaltrials.gov as #NCT02312206.

Conflict of interest: COI declared - see note

COI notes: M.A.G.: personal fees from Alnylam, Aptitude Health, Ashfield, Celgene, Ionis/Akcea, Janssen, Johnson & Johnson, Juno, Physicians Education Resource, Prothena, Research to Practice, Sanofi, Sorrento; personal fees for Data Safety Monitoring board from Abbvie; payment for development of educational materials for i3Health and educational program development for i3Health; grant funding from NCI SPORE MM SPORE 5P50 CA186781-04. A.D.C.: Consultant for Abbvie, Arcellx, AstraZeneca, BMS, Celgene, Genentech/Roche, GlaxoSmithKline, Ichnos, Janssen, Kite Pharma, Oncopeptides, Pfizer, Seattle Genetics, Takeda; research funding from GlaxoSmithKline and Novartis; patents and royalties from Novartis. R.L.C.: Consultant and advisor for Caelum; consultant and advisor for, and research funding from Janssen, Prothena; research funding from Karyopharm, Takeda; advisor and research funding for Unum; royalties for Patent 9593332, Pending 20170008966. E.K.: Consultant for Amgen, Janssen, Genesis, Takeda, Prothena, Pfizer; received research funding from Amgen, Janssen. H.J.L.: Advisor for Caelum, Janssen, Prothena, Pfizer; consultant and advisor for Celgene; consultant for Karyopharm; advisor and research funding from Takeda; research funding from Amgen. E.N.L.: Consultant for Abbvie, Akcea, Alnylam, Pharmacyclics, Janssen. M.L.: Research funding from Alexion, Caelum, Prothena; advisor for Janssen, Takeda. V.S.: Consultant and advisor for and research funding from Caleum; consultant for Attralus, Pfizer; research funding from Celgene, Karyopharm, Millennium-Takeda, Oncopeptides, Prothena, Sorrento; research funding from and advisor for Janssen; advisor for Abbvie, Proclara, Protego, Pharmatrace, Prothena, Regeneron, Telix. S.S.: Advisor for, travel grant, honoraria, and research funding from Janssen, Prothena; research funding from Sanofi; honoraria from Pfizer, Takeda; advisor for Telix; travel grant from Binding Site, Celgene, Jazz. A.W.: Research funding from Amgen; honoraria from Alexion, Celgene, GlaxoSmithKline, Janssen-Cilag, Prothena, Takeda. J.A.Z.: Consultant/advisor for Alnylam, Amgen, BMS, Caelum, Celgene, Intellia, Janssen, Oncopeptides, Takeda; research funding from BMS, Celgene. GP: advisor for and honoraria from Janssen, Protego, Zentalis; honoraria from Pfizer, Sebia, Siemens, The Binding Site. J.W.: employed by Consulting JW LLC; consultant for 4DMT, Aduro/Chinook, Alpha Holdings, Ambagon, Aminex, Arch Oncology, AroBio, Benevolent/BAI, CALIBR - Scripps, Cocept, Crinetics Pharmaceuticals, Crown BioScience, Cumulus Oncology Limited, Cybrexa, CytomX, Dren Bio, Entos, Excure Inc., Flag, Fulgent, Harpoon, Immune Onc, ImmuNext, InClin, January Therapeutics, Janux Therapeutics, Myovant, Nurix, Nuvation Bio, Inc. OPNA Bio LLC., Orbus, Orphagen, Oryzon, Plexxikon, Propella, Que Oncology, Sagamore, Sesen Bio, Shape Therapeutics, Sonnet Bio Therapeutics, Trex, venBio Partners LLC; payment for expert testimony from Puma Biotechnology. S.G.: founder of and employed by Attralus, Inc.; owns stock or stock options in Prothena and Attralus; named inventor on patents related to birtamimab and Attralus. C.N., C.K., Y.J.: Employment and stockholder at Prothena. G.G.K.: Employment and equity ownership in Prothena; named inventor on multiple patents and patent applications related to birtamimab. G.M.: consultant for GlaxoSmithKline, Novo Nordisk; travel grant from Janssen. The current affiliation for J.W. is Consulting JW LLC, San Francisco, CA, USA. The current affiliation for S.G. is Attralus Inc., San Francisco, CA, USA.

Preprint server: No;

Author contributions and disclosures: M.G, J.W., and G.K. designed the study; M.G., J.W., S.G. and G.K. were responsible for the conduct of the study; C.N. was responsible for data collection; and Y.J. performed analysis of data. All authors had full access to study data and take responsibility for the integrity of the data and the accuracy of the data analysis. C.N., C.K., Y.J., G.K. and M.G. assisted in preparation of the manuscript. All authors revised the manuscript and reviewed and approved the final version for submission.

Non-author contributions and disclosures: Yes; Medical writing and editorial support were provided by Emily Mercadante PhD of AMICULUM Ltd., funded by Prothena Biosciences, Ltd. The authors would like to thank Peloton Advantage, LLC, an OPEN Health company, for their contributions toward initial drafting of the manuscript. We thank Martin Koller for his contributions to the VITAL Study design and data acquisition. This clinical trial was sponsored by Prothena Biosciences, Ltd, Dublin, Ireland, a member of the Prothena Corporation plc group. Prothena Biosciences, Ltd participated in the design and conduct of the study; collection, management, analysis, and interpretation of the data; and preparation, review, and approval of the manuscript.

Agreement to Share Publication-Related Data and Data Sharing Statement: On request, and subject to certain criteria, conditions, exceptions, and applicable data privacy laws, Prothena will provide access to individual deidentified participant data from Prothena-sponsored global interventional clinical studies conducted for medicines for indications that have been approved. Please contact medicalinfo@prothena.com for inquiries

Clinical trial registration information (if any): Study ID number NCT02312206

Birtamimab plus standard of care in light chain amyloidosis: the phase 3 randomized placebo-controlled VITAL trial

Morie A. Gertz,[1] Adam D. Cohen,[2] Raymond L. Comenzo,[3] Efstathios Kastritis,[4] Heather J. Landau,[5] Edward N. Libby,[6,7] Michaela Liedtke,[8] Vaishali Sanchorawala,[9] Stefan Schönland,[10] Ashutosh Wechalekar,[11] Jeffrey A. Zonder,[12] Giovanni Palladini,[13,14] Jackie Walling,[15] Spencer Guthrie,[15] Christie Nie,[15] Carol Karp,[15] Yuying Jin,[15] Gene G. Kinney,[15] and Giampaolo Merlini,[13,14] on behalf of the VITAL Study Investigators

[1]Division of Hematology, Department of Internal Medicine, Mayo Clinic, Rochester, MN; [2]Abramson Cancer Center, The Hospital of the University of Pennsylvania, Philadelphia, PA; [3]Divison of Hematology and Oncology, Tufts Medical Center, Boston, MA; [4]Department of Clinical Therapeutics, School of Medicine, National and Kapodistrian University of Athens, Athens, Greece; [5]Adult Bone Marrow Transplantation Service, Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, NY; [6]Clinical Research Division, Fred Hutchinson Cancer Center, Seattle, WA; [7]Division of Medical Oncology, Department of Medicine, University of Washington, Seattle, WA; [8]Stanford Cancer Institute, Stanford, CA; [9] Amyloidosis Center, Boston University School of Medicine, Boston, MA; [10]Medical Department V, Amyloidosis Center, Universitätsklinikum Heidelberg, Heidelberg, Germany; [11]National Amyloidosis Centre, Division of Medicine, University College of London, Royal Free Hospital, London, UK; [12]Department of Oncology, Barbara Ann Karmanos Cancer Institute, Detroit, MI; [13]Department of Molecular Medicine, University of Pavia, Pavia, Italy; [14]Amyloidosis Research and Treatment Center, Fondazione IRCCS, Policlinico San Matteo, Pavia, Italy; [15]Prothena Biosciences Inc., South San Francisco, CA

Corresponding author:
Morie A. Gertz
Mayo Clinic
200 First Street SW
Rochester, MN 55905
Phone: 507-266-5081
Fax: 507-266-4972
E-mail: gertm@mayo.edu

Short Title: VITAL Study: Birtamimab in cardiac AL amyloidosis

Blood

Text word count: 4414/4000; Abstract: 250/250; Figures: 1; Tables: 4; References: 30; plus supplement

KEY POINTS

- The VITAL study of birtamimab in all stages of newly diagnosed AL amyloidosis was discontinued early per futility analysis
- Birtamimab improved post hoc all-cause mortality in Mayo Stage IV patients with cardiac involvement, who are at high risk of early death

Abstract

Amyloid light chain (AL) amyloidosis is a rare, typically fatal disease characterized by accumulation of misfolded immunoglobulin light chains (LCs). Birtamimab is an investigational humanized monoclonal antibody designed to neutralize toxic LC aggregates and deplete insoluble organ-deposited amyloid via macrophage-induced phagocytosis. VITAL was a phase 3 randomized, double-blind, placebo-controlled clinical trial assessing the efficacy and safety of birtamimab + standard of care (SOC) in 260 newly diagnosed, treatment-naïve patients with AL amyloidosis. Patients received 24 mg/kg intravenous birtamimab + SOC or placebo + SOC every 28 days. The primary composite endpoint was time to all-cause mortality (ACM) or centrally adjudicated cardiac hospitalization ≥91 days after first study drug infusion. The trial was terminated early after an interim futility analysis; there was no significant difference in the primary composite endpoint (hazard ratio [HR] = 0.826; 95% confidence interval [CI] 0.574-1.189; log-rank *P* = .303). A post hoc analysis in Mayo Stage IV patients, those at highest risk of early mortality, showed significant improvement in time to ACM with birtamimab at month 9 (HR = 0.413; 95% CI: 0.191-0.895; log-rank *P* = .021). At month 9, 74% of Mayo Stage IV patients treated with birtamimab and 49% of those given placebo survived. Overall, the rates of treatment-emergent adverse events (TEAEs) and serious TEAEs were generally similar between treatment arms. A confirmatory phase 3 randomized, double-blind, placebo-controlled clinical trial of birtamimab in patients with Mayo Stage IV AL amyloidosis (AFFIRM-AL; NCT04973137) is currently enrolling.

The VITAL trial was registered at www.clinicaltrials.gov as #NCT02312206.

Introduction

Amyloid light chain (AL) amyloidosis is the most common form of systemic amyloidosis, with an estimated incidence of 8-14.4 cases per million person-years. [1-4] This rare, typically fatal disease is caused by misfolded kappa (κ) or lambda (λ) immunoglobulin light chains (LCs) from an underlying plasma cell dyscrasia. [5,6] Misfolded LC proteins form toxic aggregates and amyloid fibrils that deposit in vital organs leading to dysfunction,[7] most commonly in the heart (80%) and kidneys (66%). [8] Cardiac impairment and multi-organ damage are key predictors of reduced survival in AL amyloidosis. [1,2,9-11] The mortality risk for newly diagnosed, treatment-naïve patients can be categorized using the revised 2012 Mayo Clinic Staging System. [12] Mayo Stages range from I to IV, with Stage IV patients having the highest risk for early mortality (median survival from diagnosis: 5.8 months; 5-year survival rate: 14%).[12]

Current treatment options for patients with AL amyloidosis target plasma cells in an effort to minimize the production of new LCs. [6,13-15] Subcutaneous daratumumab in combination with bortezomib, cyclophosphamide, and dexamethasone (CyBorD), is the only US Food and Drug Administration (FDA)-approved therapy for patients with newly diagnosed AL amyloidosis. [13,15] However, daratumumab is not approved for the treatment of patients with advanced cardiac AL amyloidosis outside of clinical trials. [13,15] While existing antiplasma cell therapies may provide hematologic response and partial biomarker-based organ response these agents have not demonstrated a survival benefit. [6,16-19] There remains a significant unmet need for therapies that can improve survival in patients with advanced AL amyloidosis, who are at high risk for early death. [12,20]

Birtamimab (formerly NEOD001), an investigational humanized IgG1 monoclonal antibody that binds directly to a conserved epitope in misfolded κ and λ LCs, was designed to neutralize toxic soluble LC aggregates and deplete insoluble organ-deposited amyloid via macrophage-induced phagocytosis.[21,22] Birtamimab was granted orphan drug status by the US FDA and the European Medicines Agency and received FDA Fast Track Designation.[23,24] A phase 1/2 clinical trial in AL amyloidosis patients with persistent organ dysfunction demonstrated that birtamimab is generally safe and well tolerated.[25]

The phase 3 VITAL clinical trial evaluated the efficacy and safety of birtamimab + standard of care (SOC) versus placebo + SOC in newly diagnosed, treatment-naïve patients with AL amyloidosis and cardiac involvement (including N-terminal pro-brain natriuretic peptide [NT-proBNP] ≥650 and ≤8500 pg/mL) by assessing time to all-cause mortality (ACM) or cardiac hospitalization (CH). When a futility analysis of VITAL was conducted, the independent data monitoring committee recommended discontinuation of the clinical trial, prompting early termination. A numerical trend favoring birtamimab in the primary composite endpoint for the overall population was observed, and was hypothesized to be driven by a treatment effect in the most advanced patients (Mayo Stage IV). Thus, further post hoc analyses were performed in Mayo Stage IV patients. Here, we present results of the phase 3 VITAL clinical trial, including post hoc analyses conducted in Mayo Stage IV patients. Data from Mayo Stage IV patients served as the basis for the ongoing confirmatory phase 3 AFFIRM-AL study being conducted under a special protocol assessment (SPA) agreement with the US FDA.

Methods

Study design and patients

VITAL was a phase 3, multicenter, global, double-blind, placebo-controlled clinical trial (NCT02312206) conducted between 2016– and 2018 in newly diagnosed, treatment-naïve patients with AL amyloidosis and cardiac involvement. The clinical trial was approved by the institutional review boards or ethics committees of all participating sites and was conducted in compliance with the Good Clinical Practice guidelines of the International Council for Harmonisation and the principles of the Declaration of Helsinki. Written informed consent to participate in the clinical trial was obtained from all patients.

Adults aged ≥18 years with a biopsy proven diagnosis of AL amyloidosis were enrolled. Eligibility was determined using either polarizing light microscopy of green birefringent material in Congo red-stained tissue specimens or by characteristic appearance on electron microscopy, and confirmation of AL amyloidosis by immunohistochemistry or mass spectroscopy. Additional eligibility criteria included cardiac involvement, defined by the following: (1) past or present clinical signs and symptoms supportive of a diagnosis of heart failure in the absence of an explanation for heart failure other than AL amyloidosis; (2) either an endomyocardial biopsy demonstrating AL amyloidosis or an echocardiogram demonstrating a mean left ventricular wall thickness at diastole >12 mm in the absence of other causes that would adequately explain the degree of wall thickening; (3) NT-proBNP ≥650 and ≤8500 pg/mL; and (4) estimated glomerular filtration rate (eGFR) ≥30 mL/min/1.73 m^2 as estimated by the Chronic Kidney Disease Epidemiology Collaboration equation. Key exclusion criteria were non-AL amyloidosis, meeting the diagnostic

criteria for multiple myeloma as per the International Myeloma Working Group; eligibility for and plans to undergo autologous stem cell transplant; or prior treatment with plasma cell-directed chemotherapy. The full VITAL protocol is available online at https://clinicaltrials.gov (NCT02312206).

At the last screening visit (Day −2 or Day −1 prior to randomization), the severity of AL amyloidosis was determined using the 2012 Mayo Clinic revised staging criteria, [12] and level of renal dysfunction was determined using renal staging criteria[26] (**Tables S1 and S2**). Blood samples were taken for the assessment of hematology and chemistry parameters by central laboratory, including troponin-T and NT-proBNP. Other assessments included serum free light chains (FLC), urinalysis, 6-minute walk test (6MWT), and completion of the Short Form-36 questionnaire, version 2 (SF-36v2).

Randomization and interventions

Patients were stratified by Mayo Stage (I/II vs III/IV), Renal Stage (I vs II/III), and 6WMT distance (<300 meters vs ≥300 meters) and were randomized 1:1 to receive 24 mg/kg (up to a maximum dose of 2500 mg) intravenous birtamimab + SOC or intravenous placebo + SOC every 28 days. All patients received concomitant SOC chemotherapy, consisting of a first-line bortezomib-containing regimen, administered subcutaneously on a weekly basis, with subsequent plasma-cell directed therapies prescribed as per SOC at the investigator's discretion. Antiviral prophylaxis was required in patients receiving SOC chemotherapy. Patients who discontinued study drug were to be followed until the last adjudicated event.

Endpoints

The primary composite endpoint was time to ACM or CH as centrally adjudicated by the Clinical Events Committee (CEC). For ACM, all deaths occurring after the first

infusion of study drug through the clinical trial's last subject last visit, and for CH, all events occurring ≥91 days after first study drug infusion, were included.

Key secondary endpoints were the change from baseline to month 9 in the SF36v2 Physical Component Summary (PCS) score, 6MWT distance, and cardiac best response, measured by NT-proBNP (see Supplementary Methods). Safety evaluations included frequency and severity/seriousness of adverse events (AEs). Post hoc analyses in Mayo Stage IV patients were time to ACM at month 9 and change from baseline to month 9 in SF-36v2 PCS, 6MWT, and cardiac best response. Hematologic responses better than or equal to a very good partial response (VGPR) by month 3 were assessed in Mayo Stage IV patients, defined as reduction in difference between involved and uninvolved serum free light chains (dFLC) to <4.0mg/dL for patients with baseline dFLC >5mg/dL. For the post hoc analyses of ACM in Mayo Stage IV patients, all adjudicated deaths occurring after the first infusion of study drug up to month 9 were included. Deaths were censored at month 9, given the observed median survival of 8.3 months in the Mayo Stage IV placebo group in VITAL, and to align with key secondary endpoints.

Statistical analysis

Efficacy results were analyzed in the intention-to-treat population, which included all randomized patients who received any amount of study drug and was equivalent to the safety analysis population. For the primary composite endpoint of time to ACM or CH, the assumed 18-month event rate in the placebo arm was 60%, based on Kumar et al[12] and was assumed to be 42% in the birtamimab arm, corresponding to a hazard ratio (HR) of 0.594. For a two-arm clinical trial with 1:1 randomization and based on the use of a two-sided test at the alpha = 0.05 level of significance, a total

of 156 events (both arms combined) were required for 90% power. The distribution of the primary endpoint in the two treatment groups was summarized using the Kaplan-Meier method. The treatment groups were compared using a two-sided stratified (by the randomization stratification factors) log-rank test at the alpha = 0.05 level of significance. Each component of the primary endpoint was also analyzed. The SF-36v2 PCS score change from baseline at month 9 was analyzed as pre-specified, using a restricted maximum likelihood based mixed-effect model for repeated measures (MMRM) model including fixed effects for randomization strata, treatment group, categorical time point, and the treatment group × time point interaction, with the baseline value included as a covariate. The 6MWT distance (meters) change from baseline at month 9 was analyzed as pre-specified using a rank analysis of covariance (ANCOVA) model including fixed effects for randomization strata and treatment group, with the ranked baseline value included as a covariate, to address missing data. See Supplementary Methods for ranking of 6MWT distance values. The 6MWT distance (meters) change from baseline at month 9 was analyzed using the same MMRM model applied to the SF-36v2 PCS score, to estimate the change from baseline.

For the post hoc analyses in Mayo Stage IV patients, the same methods were applied as above but only included stratification factors of Renal Stage (I vs II/III) and 6MWT distance (<300 meters vs ≥300 meters). Sensitivity analyses of ACM in Mayo Stage IV patients were also performed, adjusting for key baseline variables. HRs and 90% two-sided confidence intervals (CIs) were estimated from the semiparametric Cox Regression model stratified by randomization strata (ie, Renal Stage I vs II/III, and 6MWT distance), and with baseline variables including age, sex, race, ethnicity, age at diagnosis, duration since diagnosis, NT-proBNP, dFLC, FLC, New York Heart

Association (NYHA) class, troponin-T, and 6MWT distance added separately. All baseline variables except categorical variables (ie, sex, race, ethnicity, NYHA class) were adjusted as continuous variables. An effect modification analysis comparing HRs of ACM at month 9 was performed to determine whether the observed post hoc treatment effect in Mayo Stage IV patients was due to chance. The Cox Regression model included treatment (birtamimab vs placebo), Mayo Stage (I-III vs IV), and the interaction between treatment and Mayo Stage, with stratification factors of Renal Stage and 6MWT distance. Effect modification of Mayo Stage (I-III vs IV) was considered present if the interaction term had a statistically significant *P*-value ($P \leq .05$). Number and percentage, along with two-sided 95% CIs of patients in each category of hematologic ≥VGPR are presented by treatment group. A Cochran–Mantel–Haenszel test stratified by the randomization stratification factors was used to compare hematologic ≥VGPR rate at month 3.

Treatment-emergent AEs (TEAEs) were summarized. The incidence of TEAEs was tabulated by system organ class and preferred term for each treatment group, and by severity/seriousness and relationship to treatment. TEAEs leading to death or study drug discontinuation, with grade ≥3 severity, and serious TEAEs were summarized and listed. TEAEs occurring at any dose that resulted in any of the following outcomes were considered serious: death; life-threatening TEAE; inpatient hospitalization, or prolongation of existing hospitalization; persistent or significant disability/incapacity; congenital anomaly/birth defect; or important medical events. Severity of TEAEs was assessed using the National Cancer Institute-Common Terminology Criteria for Adverse Events (NCI-CTCAE) version 4.0.

Data sharing statement

On request, and subject to certain criteria, conditions, exceptions, and applicable data privacy laws, Prothena will provide access to individual deidentified participant data from Prothena-sponsored global interventional clinical studies conducted for medicines for indications that have been approved. Please contact medicalinfo@prothena.com for inquiries.

Results

Patient disposition and baseline characteristics

A total of 260 patients were randomized, 130 to birtamimab and 130 to placebo, from 70 study sites over approximately 2 years. Randomized patients received at least one infusion of study drug and were included in the efficacy and safety analyses. The primary reason for study discontinuation was study termination by the sponsor (148 patients [57%]) (**Figure S1**).

The baseline demographic and clinical characteristics of all patients in the VITAL clinical trial are summarized in **Table 1** by treatment group. Patient demographics and baseline disease characteristics were generally well-balanced between the birtamimab and placebo groups: median (quartile [Q] 1, Q3) age at AL amyloidosis diagnosis was 64.1 (57.5, 70.9) and 62.4 (56.8, 69.3) years, time since disease diagnosis was 1.31 (0.92, 1.87) and 1.48 (0.95, 2.17) months, and baseline NT-proBNP was 3146.2 (1650.0, 5173.0) and 3183.7 (1910.0, 5551.0) pg/mL, respectively. Approximately 30% of patients enrolled had Mayo Stage IV AL amyloidosis (77/260); patient demographics and baseline disease characteristics in

this subset of patients were generally balanced between birtamimab (n=38) and placebo (n=39) arms (**Table 1**).

All patients received concomitant bortezomib-containing chemotherapy regimens; 87.7% of patients received first-line CyBorD. SOC regimens given in the second-line setting varied, but most commonly consisted of lenalidomide-containing regimens (12.7%). Overall, patients in the birtamimab and placebo arms received a similar median (range) number of study drug infusions, 15.5 (1-35) and 14.0 (1-35) infusions, respectively. The mean (standard deviation) duration of exposure was 389.4 (245.7) days for patients treated with birtamimab and 352.7 (248.3) days for patients given placebo. In the overall population, median (Q1, Q3) follow-up was 15.7 (12.0, 22.1) months for birtamimab and 14.5 (10.3, 19.8) months for placebo; in Mayo Stage IV patients, median follow-up was 15.2 (9.4, 21.4) and 11.3 (2.3, 17.7) months, respectively.

Efficacy

In the overall population, analysis of the primary composite endpoint of time to ACM or CH favored birtamimab, but the difference between birtamimab + SOC and placebo + SOC was not statistically significant ([HR = 0.826; 95% CI 0.574-1.189; log-rank *P* = .303]; **Figure S2**). Table S3 shows results for individual components of the primary endpoint in the overall population. There were no differences between birtamimab and placebo in the three key secondary endpoints in the overall population (**Table S4**).

Post hoc analyses of time to ACM in Mayo Stage IV patients were subsequently performed to better understand the treatment effect in high-risk patients, with survival censored at 9 months, as explained in the methods. The post hoc analysis showed significant improvement in ACM at month 9 for birtamimab + SOC compared with

placebo + SOC (HR = 0.413; 95% CI 0.191-0.895; log-rank *P* = .021; **Figure 1A**). The median survival in Mayo Stage IV patients was not reached (>9 months) in the birtamimab + SOC arm versus 8.3 months in the placebo + SOC arm. At month 9, the proportion of Mayo Stage IV patients surviving was 74% versus 49% in the birtamimab and placebo arms, respectively. Separation of the birtamimab survival curve from the placebo curve occurred early (ie, starting at approximately 1 month) and was sustained throughout the study. Sensitivity analyses by baseline characteristics confirmed the robustness of the ACM result in Mayo Stage IV patients (**Figure 1B**). The effect modification analysis comparing HRs for ACM at month 9 between Mayo Stage I-III and Stage IV patients yielded a statistically significant interaction *P*-value for treatment and Mayo Stage (*P* = .040), suggesting that disease severity at baseline modified the treatment effect of birtamimab.

The post hoc analysis of SF-36v2 PCS scores for Mayo Stage IV patients showed significantly less worsening at month 9 in the birtamimab versus placebo arm (least squares [LS] mean [standard error (SE)]: −0.75 [1.749] vs −5.40 [1.597]; between-group difference, +4.65 [2.325], *P* = .046; **Table 2**). In a post hoc analysis in Mayo Stage IV patients, the LS mean 6MWT distance increased by 15.22 meters at month 9 with birtamimab and decreased by 21.15 meters with placebo (between-group difference, +36.37 [26.310], *P* = .022 from rank ANCOVA; **Table 2**). Rank analysis scores for 6MWT distance at month 9 and change from baseline for Mayo Stage IV patients are shown in **Table S5**. There was no difference between birtamimab and placebo for cardiac best response in Mayo Stage IV patients as assessed by changes in NT-proBNP, a biomarker which to date has not been established as a surrogate endpoint for product registration (**Table S6**). This analysis is limited by missing laboratory data due to early termination of the study. There was no

significant difference in the proportion of Mayo Stage IV patients who achieved a hematologic response ≥VGPR by month 3 between treatment arms (relative risk [95% CI], 1.08 [0.56, 2.07]; *P* = .822). In the birtamimab and placebo arms, 12/38 patients and 11/39 patients, respectively, achieved a hematologic response ≥VGPR.

Safety

Multiple intravenous infusions of birtamimab were generally safe and well tolerated overall and in Mayo Stage IV patients. The rates of TEAEs (all events and serious events) were balanced between treatment groups in the overall population (**Table 3**). Fatal TEAEs occurred in 15% of patients with birtamimab and 22% of patients with placebo. Consistent with the underlying disease, cardiac disorders was the most common class of fatal TEAEs, occurring in 9 patients (7%) in the birtamimab arm and 18 patients (14%) in the placebo arm. The most common (≥10% of patients in either treatment group) grade ≥3 TEAEs by preferred term are shown in **Table 4**, and in the overall population included cardiac failure (birtamimab 13%; placebo 20%), pneumonia (birtamimab 11%; placebo 9%), and congestive cardiac failure (birtamimab 10%; placebo 7%).

In Mayo Stage IV patients, all patients reported at least one TEAE; serious TEAEs were reported in 27 patients (71%) in the birtamimab group and 29 patients (74%) in the placebo group (**Table 3**). As with the overall population, serious TEAEs in Mayo Stage IV patients were generally assessed by the investigator as unrelated to study drug. Among Mayo Stage IV patients, 4 patients (11%) in the birtamimab group and 14 patients (36%) in the placebo group experienced a TEAE resulting in death. The percentages of patients with grade ≥3 TEAEs was 79% in the birtamimab arm and 90% in the placebo arm and were generally assessed by the investigator as

unrelated to study drug (**Table 3**). The three most common grade ≥3 TEAEs in Mayo Stage IV patients were syncope (birtamimab 16%; placebo 15%), cardiac failure (birtamimab 13%; placebo 28%), and congestive cardiac failure (birtamimab 13%; placebo 8%) (**Table 4**).

In the overall population, TEAEs associated with infusions were reported for 5 patients (4%) with birtamimab and 3 patients (2%) with placebo. All infusion-associated TEAEs were nonserious and mild or moderate in severity, except for a grade 3 infusion-related reaction that occurred in a birtamimab-treated patient on day 226 and resolved on the same day. All other infusion-associated TEAEs in the birtamimab group occurred on day 1. In Mayo Stage IV patients, infusion-associated TEAEs were reported in 3 patients in the birtamimab group and included dyspnea (n = 1), chest discomfort (n = 1) and hypoxia concurring with infusion-related reaction (n = 1).

In the overall study population, 41 patients (32%) in the birtamimab arm and 42 patients (32%) in the placebo arm died during the study; except for one death in the placebo arm, all were adjudicated by the CEC. Cardiac disorders were the most common cause of death, occurring in 21 patients in the birtamimab arm and 28 patients in the placebo arm of the overall study population, consistent with the underlying disease and the known risk of cardiac complications in AL amyloidosis. Among Mayo Stage IV patients, there were 14 (37%) deaths in the birtamimab arm and 22 (56%) in the placebo arm, of which 8 and 15, respectively, were attributed to cardiac events. The largest proportion of adjudicated deaths among Mayo Stage IV patients occurred in the first 3 months of the study: 2 (5%) patients in the birtamimab arm and 12 (31%) patients in the placebo arm.

Discussion

VITAL was the first randomized, placebo-controlled phase 3 trial of an amyloid-depleter therapy combined with SOC chemotherapy in AL amyloidosis patients with cardiac involvement (NT-proBNP ≥650 and ≤8500 pg/mL). It is unlikely the study would have been able to detect a difference in survival between treatment groups in Mayo Stages I-III without a considerably longer duration of treatment, given the reported median survival for Mayo Stage I, II and III patients of approximately 94, 40, and 14 months, respectively.[12] The primary composite endpoint of time to ACM or CH favored birtamimab, although the difference between treatment arms did not reach pre-specified significance. Post hoc analyses in Mayo Stage IV patients showed a potential effect of birtamimab on mortality in patients with the highest risk of early mortality. Analyses in Mayo Stage IV patients were conducted using time to ACM at month 9 as the efficacy endpoint, based on median survival in the Mayo Stage IV placebo group of 8.3 months, and to align with the key secondary endpoints (change from baseline to 9 months). In patients with Mayo Stage IV disease, significant improvement in survival with birtamimab + SOC was observed at month 9 (HR = 0.413; 95% CI 0.191-0.895; log-rank *P* = .021). An effect modification analysis confirmed that severity of disease at baseline impacted the observed treatment effect of birtamimab, which may be attributable to the paucity of events in Mayo Stage I-III patients over the duration of the study.

Treatment with birtamimab in Mayo Stage IV patients was associated with significantly less deterioration in QoL, as measured by SF-36v2 PCS, and improved cardiac functioning, per 6MWT. In this subgroup, treatment with placebo + SOC led to a substantial decline in 6MWT distance over 9 months (approximately 21 meters),

whereas distance increased by approximately 15 meters with birtamimab + SOC during the same period. This suggests, in addition to potentially imparting a survival benefit, birtamimab may also confer a clinically meaningful impact on QoL and functional capacity in patients with advanced disease.

Newly diagnosed Mayo Stage IV patients are at high risk for early death within 6 months of diagnosis (median overall survival, 5.8 months), with cardiac failure being the leading cause of death. [6,12,18] Consistent with this previously reported mortality risk, over 50% (12/22) of the deaths in Mayo Stage IV patients treated with placebo + SOC during this clinical trial occurred within the first 3 months. Current SOC in AL amyloidosis consists of repurposed multiple myeloma therapies and is aimed at reducing or eliminating the plasma cell dyscrasia rather than directly depleting existing AL amyloid deposits or targeting toxic soluble LC aggregates. [6,13,20] In contrast, birtamimab is a humanized IgG1 monoclonal antibody that directly targets a shared cryptic epitope on misfolded κ and λ immunoglobulin LCs and is designed to neutralize toxic soluble aggregates of misfolded LCs, prevent aggregation of newly produced LCs, and deplete existing insoluble organ-deposited amyloid. [21,22] SOC therapies typically require ≥6 months to achieve organ responses, which are evaluated using biomarkers. [17] Notably, we observed no difference in the hematologic response rates between treatment arms, which suggests the observed potential survival benefit with birtamimab was not due to higher hematologic response, consistent with birtamimab's mechanism of action.

As Mayo Stage IV patients are at the highest risk for early mortality, novel safe and effective therapies to rapidly deplete organ-deposited amyloid are urgently needed. [12,13,20] To our knowledge, no other investigational or approved therapy for AL amyloidosis has demonstrated a survival benefit in Mayo Stage IV patients. Our post

hoc analysis was restricted to patients with NT-proBNP between 1800 and 8500 pg/mL; nonetheless, mortality in the placebo arm was generally consistent with historical survival rates in Mayo Stage IV patients, who are at high risk for early death. [12,27] Furthermore, the post hoc result observed here with birtamimab + SOC in Mayo Stage IV patients (median survival not reached, >9 months) suggests that birtamimab could play a role in achieving an early survival benefit in patients with advanced AL amyloidosis.

Patients with advanced AL amyloidosis are typically frail and have numerous underlying comorbidities, making them less tolerant of SOC. [6,20,28] Many SOC therapies are associated with AEs that can worsen patients' clinical status. [13,20] Poor tolerability can lead to treatment discontinuation and detrimentally impact the ability to achieve a robust hematologic response, [26,28] highlighting the unmet need for novel therapeutics with a favorable benefit–risk profile for advanced AL amyloidosis. In VITAL, once-monthly intravenous infusions of birtamimab (median, 15.5 infusions) over a median follow-up of approximately 15 months were generally safe and well tolerated, and the safety profile in patients with Mayo Stage IV disease was generally consistent with that in the overall study population. Additionally, infusion-associated TEAEs occurred with relatively low frequency: in 5 patients with birtamimab and 3 patients with placebo; all were mild or moderate with birtamimab and generally occurred early during treatment.

Limitations of post hoc analyses are well known; by nature, they have greater potential for type 1 error, meaning there is increased potential for a false-positive result. Thus, the findings from these post hoc analyses should be interpreted with caution. Due to early termination of the trial based on futility analysis, post hoc efficacy analyses were limited to 9 months, and longer-term follow-up for survival

was not possible. Immunogenicity and pharmacokinetic data from VITAL were not analyzed; however, previous data from the phase 1/2 clinical trial of birtamimab demonstrated a well-behaved IgG1-like pharmacokinetic profile that did not appear affected by underlying renal, cardiac, or neurological involvement and showed no antidrug antibodies among 27 birtamimab-treated patients.[25] These results are consistent with birtamimab being a humanized monoclonal antibody, as reducing the amount of non-human sequence in monoclonal antibodies has been associated with decreased risk of immunogenicity.[29]

Because of the significant survival and clinical benefits observed with birtamimab + SOC in the post hoc analyses of VITAL reported here, a confirmatory global phase 3 randomized, double-blind, placebo-controlled clinical trial of birtamimab in patients with Stage IV AL amyloidosis, AFFIRM-AL (ClinicalTrials.gov Identifier: NCT04973137), is being conducted under a SPA agreement with the US FDA.[30]

Conclusion

The phase 3 VITAL clinical trial was stopped early based on a recommendation from the independent data monitoring committee following the results of a futility analysis that suggested the primary endpoint was unlikely to be met. Post hoc analyses demonstrated a significant survival benefit with birtamimab and significant improvements in QoL and functional capacity in patients at the highest risk for early mortality (Mayo Stage IV). Overall, the incidence, severity, and seriousness of AEs were similar in each treatment group, indicating that birtamimab was generally safe and well tolerated. Given the urgent unmet need for treatments that improve survival in patients with advanced AL amyloidosis, the confirmatory AFFIRM-AL study of

birtamimab (NCT04973137) in this patient population is ongoing under a SPA agreement with the US FDA.

Acknowledgments

Medical writing and editorial support were provided by Emily Mercadante PhD of AMICULUM Ltd., funded by Prothena Biosciences, Ltd. The authors would like to thank Peloton Advantage, LLC, an OPEN Health company, for their contributions toward initial drafting of the manuscript. We thank Martin Koller for his contributions to the VITAL Study design and data acquisition.

This clinical trial was sponsored by Prothena Biosciences, Ltd, Dublin, Ireland, a member of the Prothena Corporation plc group. Prothena Biosciences, Ltd participated in the design and conduct of the study; collection, management, analysis, and interpretation of the data; and preparation, review, and approval of the manuscript.

Authorship

Contributions: M.G, J.W., and G.K. designed the study; M.G., J.W., S.G. and G.K. were responsible for the conduct of the study; C.N. was responsible for data collection; and Y.J. performed analysis of data. All authors had full access to study data and take responsibility for the integrity of the data and the accuracy of the data analysis. C.N., C.K., Y.J., G.K. and M.G. assisted in preparation of the manuscript. All authors revised the manuscript and reviewed and approved the final version for submission.

Conflict-of-interest disclosure: M.A.G.: personal fees from Alnylam, Aptitude Health, Ashfield, Celgene, Ionis/Akcea, Janssen, Johnson & Johnson, Juno, Physicians Education Resource, Prothena, Research to Practice, Sanofi, Sorrento; personal fees for Data Safety Monitoring board from Abbvie; payment for development of educational materials for i3Health and educational program development for i3Health; grant funding from NCI SPORE MM SPORE 5P50 CA186781-04. A.D.C.: Consultant for Abbvie, Arcellx, AstraZeneca, BMS, Celgene, Genentech/Roche,

GlaxoSmithKline, Ichnos, Janssen, Kite Pharma, Oncopeptides, Pfizer, Seattle Genetics, Takeda; research funding from GlaxoSmithKline and Novartis; patents and royalties from Novartis. R.L.C.: Consultant and advisor for Caelum; consultant and advisor for, and research funding from Janssen, Prothena; research funding from Karyopharm, Takeda; advisor and research funding for Unum; royalties for Patent 9593332, Pending 20170008966. E.K.: Consultant for Amgen, Janssen, Genesis, Takeda, Prothena, Pfizer; received research funding from Amgen, Janssen. H.J.L.: Advisor for Caelum, Janssen, Prothena, Pfizer; consultant and advisor for Celgene; consultant for Karyopharm; advisor and research funding from Takeda; research funding from Amgen. E.N.L.: Consultant for Abbvie, Akcea, Alnylam, Pharmacyclics, Janssen. M.L.: Research funding from Alexion, Caelum, Prothena; advisor for Janssen, Takeda. V.S.: Consultant and advisor for and research funding from Caleum; consultant for Attralus, Pfizer; research funding from Celgene, Karyopharm, Millennium-Takeda, Oncopeptides, Prothena, Sorrento; research funding from and advisor for Janssen; advisor for Abbvie, Proclara, Protego, Pharmatrace, Prothena, Regeneron, Telix. S.S.: Advisor for, travel grant, honoraria, and research funding from Janssen, Prothena; research funding from Sanofi; honoraria from Pfizer, Takeda; advisor for Telix; travel grant from Binding Site, Celgene, Jazz. A.W.: Research funding from Amgen; honoraria from Alexion, Celgene, GlaxoSmithKline, Janssen-Cilag, Prothena, Takeda. J.A.Z.: Consultant/advisor for Alnylam, Amgen, BMS, Caelum, Celgene, Intellia, Janssen, Oncopeptides, Takeda; research funding from BMS, Celgene. GP: advisor for and honoraria from Janssen, Protego, Zentalis; honoraria from Pfizer, Sebia, Siemens, The Binding Site. J.W.: employed by Consulting JW LLC; consultant for 4DMT, Aduro/Chinook, Alpha Holdings, Ambagon, Aminex, Arch Oncology, AroBio, Benevolent/BAI, CALIBR – Scripps, Cocept, Crinetics Pharmaceuticals, Crown BioScience, Cumulus Oncology Limited, Cybrexa, CytomX, Dren Bio, Entos, Excure Inc., Flag, Fulgent, Harpoon, Immune Onc, ImmuNext, InClin, January Therapeutics, Janux Therapeutics, Myovant, Nurix, Nuvation Bio, Inc. OPNA Bio LLC., Orbus, Orphagen, Oryzon, Plexxikon, Propella, Que Oncology, Sagamore, Sesen Bio, Shape Therapeutics, Sonnet Bio Therapeutics, Trex, venBio Partners LLC; payment for expert testimony from Puma Biotechnology. S.G.: founder of and employed by Attralus, Inc.; owns stock or stock options in Prothena and Attralus; named inventor on patents related to

birtamimab and Attralus. C.N., C.K., Y.J.: Employment and stockholder at Prothena. G.G.K.: Employment and equity ownership in Prothena; named inventor on multiple patents and patent applications related to birtamimab. G.M.: consultant for GlaxoSmithKline, Novo Nordisk; travel grant from Janssen.

The current affiliation for J.W. is Consulting JW LLC, San Francisco, CA, USA.

The current affiliation for S.G. is Attralus Inc., San Francisco, CA, USA.

References

1. Kyle RA, Gertz MA. Primary systemic amyloidosis: clinical and laboratory features in 474 cases. *Semin Hematol*. 1995;32(1):45-59.

2. Kyle RA, Linos A, Beard CM, et al. Incidence and natural history of primary systemic amyloidosis in Olmsted County, Minnesota, 1950 through 1989. *Blood*. 1992;79(7):1817-1822.

3. Pinney JH, Smith CJ, Taube JB, et al. Systemic amyloidosis in England: an epidemiological study. *Br J Haematol*. 2013;161(4):525-532.

4. Quock TP, Yan T, Chang E, Guthrie S, Broder MS. Epidemiology of AL amyloidosis: a real-world study using US claims data. *Blood Adv*. 2018;2(10):1046-1053.

5. Dittrich T, Kimmich C, Hegenbart U, Schonland SO. Prognosis and staging of AL amyloidosis. *Acta Haematol*. 2020;143(4):388-400.

6. Muchtar E, Dispenzieri A, Gertz MA, et al. Treatment of AL amyloidosis: Mayo Stratification of Myeloma and Risk-Adapted Therapy (mSMART) Consensus Statement 2020 update. *Mayo Clin Proc*. 2021;96(6):1546-1577.

7. Merlini G, Dispenzieri A, Sanchorawala V, et al. Systemic immunoglobulin light chain amyloidosis. *Nat Rev Dis Primers*. 2018;4(1):38.

8. Milani P, Merlini G, Palladini G. Light chain amyloidosis. *Mediterr J Hematol Infect Dis.* 2018;10(1):e2018022.

9. Janssen S, Van Rijswijk MH, Meijer S, Ruinen L, Van der Hem GK. Systemic amyloidosis: a clinical survey of 144 cases. *Neth J Med*. 1986;29(11):376-385.

10. Quock TP, Chang E, Munday JS, D'Souza A, Gokhale S, Yan T. Mortality and healthcare costs in Medicare beneficiaries with AL amyloidosis. *J Comp Eff Res*. 2018;7(11):1053-1062.

11. Weiss BM, Lund SH, Bjorkholm M, et al. Improved survival in AL amyloidosis: a population-based study on 1,430 patients diagnosed in Sweden 1995-2013. *Blood*. 2016;128(22):4448-4448.

12. Kumar S, Dispenzieri A, Lacy MQ, et al. Revised prognostic staging system for light chain amyloidosis incorporating cardiac biomarkers and serum free light chain measurements. *J Clin Oncol*. 2012;30(9):989-995.

13. Wechalekar AD, Cibeira MT, Gibbs SD, et al. Guidelines for non-transplant chemotherapy for treatment of systemic AL amyloidosis: EHA-ISA working group. *Amyloid*. 2022:1-15.

14. Sanchorawala V, Boccadoro M, Gertz M, et al. Guidelines for high dose chemotherapy and stem cell transplantation for systemic AL amyloidosis: EHA-ISA working group guidelines. *Amyloid*. 2022;29(1):1-7.

15. Janssen Biotech. Darzalex faspro [package insert]. Horsham, PA; 2022. Accessed December 2022.

16. Kastritis E, Palladini G, Minnema MC, et al. Daratumumab-based treatment for immunoglobulin light-chain amyloidosis. *N Engl J Med*. 2021;385(1):46-58.

17. Muchtar E, Dispenzieri A, Leung N, et al. Depth of organ response in AL amyloidosis is associated with improved survival: grading the organ response criteria. *Leukemia*. 2018;32(10):2240-2249.

18. Staron A, Zheng L, Doros G, et al. Marked progress in AL amyloidosis survival: a 40-year longitudinal natural history study. *Blood Cancer J*. 2021;11(8):139.

19. Muchtar E, Dispenzieri A, Wisniowski B, et al. Graded Cardiac Response Criteria for Patients With Systemic Light Chain Amyloidosis. *J Clin Oncol*. 2022:JCO2200643.

20. Palladini G, Merlini G. How I treat AL amyloidosis. *Blood*. 2022;139(19):2918-2930.

21. Renz M, Torres R, Dolan PJ, et al. 2A4 binds soluble and insoluble light chain aggregates from AL amyloidosis patients and promotes clearance of amyloid deposits by phagocytosis (dagger). *Amyloid*. 2016;23(3):168-177.

22. Wall JS, Kennel SJ, Williams A, et al. AL amyloid imaging and therapy with a monoclonal antibody to a cryptic epitope on amyloid fibrils. *PLoS One*. 2012;7(12):e52686.

23. Prothena achieves orphan drug status in EU for lead program NEOD001 [press release]. Dublin, Ireland: Prothena; 2013. https://ir.prothena.com/news-releases/news-release-details/prothena-achieves-orphan-drug-status-eu-lead-program-neod001. Accessed 29 January 2021

24. Prothena receives FDA fast track designation for NEOD001, a monoclonal antibody for the treatment of patients with AL amyloidosis [press release]. Dublin, Ireland: Prothena; 2014. https://ir.prothena.com/news-releases/news-release-details/prothena-receives-fda-fast-track-designation-neod001-monoclonal. Accessed 29 January 2021

25. Gertz MA, Landau H, Comenzo RL, et al. First-in-human Phase I/II study of NEOD001 in patients with light chain amyloidosis and persistent organ dysfunction. *J Clin Oncol*. 2016;34(10):1097-1103.

26. Palladini G, Hegenbart U, Milani P, et al. A staging system for renal outcome and early markers of renal response to chemotherapy in AL amyloidosis. *Blood*. 2014;124(15):2325-2332.

27. Abdallah N, Dispenzieri A, Muchtar E, et al. Prognostic restaging after treatment initiation in patients with AL amyloidosis. *Blood Adv*. 2021;5(4):1029-1036.

28. Rizio AA, White MK, McCausland KL, et al. Treatment tolerability in patients with immunoglobulin light-chain amyloidosis. *Am Health Drug Benefits*. 2018;11(8):430-437.

29. Foltz IN, Karow M, Wasserman SM. Evolution and emergence of therapeutic monoclonal antibodies: what cardiologists need to know. *Circulation*. 2013;127(22):2222-2230.

30. Prothena announces confirmatory Phase 3 AFFIRM-AL study of birtamimab in Mayo Stage IV patients with AL amyloidosis under SPA Agreement with FDA [press release]. Dublin, Ireland: Prothena Corporation; 2021. https://www.globenewswire.com/news-release/2021/02/01/2167691/0/en/Prothena-Announces-Confirmatory-Phase-3-AFFIRM-AL-Study-of-Birtamimab-in-Mayo-Stage-IV-Patients-with-AL-Amyloidosis-under-SPA-Agreement-with-FDA.html?print=1. Accessed 9 February 2021

TABLES

Table 1. Demographics and baseline disease characteristics

	All patients (n = 260)		Mayo Stage IV patients (n = 77)	
	Birtamimab + SOC (n = 130)	**Placebo + SOC (n = 130)**	**Birtamimab + SOC (n = 38)**	**Placebo + SOC (n = 39)**
Age, years, median (Q1, Q3)	64.2 (57.6, 70.9)	62.6 (57.0, 69.3)	63.6 (55.7, 69.8)	63.7 (57.0, 68.4)
Gender, n (%)				
Male	82 (63)	90 (69)	25 (66)	28 (72)
Female	48 (37)	40 (31)	13 (34)	11 (28)
Ethnicity, n (%)				
Hispanic or Latino	2 (2)	2 (2)	0	0
Not Hispanic or Latino	116 (89)	122 (94)	34 (90)	36 (92)
Not provided or unknown	12 (9)	6 (5)	4 (11)	3 (8)
Race, n (%)				
White	118 (91)	120 (92)	36 (95)	36 (92)
Black or African American	9 (7)	3 (2)	2 (5)	2 (5)
Asian	2 (2)	2 (2)	0	0
Other	1 (1)	5 (4)	0	1 (3)
Age at AL amyloidosis diagnosis, years, median (Q1, Q3)	64.1 (57.5, 70.9)	62.4 (56.8, 69.3)	63.5 (55.6, 69.7)	63.8 (56.8, 68.4)
Duration since AL amyloidosis diagnosis, months, median (Q1, Q3)	1.31 (0.92, 1.87)	1.48 (0.95, 2.17)	1.15 (0.69, 1.58)	1.45 (0.89, 1.81)
Number of derived involved organs at baseline, median (Q1, Q3)	2.0 (1.0, 2.0)	2.0 (1.0, 2.0)	1.5 (1.0, 2.0)	2.0 (1.0, 2.0)
Baseline NT-proBNP ≥1800 pg/mL, n (%)	95 (73)	100 (77)	38 (100)	39 (100)

Baseline NT-proBNP, pg/mL, median (Q1, Q3)	3146.2 (1650.0, 5173.0)	3183.7 (1910.0, 5551.0)	5141.3 (3228.0, 5939.4)	5415.0 (4054.0, 8073.0)
Baseline troponin-T ng/mL,* median (Q1, Q3)	0.03 (0.02, 0.06)	0.02 (0.02, 0.08)	0.05 (0.04, 0.09)	0.09 (0.06, 0.13)
Baseline FLC ratio, median (Q1, Q3)	0.10 (0.03, 0.32)	0.11 (0.04, 0.51)	0.05 (0.02, 0.08)	0.05 (0.03, 11.14)
Baseline dFLC,† mg/dL, median (Q1, Q3)	26.31 (13.83, 53.05)	38.18 (18.00, 63.06)	44.44 (25.13, 56.17)	57.42 (35.52, 106.28)
Mayo Stage, n (%)				
I	11 (8)	10 (8)	NA	NA
II	34 (26)	28 (22)	NA	NA
III	47 (36)	53 (41)	NA	NA
IV	38 (29)	39 (30)	38 (100)	39 (100)
Renal Stage, n (%)				
I	91 (70)	96 (74)	28 (74)	29 (74)
II	33 (25)	26 (20)	9 (24)	9 (23)
III	6 (5)	8 (6)	1 (3)	1 (3)
Baseline 6MWT distance, n (%)				
<300 meters	44 (34)	43 (33)	13 (34)	16 (41)
≥300 meters	86 (66)	87 (67)	25 (66)	23 (59)

*Mayo Stage criteria for troponin-T levels were modified from a value of 0.025 ng/mL cited in Kumar et al[12] to 0.03 ng/mL, the lowest validated determination for the commercially available test.

†Baseline dFLC is calculated only for patients with an abnormal baseline FLC ratio (kappa/lambda <0.26 or >1.65) and is defined as the difference between involved and uninvolved FLCs.

NA indicates not applicable.

Table 2. Change from baseline in QoL and functional capacity at month 9 in Mayo Stage IV patients

Endpoints	Mayo Stage IV patients (n = 77)			
	Birtamimab + SOC (n = 38)	Placebo + SOC (n = 39)	Group difference	*P*
SF-36v2 PCS				
Baseline score, mean (SD)	33.61 (8.753)	33.75 (9.972)	NA	
Change from baseline at month 9, LS mean (SE)*	−0.75 (1.749)	−5.40 (1.597)	+4.65 (2.325)†	.046
6MWT distance				
Baseline distance (meters), mean (SD)	336.10 (101.722)	322.63 (100.484)	NA	
Change from baseline at month 9 (meters), LS mean (SE)*	15.22 (20.010)	−21.15 (20.632)	+36.37 (26.310)†	.022‡

*Estimates of the LS mean and SE for each treatment group were estimated using an MMRM methodology including fixed effects for treatment group, categorical time point (all postbaseline visits), treatment group by visit interaction, IWRS stratification factors (Renal Stage: I, II/III; baseline 6MWT distance: <300 meters, ≥300 meters), the associated baseline value as a covariate, and a compound symmetry covariance structure to model the within-patient errors.
†Group difference favors birtamimab.
‡*P*-value from rank ANCOVA; prior to analysis, patients were ranked from worst to best following the 7-step algorithm.
IWRS indicates interactive web response system; QoL, quality of life; and SD, standard deviation.

Table 3. Overall summary of TEAEs (safety population) and most commonly reported TEAEs by preferred term

	All patients (n = 260), n (%)		Mayo Stage IV patients (n = 77), n (%)	
	Birtamimab + SOC (n = 130)	Placebo + SOC (n = 130)	Birtamimab + SOC (n = 38)	Placebo + SOC (n = 39)
Patients reporting ≥1 of the following:				
TEAE*	127 (98)	130 (100)	38 (100)	39 (100)
Treatment-related TEAE	41 (32)	50 (38)	12 (32)	10 (26)
TEAE grade ≥3	96 (74)	102 (78)	30 (79)	35 (90)
Treatment-related TEAE grade ≥3	6 (5)	12 (9)	1 (3)	4 (10)
Serious TEAE	88 (68)	91 (70)	27 (71)	29 (74)
Treatment-related serious TEAE	4 (3)	5 (4)	1 (3)	1 (3)
TEAE leading to study drug withdrawal	6 (5)	14 (11)	3 (8)	2 (5)
TEAE leading to death	19 (15)	28 (22)	4 (11)	14 (36)
Treatment-related TEAE leading to death	0	0	0	0
Most commonly reported TEAE by preferred term†				
Fatigue	57 (44)	52 (40)	15 (39)	16 (41)
Nausea	56 (43)	44 (34)	16 (42)	12 (31)
Peripheral edema	56 (43)	56 (43)	21 (55)	19 (49)
Constipation	54 (42)	55 (42)	16 (42)	13 (33)

Diarrhea	52 (40)	54 (42)	12 (32)	17 (44)
Dyspnea	40 (31)	41 (32)	16 (42)	12 (31)
Insomnia	39 (30)	30 (23)	12 (32)	8 (21)
Cough	31 (24)	27 (21)	11 (29)	5 (13)
Hypokalemia	26 (20)	27 (21)	12 (32)	10 (26)
Dizziness	25 (19)	39 (30)	3 (8)	11 (28)
Cardiac failure	24 (18)	30 (23)	8 (21)	12 (31)
Hypotension	19 (15)	33 (25)	4 (11)	12 (31)

*Patients reporting more than 1 TEAE are counted only once using the closest relationship to study drug, as assessed by the investigator.
†Occurring in ≥25% of patients in either treatment group (overall population or Mayo Stage IV patients) and ordered from highest to lower percentage in the birtamimab overall study population (all patients).

Table 4. Most commonly reported Grade ≥3 TEAEs by preferred term*

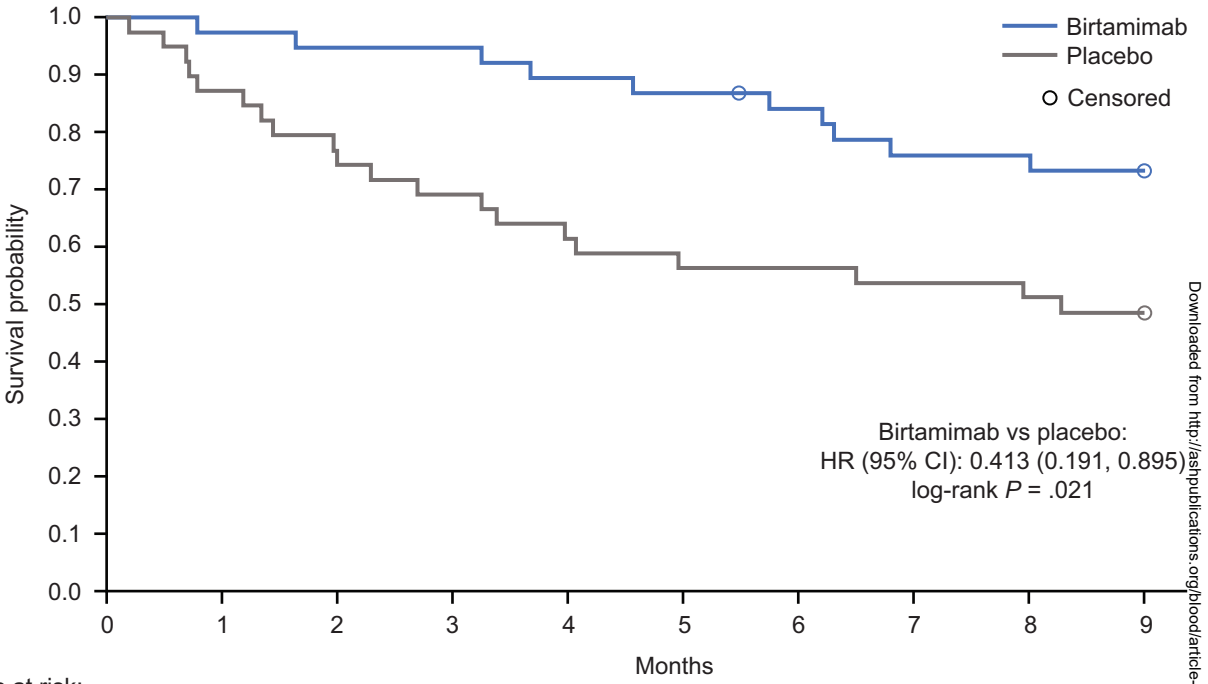
	All patients (n = 260), n (%)		Mayo Stage IV patients (n = 77), n (%)	
	Birtamimab + SOC (n = 130)	Placebo + SOC (n = 130)	Birtamimab + SOC (n = 38)	Placebo + SOC (n = 39)
Cardiac failure	17 (13)	26 (20)	5 (13)	11 (28)
Pneumonia	14 (11)	12 (9)	4 (11)	1 (3)
Congestive cardiac failure	13 (10)	9 (7)	5 (13)	3 (8)
Syncope	13 (10)	17 (13)	6 (16)	6 (15)
Peripheral edema	8 (6)	10 (8)	1 (3)	5 (13)
Diarrhea	6 (5)	7 (5)	2 (5)	5 (13)
Hypokalemia	5 (4)	7 (5)	2 (5)	4 (10)
Lymphopenia	5 (4)	8 (6)	4 (11)	3 (8)
Hypotension	4 (3)	7 (5)	1 (3)	5 (13)

*Occurring ≥10% patients in either treatment group (overall population or Mayo Stage IV patients) and ordered from highest to lower percent in the birtamimab overall study population (all patients). Patients reporting more than one TEAE are counted only once using the closest relationship to study drug, as assessed by the investigator.

Figure Legends

Figure 1. All-cause mortality at month 9 among Mayo Stage IV patients. (A) Kaplan-Meier estimate of ACM with data censored at 9 months and (B) Forest plot of ACM at month 9 adjusted for baseline characteristics in Mayo Stage IV patients. HR and 90% two-sided CIs were estimated from the semiparametric Cox Regression model stratified by randomization strata (ie, Renal Stage I vs II/III, and 6MWT distance), and with baseline variables including age, sex, race, ethnicity, age at diagnosis, duration since diagnosis, NT-proBNP, dFLC, FLC, NYHA class, troponin-T, and 6MWT distance added separately. All baseline variables except for categorical variables (ie sex, race, ethnicity, NYHA class) are adjusted as continuous variables.

A



No. of patients at risk:

Birtamimab	38	37	36	36	34	33	31	28	28	27
Placebo	39	34	30	27	24	22	22	21	20	19

B