**Original Research**
# Rationale and Design of the Alpha-1 Biomarkers Consortium Study

Monica P. Goldklang[1] Cheryl Pirozzi[2] Igor Barjaktarevic[3] Surya Bhatt[4] Sandeep Bodduluri[4] M. Bradley Drummond[5] D. Kyle Hogarth[6] Alison Keaveny[7] Zhonghua Liu[1] Noel G. McElvaney[8] Oliver J. McElvaney[8] Nadine Nuchovich[7] Sabrina Palumbo[1] Randel Plant[7] Robert Sandhaus[9] J. Michael Wells[4] Andrew Wilson[10] Charlie Strange[11] Jeanine M. D'Armiento[1] for the Alpha-1 Biomarker Consortium Study Group*

[1]Columbia University, New York, New York, United States

[2]University of Utah, Salt Lake City, Utah, United States

[3]University of California, Los Angeles, Los Angeles, California, United States

[4]University of Alabama at Birmingham, Birmingham, Alabama, United States

[5]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States

[6]University of Chicago, Chicago, Illinois, United States

[7]Alpha-1 Foundation, Coral Gables, Florida, United States

[8]Royal College of Surgeons in Ireland, Dublin, Ireland

[9]National Jewish Health, Denver, Colorado, United States

[10]Boston University, Boston, Massachusetts, United States

[11]Medical University of South Carolina, Charleston, South Carolina, United States

**Members of the Alpha-1 Biomarker Consortium Study Group are listed in the Acknowledgements section.*

***Address correspondence to:***
Jeanine M. D'Armiento
Columbia University
New York, New York
Phone: (212) 305-3745
Email: jmd12@cumc.columbia.edu

***Running Head:*** **The Alpha-1 Biomarkers Consortium Study**

***Keywords:*** alpha-1 antitrypsin deficiency; emphysema; computed tomography; biomarkers; spirometry




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

***Abbreviations :*** AATD: Alpha-1 antitrypsin deficiency*;* COPD: Chronic obstructive pulmonary disease*;* CT: Computed tomography *;* AECOPD: Acute exacerbation of COPD*;* PBMCs: Peripheral blood mononuclear cells*;* iPSCs: inducible pluripotent stem cells *;* NHLBI: National Heart, Lung, and Blood Institute *;* QUANTUM-1: the QUANTitative lung CT UnMasking emphysema progression in AATD*;* EARCO: European Alpha-1 Research Collaboration; NE: Neutrophil elastase; ECM: Extracellular matrix*;* Perc15: 15$^{th}$ percentile Hounsfield Unit value*;* AFD: Airway fractal dimension*;* $FEV_1$: Forced expiratory volume in one second*;* A1BC: Alpha-1 Biomarkers Consortium*;* QOL: Quality of life*;* A1F: Alpha-1 Foundation*;* WCG: Western Institutional Review Board-Copernicus Group*;* PRO: Patient reported outcome*;* PHQ-9: Patient Health Questionnaire*;* SGRQ: St George's Respiratory Questionnaire*;* CAT: COPD Assessment Test*;* SOBQ: University of California San Diego, Shortness of Breath Questionnaire*;* mMRC: Modified Medical Research Council Dyspnea scale*;* BCSS: Breathlessness, Cough, and Sputum Scale*;* CLDQ: Chronic Liver Disease Questionnaire*;* AUDIT-C: Alcohol Use Disorder Identification Test*;* TLC: Total lung capacity*;* RV: Residual volume*;* BMI: Body mass index*;* UAD: United airway disease*;* ECLIPSE: Evaluation of COPD Longitudinally to Identify Predictive Surrogate Endpoints*;* AcPGP: Acetylated proline-glycine-proline*;* MMPs: Matrix metalloproteinases*;* REDCap: Research Electronic Data Capture*;* OSMB: Observational Safety Monitoring Board

***Funding Support:*** This work was supported by the National Institutes of Health UG3/UH3 HL152323 and the Alpha-1 Foundation.

***Date of Acceptance***: April 17, 2025 | ***Published Online***: April 24, 2025

***Citation***: Goldklang MP, Pirozzi C, Barjaktarevic I, et al. Rationale and design of the alpha-1 biomarkers consortium study. *Chronic Obstr Pulm Dis*. 2025; Published online April 24, 2025. https://doi.org/10.15326/jcopdf.2025.0603

***This article has an online supplement.***




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## ABSTRACT

**Rationale**: Alpha-1 antitrypsin deficiency (AATD) is the most common genetic cause of chronic obstructive pulmonary disease (COPD), but considerable phenotypic variability exists among affected individuals who share disease-causing variants. Therefore, a multi-center longitudinal cohort study of 270 adult participants with PiZZ AATD will be established with goal of examining how computed tomography (CT) imaging and serum and airway biomarkers can be used to explain differences in phenotypic manifestations and outcomes.

**Methods:** Study visits at enrollment, 18 months and 36 months will obtain spirometry, patient-reported outcomes and biosampling from blood, nasal mucosa and sputum. Chest CT image acquisition will be utilized for whole lung and lobar estimations of emphysema based on lung density and to test novel measurements of airway remodeling and lung tissue mechanics. Dried blood spot cards will be collected if the participant experiences an acute exacerbation of COPD (AECOPD) during the study. Genetic analysis will be performed with complete SERPINA1 sequencing, and peripheral blood mononuclear cells (PBMCs) will be isolated to generate a repository of inducible pluripotent stem cells (iPSCs).

**Results:** The cohort will be deeply characterized including imaging, physiology, and symptomatology cross-sectionally and longitudinally over a 3-year follow-up period. A validation cohort from Ireland will independently enroll patients with identical procedures.

**Conclusion:** This is the first cohort of AATD to incorporate such detailed metrics of disease including quantitative emphysema measures with the overarching goal of improving the understanding of disease heterogeneity in AATD and identifying factors associated with disease severity and progression.





Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## INTRODUCTION

Alpha-1 antitrypsin deficiency (AATD) is the most common genetic cause of chronic obstructive pulmonary disease (COPD). Although AATD is regarded as a classical Mendelian disorder, marked variability in the development and severity of disease-associated phenotypes exists[1-3]. This variability underscores the need for an improved characterization of the range of clinical outcomes associated with specific disease-causing mutations and the influence of additional modifying genes and environmental exposures on disease presentation. To date, the National Heart, Lung, and Blood Institute (NHLBI) Registry of alpha-1 antitrypsin deficiency[4,5], the QUANTitative lung CT UnMasking emphysema progression in AATD (QUANTUM-1)[6], and the control arms of some comparative studies[7-9], are the largest longitudinal studies of patients with AATD. These studies were performed before widespread quantitative computed tomography (CT) scans in the measurement of emphysema, prior to adoption of triple therapy in the management of COPD and reflect historical smoking rates in the United States population. Contemporaneously, the European Alpha-1 Research Collaboration (EARCO) International Registry[10,11] has recruited a larger numbers of patients with AATD in Europe, with extensive analyses regarding clinical characterization and blood biomarkers, but the study contains only qualitative, not quantitative measures of emphysema, in only a subset of patients. Furthermore, augmentation therapy is not widely available in all countries within the EARCO registry. Therefore, there is still much to be defined regarding the current natural history, treatment strategies for, and the clinical course of AATD for patients in the United States.

### *Pathophysiology of AATD*

Based on population studies[12,13], it is estimated that one in 2,800 to one in 5,000 individuals in the United States has a severely deficient genetic variant on both SERPINA1 sites (ZZ, or rare null alleles), extrapolating to between 67,000 and 117,000 severely deficient patients. Alpha-1 antitrypsin is a serine protease inhibitor that inactivates neutrophil elastase and matrix metalloproteinases to maintain the protease-antiprotease balance in the lung. Individuals with AATD who inherit variant alleles of *SERPINA1* that cause misfolding of the AAT protein within the hepatocyte with resultant low levels of functional plasma AAT[14,15]. This misfolded protein triggers an endoplasmic reticulum stress response in the hepatocyte with resultant liver inflammation and fibrosis[16]. Initial interest in imbalances between proteases and their




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

endogenous inhibitors stemmed from the observation of an increased incidence of emphysema in smokers with AATD[17-19]. Neutrophil elastase (NE) is well known to be a major protease involved in tissue destruction of emphysema[20,21] and AAT is a key inhibitor of elastase with its functional loss resulting in damage of extracellular matrix (ECM).

*CT Imaging in AATD*

Unlike the general COPD population that can enroll large numbers of participants to test and validate new therapies based on spirometric measures[22,23], the limited patient pool in AATD creates a challenge. Previously, CT-based measures emphysema and lung density have been utilized in AATD clinical trials. The RAPID[7] and RAPID-OLE[24] trials tested the efficacy of weekly alpha-1 antitrypsin augmentation therapy on altering changes in CT lung density (e.g. the 15$^{th}$ percentile Hounsfield Unit value, Perc15) in 180 participants and to date has established Perc15 as the most widely accepted biomarker of lung destruction in AATD. Recent advances in CT image post-processing techniques now facilitate the estimation of lung tissue biomechanics through paired inspiratory and expiratory high resolution chest CT scans. The Jacobian determinant is a measure of local volume change and substantially explains differences between density-based measures of emphysema and the degree of airflow obstruction on spirometry[25,26], but has not be evaluated in AATD. Furthermore, complex branching patterns of the airways and subtle variations in these patterns due to disease presence can be quantified using the airway fractal dimension (AFD)[27], which has been independently associated with respiratory quality of life, functional capacity, exacerbations, lung function decline, and mortality[27], again never evaluated in AATD. This study presents unique opportunities for exploring CT imaging metrics to better understand disease severity and progression. The identification of novel imaging-based biomarkers with clinical and/or pathological relevance has the potential to accelerate the pipeline of therapies available to this vulnerable patient population.

*Endpoints in AATD Clinical Trials*

General COPD studies have focused on endpoints such as change in forced expiratory volume in one second ($FEV_1$)[28] but require large numbers of patients for statistical significance given variation in $FEV_1$ over time. Interventional clinical trials require large numbers of patients (in some cases over 1,000) to demonstrate an effect on exacerbation rate[22,29]. In AATD, this




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

approach is infeasible due to a limited patient pool. The AATD clinical research community has therefore turned to methods including change in CT lung density as a surrogate marker of lung destruction in clinical trials, including the evaluation of the clinical efficacy of AAT augmentation therapy[7,24]. The identification of additional blood or imaging biomarkers with clinical or pathological relevance as meaningful intermediate endpoints that may decrease sample size and/or trial length has the potential to improve the pipeline of therapies available to this patient population.

## Methods

### *Study Objectives*

The goal of the Alpha-1 Biomarkers Consortium (A1BC) is to better understand the heterogeneity of AATD and associate blood or imaging biomarkers with specific disease phenotypes that might be applied to characterize disease severity or predict clinical outcomes. Biomarkers have been defined by an NIH working group as a "characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention"[30]. In the A1BC, biomarkers are broadly defined to include CT-based measurements of emphysema, serum, plasma and sputum biomarkers of inflammation and lung destruction, alongside outcomes such as spirometry and quality of life (QOL) questionnaires. The study further aims to determine if there are modifying genetic relationships in disease progression of AATD. Sequencing of the *SERPINA1* gene will be performed including in the promoter and enhancer region on all A1BC patients to test the hypothesis that genotype-phenotype correlations exist among PiZZ AATD patients.

### *Study design*

In 2019, the Alpha-1 Foundation (A1F) established a research registry (A1F Registry, NCT04157049) housed within A1F containing over 1,200 discrete data including contact information, extensive patient-reported, and medical record verified clinical data. The first phase of this study was to establish a clinical cohort of Alpha-1 patients with methodology to link A1BC participants to the A1F Research Registry. All self-reported PiZZ AATD participants in the A1F Registry 18 years or older were invited to join the A1BC, with a final enrollment goal of


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

270 participants. Nine A1F Clinical Resource Centers will be recruiting geographically dispersed United States patients (Table 1 and Figure 1). A separate validation cohort in Ireland has been established. Study design was approved by the Western Institutional Review Board-Copernicus Group (WCG) and both the A1F Registry (NCT04157049) and the A1BC (NCT05297812) are enrolled in clinicaltrials.gov. A1F Registry and A1BC registration numbers will be linked for data sharing. After study completion, patient data and biomaterials will be transferred to the A1F Registry and will be used in accordance with A1F's mission.

Liberal inclusion and exclusion criteria have been selected to ensure that the entire range of AATD pulmonary manifestations in the PiZZ cohort are included (Appendix Table 1). Inclusion criteria are largely based on genotype, adult age (owing to the low incidence of emphysema in children and radiation risk exposure over a lifetime), and a willingness to share data with the A1F. Participants will be included regardless of augmentation therapy status, with data collection regarding dose and interval collected on relevant patients. Crossover of augmentation therapy status during the study will be collected. Patients are excluded if listed for lung or liver transplant. Since CT metrics of emphysema serve as the primary endpoint of this study, patients with diagnoses that could cause alterations in CT imaging were specifically excluded from the cohort; such exclusions have been implemented in other trials utilizing CT densitometry including in COPDGene[31]. Therefore, patients with major thoracic surgeries, lung volume reduction procedures, and automatic internal cardiac defibrillators and similar devices were excluded. Additionally, patients with clinically significant bronchiectasis were excluded[31]. While patients cannot be enrolled in therapeutic clinical trials at the time of A1BC consent and first visit, to not hamper enrollment in the current and upcoming clinical trials across the AATD landscape, the A1BC does not prohibit enrollment in interventional clinical studies after A1BC initial visit. At each 6-month call, patients are queried regarding clinical trial enrollment, and if newly enrolled in additional clinical trials, the clinicaltrials.gov reference number is collected.

In-person visits with blood, spirometry with and without bronchodilators, and CT will occur at baseline, 18 months, and 36 months. Optional induced sputum will be performed on appropriate patients at participating centers (only 3 of 8 centers due to COVID-19 restrictions). Extensive patient reported outcome (PRO) measures will be administered. Participants will be contacted by




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

telephone every 6 months to update clinical history, obtain current medication lists, and assess for any adverse events. Patients will be invited to complete monthly surveys of exacerbation history. A Whatman 903 blood spot card and fingerstick lancet is provided to all participants at baseline, and participants will be instructed on self-collection of blood on a dried blood spot card at the day of onset, day 3, and day 7 of exacerbations. A schedule of procedures is provided in Appendix Table 2.

**Measurements**

*Questionnaires*

This cohort will undergo extensive analysis including clinical characterization of exposure history and health status (Patient Health Questionnaire [PHQ-9], A1BC Exposure Questionnaire). Table 2 lists the questionnaires used to capture clinical manifestations of lung and liver disease associated with AATD; more details are available in the online supplement.

In addition to the above, extensive AATD-related clinical questions are obtained regarding diagnosis, including method of diagnosis, familial index case, and duration between symptom onset and diagnosis. Information regarding childhood or adult asthma, bronchiectasis, and NTM infection is obtained. Specific information about augmentation dosing, intervals, and location of administration will be collected. Other non-pulmonary AATD-related clinical findings are directly queried including neonatal and childhood liver disease, liver fibrosis and cirrhosis, vasculitis, and panniculitis. Clinician-obtained and questionnaire-based information regarding smoking history and alcohol use will be collected.

*CT Imaging*

All participants enrolled in the study will undergo acquisitions of high-resolution CT scans of the chest at total lung capacity (TLC) and residual volume (RV) at the baseline visit and subsequently at 18 months and 36 months of follow-up. The acquisition protocol will be standardized, and dose adjusted according to body mass index (BMI) using the SPIROMICS chest CT protocol[32]. Subsequent CT scans will be acquired using the same scanner and the same protocol for each participant at each site, unless there is a significant change in body weight. The




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

COPDGene phantom will be used to calibrate CT scanners for quality assurance[33]. DICOM images will be transferred to the UAB Lung Imaging Laboratory using a HIPAA-compliant secure web portal system[32].

*Nasal Transcriptome*

Based on the united airway disease (UAD) hypothesis which proposes that specific inflammatory process within the respiratory tract manifests in both the upper and lower tracts[34], we hypothesized that the nasal transcriptome will be a minimally invasive "window" to view the pathobiology of AATD-related respiratory disease development. Study participants will undergo nasal swab sampling of each nare prior to sputum induction for RNA sequencing and nasal transcriptome analysis. We plan to evaluate the potential of nasal swab-based sampling to reflect the immunological and inflammatory patterns associated with development of lung disease and aim to compare the nasal to peripheral blood transcriptome patterns.

*Plasma and Serum Biomarkers*

Identification and validation of biomarkers in AATD, especially those present in blood, has remained a challenge. While Evaluation of COPD Longitudinally to Identify Predictive Surrogate Endpoints (ECLIPSE) and other studies have shown differences in lung-derived and liver derived biomarkers between patients with COPD and smoking controls[35-43] and some of these markers have been associated with disease severity (fibrinogen and CRP)[44,45], rate of decline in lung function (CC-16), exacerbation frequency (SP-D and CRP)[38,44,46-49] and risk of mortality (PARC/CCL-18)[41,50], they excluded patients with PiZZ AATD. Furthermore, although sRAGE and SP-D are associated with loss of lung tissue as assessed by CT lung density, these biomarkers are understudied in AATD-associated emphysema. Therefore, these biomarkers need to be assessed in individuals with AATD to clarify whether the associations observed in usual COPD are maintained in this rare condition. Of particular interest will be the ability of the ECLIPSE biomarkers to correlate with clinical outcome measures including rate of loss of CT lung density, as well as their ability to identify patients with frequent exacerbator and/or rapid decliner phenotypes. The optimal technique to measure blood biomarkers that associate with CT density, the hallmark of AAT lung disease, is an unbiased proteomic approach. While the A1BC




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

may be underpowered for such analyses, an unbiased proteomic analysis will be performed on all A1BC serum samples.

Central to the present investigation is the concept of a loss of balance between pro-inflammatory and anti-inflammatory mediators and between proteases and antiproteases in AATD and multiple potential biomarkers reflective of this derangement will be evaluated. Collagen breakdown by prolyl endopeptidases produce the biologically active extracellular matrix peptide acetylated proline-glycine-proline (AcPGP), a potent chemokine associated with neutrophilic inflammation in cigarette-smoke-mediated emphysema development[51-55]. In addition to neutrophilic inflammation and elastase activity, it is important to note other proteinases have been implicated in COPD pathogenesis; cathepsins and matrix metalloproteinases (MMPs) play important roles in the protease imbalance in human emphysema[56-64]. Therefore, AcPGPs and levels of MMPs in the circulating blood will be evaluated.

*Peripheral Blood Mononuclear Cell Banking and Induced Pluripotent Stem Cells*

Patient-derived iPSCs allow disease modeling in differentiated cells that contain the genome of the patient from whom they were derived including any disease-relevant variants. In recent years, iPSC models have been applied to the study of AATD-associated hepatocyte injury and have both recapitulated known disease features and extended understanding of cellular injury mechanisms[65-69]. PBMCs that can be used to generate iPSCs will be isolated from A1BC study participants and cryopreserved as live cells for banking in an established PBMC/iPSC repository[70,71]. Specific participant samples with distinct clinical phenotypes or novel genetic variants will be identified for iPSC generation and differentiation to disease-relevant lineages based on phenotypes observed in study data.

*Genetic Testing and Detection of Single Nucleotide Polymorphisms (SNPs)*

Recent advances in sequencing have led to the description of new mutations in both the exon and intron portions of the SERPINA1 gene[72]. It has been hypothesized that mutations in the promoter region can account for changes in the acute phase response and subsequent increase in AAT during infection. The functional effects of polymorphisms within both the intron and promoter region are unknown, and it is possible that there are mutations within SERPINA1 to account for


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

the phenotypic differences between patients with the PiZZ allele.

**Data Management and Analysis**

Columbia University and the A1F have established a Research Electronic Data Capture (REDCap) data entry system. REDCap is a public sourced database, which was a requirement of the NIH study. The REDCap for A1BC has linkage numbers with the A1F Registry to connect current data with historical clinical data.

*Statistical Considerations*

The goal for powering this study was to assure that longitudinal CT density changes would be seen over 3 years. Two recent studies have used 180 participants over 2 years in a two-arm therapeutic trial (RAPID) and 330 participants over 3 years in a three-arm therapeutic trial (SPARTA). Since our study is not a clinical trial and individuals with normal lung function are invited for participation, the number of 270 participants to follow over 3 years was chosen as a pragmatic goal that was also based on the number of PiZZ patients at participating sites. While the number of patients required to observe clinical and biochemical effects in augmentation naïve patients and/or those stable on augmentation therapy will be higher than the aforementioned AATD proof-of-concept studies, we expect that this number is again covered by the proposed cohort size. In RAPID evaluation of 54 patients from the placebo arm was sufficient to detect a significant change in desmosine/isodesmosine at 24 months[7,73]. Since large prospective studies correlating inflammatory biomarkers with clinical outcomes and quantitative CT metrics of emphysema in AATD are lacking, this will be the largest prospective biomarker study ever conducted in AATD.

*Association of predictors with clinical outcomes*

Marginal association analysis will be conducted using linear regression models by regressing the CT measurement of Perc15 on each of the K (K= 28~30 in total) blood biomarkers and lung function parameters (FEV1% predicted using GLI "other" equations and $FEV_1$/FVC ratio), adjusted for important baseline covariates including ever smoking (100 cigarettes in a lifetime), a categorical variable of 10 pack years of smoking, baseline CRP, age, and sex. We will utilize




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

different sets of baseline covariates for sensitivity analysis including smoking pack years. To correct for multiple testing, we plan to use both Bonferroni correction to control for family-wise error rate and use q-value to control for false discovery rate. To account for possible correlations among biomarkers, we will use the elastic-net penalized regression models with 5-fold cross-validation to select important biomarkers jointly related to the baseline Perc15 using the R package *glmnet.* Moreover, we will apply the weighted correlation network analysis method *using the R package WGCNA* to discover clusters (modules) of highly correlated biomarkers that are associated with Perc15. For each cluster consisting of multiple biomarkers, we will use kernel machine regression to capture complex nonlinear and interaction effects of multiple biomarkers associated with Perc15. Any missing data will be imputed using the multiple imputation technique implemented in the R package *mice.* We will begin with cross-sectional analysis for data collected at each of the three visits and will use linear mixed models for the repeated measurements of all three visits to assess any longitudinal change over time. Important covariates from repeated measurement analysis including the number and severity of pulmonary exacerbations will be explored.

Findings will be validated by a companion cohort of participants from the National Alpha-1 Registry of Ireland, based at the Irish Centre for Genetic Lung Disease (Beaumont Hospital/Royal College of Surgeons in Ireland, Dublin). Methods for validation will be modeled to explore significant findings from the United States cohort and discrepancies will be probed to determine the differences in covariates between the two cohorts.

*Genetic Analyses*

With sequencing data of the *SERPINA1* gene, we will first perform standard quality control and data preprocessing. SERPINA1 sequencing will be performed on the entire gene, with promoter region, exon and introns included. Then, we will perform genetic association analysis using a linear regression model for Perc15 measured at each visit and linear mixed models for the three repeatedly measured Perc15 to identify putative causal genetic variants, adjusted for sex age, and smoking. Gene-environment analysis will be performed with inclusion of an interaction term between genetic variants and important environmental variables (e.g., cigarette smoking). Additionally, we will also perform genetic interaction analysis by including interaction terms


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

between genetic variants to explore possible epistatic effects.

*Study oversight*

An observational safety monitoring board (OSMB) has been established to include two pulmonologists and one statistician. The OSMB meets approximately once every 6 months to review patient enrollment, safety related to the study procedures, and adverse events. The open OSMB meeting is attended by NIH and A1F representatives. Adverse events are defined primarily as related to study procedures. Special events of clinical interest will be captured including AECOPD, decompensated liver disease, and enrollment in clinical trials. Furthermore, incidental findings primarily related to lung nodules on CT scans will be collected.

**Potential outcomes and conclusions**

The above-described study will be the largest cohort of AATD patients with PiZZ genotype prospectively assembled, longitudinally followed and systematically analyzed with regards to biomarkers of lung disease. The characterization of a full complement of biomarkers, including blood biomarkers, CT imaging biomarkers, patient reported outcomes, exposure history, lung function, and genomics in a cohort of individuals with alpha-1 antitrypsin deficiency has never been done at this large scale in a longitudinal fashion. The evaluation of associations between these characteristics and clinical outcomes in cross-sectional analysis as well as over time in the 3-year longitudinal follow-up will improve understanding of the heterogeneity of AATD and identify factors associated with disease severity and progression. The results have the potential to improve care for individuals with AATD by identifying individuals at risk for more severe lung or liver disease manifestations earlier before deterioration and may lead to future interventions that will improve clinical outcomes.




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Acknowledgements**

**Author Contributions**

M.P.G, C.P., I.B., S.Bh., M.B.D., N.G.M, O.J.M., J.M.W, A.W., C.S., and J.M.D. conceived the study and developed the theoretical framework. All authors contributed to and approved the final manuscript.

The authors wish to thank Drs. Antonello Punturieri and Lisa Viviano from the NIH and the Alpha-1 Foundation leadership for their support if this consortium.

**Alpha-1 Biomarker Consortium Study Group**

**Columbia University:** Jeanine M. D'Armiento, Monica P. Goldklang, Zhonghua Liu, Laura D. Fonseca, Tina Zelonina, Alexis C. Fisher, Niki Trivedi, Sydney Harris, Sabrina Palumbo, Kiran K. Chada, Devipriya Harinath, Sanjiv Prasad

**University of Utah**: Cheryl Pirozzi, Cassie E Larsen, Alexis G. Vanwagoner

**University of California, Los Angeles**: Igor Barjaktarevic, Russell Buhr, John Belperio, Semi Yoon, Vyacheslav Palchevskiy, Lung Wing

**University of Alabama at Birmingham**: Mike Wells, Surya P. Bhatt, Sandeep Bodduluri, Abhiraj Pudhota

**University of North Carolina at Chapel Hill**: M. Bradley Drummond, Caleb C. Hemphill, Kelsey B. Haywood, Walker A. Long

**University of Chicago**: D. Kyle Hogarth, MD, Vanita Patel, MPH

**Alpha-1 Foundation**: Randel Plant, Alison Keaveny, Nadine Nuchovich, Jennifer Illarramendi, Charles Emala, Jon Hagstrom, Scott Santarella

**Royal College of Surgeons in Ireland**: Noel G. McElvaney, Oliver J. McElvaney, Suzanne M. Roche, Daniel D. Fraughen

**National Jewish Health**: Robert A. Sandhaus, Ariana K. Talaie

**Boston University**: Andrew Wilson, Joseph E. Kaserman, Mark Dodge, Shumin Guan

**Medical University of South Carolina**: Charlie Strange, Leah Benn, Allison Burton, Gwen Hayden, Chloe Jacobs, Kristen Neff, Melinda M. Talley, Andrea J. Swartz


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Declaration of Interest**

In the last 36 months, the following disclosures were provided. M.P.G. has contracted research support for clinical trials from Sanofi, Arrowhead, NovoNordisc, Takeda, InhibRx, Mereo, Vertex and Grifols; research grant support from the Alpha-1 Foundation, and has been paid fees for advisory work from Takeda, GSK, Sanofi, Korro, Inhibrx, Bridge Bio, and Grifols, and received support for medical writing from Takeda. C.S.P. has contracted research support for clinical trials from Inhibrx, Vertex, AstraZeneca, has received consulting fees from Takeda, and has received honoraria for lectures from Medscape, Advancing Knowledge in Healthcare, the Academy for Continued Healthcare Learning, Medscape Education, Cleveland Clinic, Alpha-1 Foundation, and Projects in Knowledge. I.B. has contracted research support for clinical trials from Theravance and Viatris, Aerogen, Takeda, Amgen, the Alpha-1 Foundation, and Johnny Carson's Foundation, has received consulting fees from AstraZeneca, Sanofi, Regeneron, Grifols, Verona Pharma, Inhibrx, Takeda, Genentech, Aerogen, Theravance and Viatris. S.Bh. has received consulting fees from Sanofi, Regeneron, GSK, Genentech, Boehringer Ingelheim, Apreo, AstraZeneca, Chiesi, Verona, Merck, has received honoraria for lectures from MedScape, IntegrityCE, Integratis Communications, Illuminate Help and Horizon CME. M.B.D. has received grants to institution for research by PCORI, Vertex, Teva Pharmaceuticals, American Lung Association, Boehringer-Ingelheim, Midmark, Inc, and National Institutes of Health, has received consulting fees from AstraZeneca, Verona, Takdea, Becker Pharma, GlaxoSmithKline, Stratos, Genentech, and Amgen, and serves on the medical and scientific advisory boards for the COPD Foundation and Alpha-1 Foundation. D.K.H. has received honoraria for lectures for Grifols, Takeda and Sanofi, and has received consulting fees from Advanced Infusion Care and Wave Life Sciences. N.G.M. has received grants for investigator-initiated studies from Grifols and the Alpha-1 Foundation, consulting fees from CSL Behring, BEAM Therapeutics, Intellia Therapeutics and Glaxo Smith Kline, support to attend meetings from Grifols. O.J.M. has received grants from the Cystic Fibrosis Foundation and the University of Washington, consulting fees from Grifols, and serves on the scientific advisory committee for Grifols. R.P. has received honoraria for participation in the Takeda Educational Materials Working Group. R.S. has received grant support from Inhibrx and Sanofi, consulting fees from Grifols, CSL Behring, Takeda, Korobio, Beam, Wave, Biomarin with payments all payments directed to the


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

not-for-profit disease management organization AlphaNet, has a pending patent through his institution for CT analysis software, is a DSMB member for Takeda, Beam and Biomarin, and has medical director roles in the Alpha-1 Foundation, AlphaNet, and AlphaNet Canada. J.M.W. has clinical trial support from the NIH, VA, ARCUS-Med, Medscape, Verona Pharma, Grifols, Alpha-1 Foundation, Inhibrx, American Lung Association, has a patent with Mereo BioPharma, has received advisory board fees from AstraZeneca, Takeda, GSK, Bavarian Nordic, Krystal Biotech, Sanofi, and Verona Pharma, and has received support for medical writing from Takeda, GSK and Verona Pharma. A.W. has received consulting fees from Takeda and is the scientific director of the Alpha-1 Foundation. C.S. has grants paid to MUSC from the Alpha-1 Foundation, Beam, Biomarin, Grifols, Krystal, Mereo, and Takeda, and is a consultant for CSL Behring, Glaxo Smith Kline, Sanofi, and Takeda. J.M.D. has grants and contracts for clinical trials from Mereo, Takeda, Arrowhead, Vertex and Inhibrx, has received consulting fees from Sanofi and Bridge Bio, and is participating in advisory boards for Sanofi and Takeda. S.Bo., A.K., Z.L., N.N., and S.P. have no disclosures.




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## REFERENCES


1. Demeo DL, Sandhaus RA, Barker AF, et al. Determinants of airflow obstruction in severe alpha-1-antitrypsin deficiency. *Thorax*. Sep 2007;62(9):806-13. doi:10.1136/thx.2006.075846
2. DeMeo DL, Silverman EK. Alpha1-antitrypsin deficiency. 2: genetic aspects of alpha(1)-antitrypsin deficiency: phenotypes and genetic modifiers of emphysema risk. *Thorax*. Mar 2004;59(3):259-64.
3. Piitulainen E, Tornling G, Eriksson S. Effect of age and occupational exposure to airway irritants on lung function in non-smoking individuals with alpha 1-antitrypsin deficiency (PiZZ). *Thorax*. Mar 1997;52(3):244-8.
4. Stoller JK, Tomashefski J, Jr., Crystal RG, et al. Mortality in individuals with severe deficiency of alpha1-antitrypsin: findings from the National Heart, Lung, and Blood Institute Registry. *Chest*. Apr 2005;127(4):1196-204. doi:10.1378/chest.127.4.1196
5. McElvaney NG, Stoller JK, Buist AS, et al. Baseline characteristics of enrollees in the National Heart, Lung and Blood Institute Registry of alpha 1-antitrypsin deficiency. Alpha 1-Antitrypsin Deficiency Registry Study Group. *Chest*. Feb 1997;111(2):394-403. doi:10.1378/chest.111.2.394
6. Beiko T, Janech MG, Alekseyenko AV, et al. Serum Proteins Associated with Emphysema Progression in Severe Alpha-1 Antitrypsin Deficiency. *Chronic Obstr Pulm Dis*. Jul 15 2017;4(3):204-216. doi:10.15326/jcopdf.4.3.2016.0180
7. Chapman KR, Burdon JG, Piitulainen E, et al. Intravenous augmentation treatment and lung density in severe alpha1 antitrypsin deficiency (RAPID): a randomised, double-blind, placebo-controlled trial. *Lancet*. Jul 25 2015;386(9991):360-8. doi:10.1016/S0140-6736(15)60860-1
8. Parr DG, Dirksen A, Piitulainen E, Deng C, Wencker M, Stockley RA. Exploring the optimum approach to the use of CT densitometry in a randomised placebo-controlled study of augmentation therapy in alpha 1-antitrypsin deficiency. *Respir Res*. Aug 13 2009;10(1):75. doi:10.1186/1465-9921-10-75
9. Dirksen A, Piitulainen E, Parr DG, et al. Exploring the role of CT densitometry: a randomised study of augmentation therapy in alpha1-antitrypsin deficiency. *Eur Respir J*. Jun 2009;33(6):1345-53. doi:10.1183/09031936.00159408
10. Ersoz H, Torres-Duran M, Turner AM, et al. Sex-Differences in Alpha-1 Antitrypsin Deficiency: Data From the EARCO Registry. *Arch Bronconeumol*. Jul 9 2024;doi:10.1016/j.arbres.2024.06.019
11. Stockley RA, Pye A, De Soyza J, Turner AM, Miravitlles M, investigators Es. The prevalence of bronchiectasis in patients with alpha-1 antitrypsin deficiency: initial report of EARCO. *Orphanet J Rare Dis*. Aug 12 2023;18(1):243. doi:10.1186/s13023-023-02830-2
12. Silverman EK, Miletich JP, Pierce JA, et al. Alpha-1-antitrypsin deficiency. High prevalence in the St. Louis area determined by direct population screening. *Am Rev Respir Dis*. Oct 1989;140(4):961-6. doi:10.1164/ajrccm/140.4.961
13. O'Brien ML, Buist NR, Murphey WH. Neonatal screening for alpha1-antitrypsin deficiency. *J Pediatr*. Jun 1978;92(6):1006-10. doi:10.1016/s0022-3476(78)80388-6
14. Crowther DC, Belorgey D, Miranda E, Kinghorn KJ, Sharp LK, Lomas DA. Practical genetics: alpha-1-antitrypsin deficiency and the serpinopathies. *Eur J Hum Genet*. Mar 2004;12(3):167-72. doi:10.1038/sj.ejhg.5201127



Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

15. Strnad P, McElvaney NG, Lomas DA. Alpha(1)-Antitrypsin Deficiency. *N Engl J Med*. Apr 9 2020;382(15):1443-1455. doi:10.1056/NEJMra1910234
16. Ordonez A, Snapp EL, Tan L, Miranda E, Marciniak SJ, Lomas DA. Endoplasmic reticulum polymers impair luminal protein mobility and sensitize to cellular stress in alpha1-antitrypsin deficiency. *Hepatology*. May 2013;57(5):2049-60. doi:10.1002/hep.26173
17. Laurell CB, Eriksson S. The electrophoretic alpha1-globulin pattern of serum in alpha1-antitrypsin deficiency. 1963. *COPD*. Mar 2013;10 Suppl 1:3-8. doi:10.3109/15412555.2013.771956
18. Wulfsberg EA, Hoffmann DE, Cohen MM. Alpha 1-antitrypsin deficiency. Impact of genetic discovery on medicine and society. *JAMA*. Jan 19 1994;271(3):217-22.
19. Silverman EK, Sandhaus RA. Clinical practice. Alpha1-antitrypsin deficiency. *N Engl J Med*. Jun 25 2009;360(26):2749-57. doi:10.1056/NEJMcp0900449
20. Sandhaus RA, Turino G. Neutrophil elastase-mediated lung disease. *COPD*. Mar 2013;10 Suppl 1:60-3. doi:10.3109/15412555.2013.764403
21. Shapiro SD, Goldstein NM, Houghton AM, Kobayashi DK, Kelley D, Belaaouaj A. Neutrophil elastase contributes to cigarette smoke-induced emphysema in mice. *Am J Pathol*. Dec 2003;163(6):2329-35. doi:10.1016/S0002-9440(10)63589-4
22. Lipson DA, Barnhart F, Brealey N, et al. Once-Daily Single-Inhaler Triple versus Dual Therapy in Patients with COPD. *N Engl J Med*. May 3 2018;378(18):1671-1680. doi:10.1056/NEJMoa1713901
23. Rabe KF, Martinez FJ, Ferguson GT, et al. Triple Inhaled Therapy at Two Glucocorticoid Doses in Moderate-to-Very-Severe COPD. *N Engl J Med*. Jul 2 2020;383(1):35-48. doi:10.1056/NEJMoa1916046
24. McElvaney NG, Burdon J, Holmes M, et al. Long-term efficacy and safety of alpha1 proteinase inhibitor treatment for emphysema caused by severe alpha1 antitrypsin deficiency: an open-label extension trial (RAPID-OLE). *Lancet Respir Med*. Jan 2017;5(1):51-60. doi:10.1016/S2213-2600(16)30430-1
25. Bhatt SP, Bodduluri S, Newell JD, et al. CT-derived Biomechanical Metrics Improve Agreement Between Spirometry and Emphysema. *Acad Radiol*. Oct 2016;23(10):1255-63. doi:10.1016/j.acra.2016.02.002
26. Bodduluri S, Newell JD, Jr., Hoffman EA, Reinhardt JM. Registration-based lung mechanical analysis of chronic obstructive pulmonary disease (COPD) using a supervised machine learning framework. *Acad Radiol*. May 2013;20(5):527-36. doi:10.1016/j.acra.2013.01.019
27. Bodduluri S, Puliyakote ASK, Gerard SE, et al. Airway fractal dimension predicts respiratory morbidity and mortality in COPD. *J Clin Invest*. Dec 3 2018;128(12):5374-5382. doi:10.1172/JCI120693
28. Zhou Y, Zhong NS, Li X, et al. Tiotropium in Early-Stage Chronic Obstructive Pulmonary Disease. *N Engl J Med*. Sep 7 2017;377(10):923-935. doi:10.1056/NEJMoa1700228
29. Criner GJ, Connett JE, Aaron SD, et al. Simvastatin for the prevention of exacerbations in moderate-to-severe COPD. *N Engl J Med*. Jun 5 2014;370(23):2201-10. doi:10.1056/NEJMoa1403086
30. Biomarkers Definitions Working G. Biomarkers and surrogate endpoints: preferred definitions and conceptual framework. *Clin Pharmacol Ther*. Mar 2001;69(3):89-95. doi:10.1067/mcp.2001.113989


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

31. Regan EA, Hokanson JE, Murphy JR, et al. Genetic epidemiology of COPD (COPDGene) study design. *COPD*. Feb 2010;7(1):32-43. doi:10.3109/15412550903499522
32. Sieren JP, Newell JD, Jr., Barr RG, et al. SPIROMICS Protocol for Multicenter Quantitative Computed Tomography to Phenotype the Lungs. *Am J Respir Crit Care Med*. Oct 1 2016;194(7):794-806. doi:10.1164/rccm.201506-1208PP
33. Sieren JP, Hoffman EA, Fuld MK, Chan KS, Guo J, Newell JD, Jr. Sinogram Affirmed Iterative Reconstruction (SAFIRE) versus weighted filtered back projection (WFBP) effects on quantitative measure in the COPDGene 2 test object. *Med Phys*. Sep 2014;41(9):091910. doi:10.1118/1.4893498
34. Simons FE. Allergic rhinobronchitis: the asthma-allergic rhinitis link. *J Allergy Clin Immunol*. Sep 1999;104(3 Pt 1):534-40. doi:10.1016/s0091-6749(99)70320-9
35. Cheng DT, Kim DK, Cockayne DA, et al. Systemic soluble receptor for advanced glycation endproducts is a biomarker of emphysema and associated with AGER genetic variants in patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. Oct 15 2013;188(8):948-57. doi:10.1164/rccm.201302-0247OC
36. Gan WQ, Man SF, Senthilselvan A, Sin DD. Association between chronic obstructive pulmonary disease and systemic inflammation: a systematic review and a meta-analysis. *Thorax*. Jul 2004;59(7):574-80. doi:10.1136/thx.2003.019588
37. Karadag F, Karul AB, Cildag O, Yilmaz M, Ozcan H. Biomarkers of systemic inflammation in stable and exacerbation phases of COPD. *Lung*. Nov-Dec 2008;186(6):403-9. doi:10.1007/s00408-008-9106-6
38. Lomas DA, Silverman EK, Edwards LD, et al. Serum surfactant protein D is steroid sensitive and associated with exacerbations of COPD. *Eur Respir J*. Jul 2009;34(1):95-102. doi:10.1183/09031936.00156508
39. Lomas DA, Silverman EK, Edwards LD, et al. Evaluation of serum CC-16 as a biomarker for COPD in the ECLIPSE cohort. *Thorax*. Dec 2008;63(12):1058-63. doi:10.1136/thx.2008.102574
40. Park HY, Churg A, Wright JL, et al. Club cell protein 16 and disease progression in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. Dec 15 2013;188(12):1413-9. doi:10.1164/rccm.201305-0892OC
41. Sin DD, Miller BE, Duvoix A, et al. Serum PARC/CCL-18 concentrations and health outcomes in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. May 1 2011;183(9):1187-92. doi:10.1164/rccm.201008-1220OC
42. Wedzicha JA, Seemungal TA, MacCallum PK, et al. Acute exacerbations of chronic obstructive pulmonary disease are accompanied by elevations of plasma fibrinogen and serum IL-6 levels. *Thromb Haemost*. Aug 2000;84(2):210-5.
43. Vestbo J, Anderson W, Coxson HO, et al. Evaluation of COPD Longitudinally to Identify Predictive Surrogate End-points (ECLIPSE). *Eur Respir J*. Apr 2008;31(4):869-73. doi:10.1183/09031936.00111707
44. Dahl M, Tybjaerg-Hansen A, Vestbo J, Lange P, Nordestgaard BG. Elevated plasma fibrinogen associated with reduced pulmonary function and increased risk of chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. Sep 15 2001;164(6):1008-11. doi:10.1164/ajrccm.164.6.2010067
45. Walter RE, Wilk JB, Larson MG, et al. Systemic inflammation and COPD: the Framingham Heart Study. *Chest*. Jan 2008;133(1):19-25. doi:10.1378/chest.07-0058




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

46. Hurst JR, Vestbo J, Anzueto A, et al. Susceptibility to exacerbation in chronic obstructive pulmonary disease. *N Engl J Med*. Sep 16 2010;363(12):1128-38. doi:10.1056/NEJMoa0909883
47. Dahl M, Vestbo J, Lange P, Bojesen SE, Tybjaerg-Hansen A, Nordestgaard BG. C-reactive protein as a predictor of prognosis in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. Feb 1 2007;175(3):250-5. doi:10.1164/rccm.200605-713OC
48. Engstrom G, Segelstorm N, Ekberg-Aronsson M, Nilsson PM, Lindgarde F, Lofdahl CG. Plasma markers of inflammation and incidence of hospitalisations for COPD: results from a population-based cohort study. *Thorax*. Mar 2009;64(3):211-5. doi:10.1136/thx.2008.102079
49. Groenewegen KH, Postma DS, Hop WC, et al. Increased systemic inflammation is a risk factor for COPD exacerbations. *Chest*. Feb 2008;133(2):350-7. doi:10.1378/chest.07-1342
50. Fibrinogen Studies C, Danesh J, Lewington S, et al. Plasma fibrinogen level and the risk of major cardiovascular diseases and nonvascular mortality: an individual participant meta-analysis. *JAMA*. Oct 12 2005;294(14):1799-809. doi:10.1001/jama.294.14.1799
51. Weathington NM, van Houwelingen AH, Noerager BD, et al. A novel peptide CXCR ligand derived from extracellular matrix degradation during airway inflammation. *Nat Med*. Mar 2006;12(3):317-23. doi:10.1038/nm1361
52. Wells JM, O'Reilly PJ, Szul T, et al. An aberrant leukotriene A4 hydrolase-proline-glycine-proline pathway in the pathogenesis of chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. Jul 1 2014;190(1):51-61. doi:10.1164/rccm.201401-0145OC
53. Snelgrove RJ, Jackson PL, Hardison MT, et al. A critical role for LTA4H in limiting chronic pulmonary neutrophilic inflammation. *Science*. Oct 01 2010;330(6000):90-4. doi:10.1126/science.1190594
54. O'Reilly PJ, Hardison MT, Jackson PL, et al. Neutrophils contain prolyl endopeptidase and generate the chemotactic peptide, PGP, from collagen. *J Neuroimmunol*. Dec 10 2009;217(1-2):51-4. doi:10.1016/j.jneuroim.2009.09.020
55. Wells JM, Jackson PL, Viera L, et al. A Randomized, Placebo-controlled Trial of Roflumilast. Effect on Proline-Glycine-Proline and Neutrophilic Inflammation in Chronic Obstructive Pulmonary Disease. *Am J Respir Crit Care Med*. Oct 15 2015;192(8):934-42. doi:10.1164/rccm.201503-0543OC
56. D'Armiento J, Dalal SS, Okada Y, Berg RA, Chada K. Collagenase expression in the lungs of transgenic mice causes pulmonary emphysema. *Cell*. Dec 11 1992;71(6):955-61. doi:10.1016/0092-8674(92)90391-o
57. Mercer BA, Kolesnikova N, Sonett J, D'Armiento J. Extracellular regulated kinase/mitogen activated protein kinase is up-regulated in pulmonary emphysema and mediates matrix metalloproteinase-1 induction by cigarette smoke. *J Biol Chem*. Apr 23 2004;279(17):17690-6. doi:10.1074/jbc.M313842200
58. Seagrave J, Barr EB, March TH, Nikula KJ. Effects of cigarette smoke exposure and cessation on inflammatory cells and matrix metalloproteinase activity in mice. *Exp Lung Res*. Jan-Feb 2004;30(1):1-15. doi:YJGGWBQLV28W701E [pii]
59. Valenca SS, da Hora K, Castro P, Moraes VG, Carvalho L, Porto LC. Emphysema and metalloelastase expression in mouse lung induced by cigarette smoke. *Toxicol Pathol*. May-Jun 2004;32(3):351-6. doi:10.1080/01926230490431466
VNNGEFT53TDX2MEM [pii]
60. Lee EJ, In KH, Kim JH, et al. Proteomic analysis in lung tissue of smokers and COPD patients. *Chest*. Feb 2009;135(2):344-52. doi:10.1378/chest.08-1583




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

61. Betsuyaku T, Tanino M, Nagai K, Nasuhara Y, Nishimura M, Senior RM. Extracellular matrix metalloproteinase inducer is increased in smokers' bronchoalveolar lavage fluid. *Am J Respir Crit Care Med*. Jul 15 2003;168(2):222-7. doi:10.1164/rccm.200301-103OC 200301-103OC [pii]
62. Finlay GA, O'Driscoll LR, Russell KJ, et al. Matrix metalloproteinase expression and production by alveolar macrophages in emphysema. *Am J Respir Crit Care Med*. Jul 1997;156(1):240-7.
63. Imai K, Dalal SS, Chen ES, et al. Human collagenase (matrix metalloproteinase-1) expression in the lungs of patients with emphysema. *Am J Respir Crit Care Med*. Mar 2001;163(3 Pt 1):786-91. doi:10.1164/ajrccm.163.3.2001073
64. Ohnishi K, Takagi M, Kurokawa Y, Satomi S, Konttinen YT. Matrix metalloproteinase-mediated extracellular matrix protein degradation in human pulmonary emphysema. *Lab Invest*. Sep 1998;78(9):1077-87.
65. Wilson AA, Ying L, Liesa M, et al. Emergence of a stage-dependent human liver disease signature with directed differentiation of alpha-1 antitrypsin-deficient iPS cells. *Stem Cell Reports*. May 12 2015;4(5):873-85. doi:10.1016/j.stemcr.2015.02.021
66. Kaserman JE, Werder RB, Wang F, et al. Human iPSC-hepatocyte modeling of alpha-1 antitrypsin heterozygosity reveals metabolic dysregulation and cellular heterogeneity. *Cell Rep*. Dec 6 2022;41(10):111775. doi:10.1016/j.celrep.2022.111775
67. Tafaleng EN, Chakraborty S, Han B, et al. Induced pluripotent stem cells model personalized variations in liver disease resulting from alpha1-antitrypsin deficiency. *Hepatology*. Jul 2015;62(1):147-57. doi:10.1002/hep.27753
68. Segeritz CP, Rashid ST, de Brito MC, et al. hiPSC hepatocyte model demonstrates the role of unfolded protein response and inflammatory networks in alpha(1)-antitrypsin deficiency. *J Hepatol*. Oct 2018;69(4):851-860. doi:10.1016/j.jhep.2018.05.028
69. Chambers JE, Zubkov N, Kubankova M, et al. Z-alpha(1)-antitrypsin polymers impose molecular filtration in the endoplasmic reticulum after undergoing phase transition to a solid state. *Sci Adv*. Apr 8 2022;8(14):eabm2094. doi:10.1126/sciadv.abm2094
70. Sommer AG, Rozelle SS, Sullivan S, et al. Generation of human induced pluripotent stem cells from peripheral blood using the STEMCCA lentiviral vector. *J Vis Exp*. Oct 31 2012;(68)doi:10.3791/4327
71. Kaserman JE, Hurley K, Dodge M, et al. A Highly Phenotyped Open Access Repository of Alpha-1 Antitrypsin Deficiency Pluripotent Stem Cells. *Stem Cell Reports*. Jul 14 2020;15(1):242-255. doi:10.1016/j.stemcr.2020.06.006
72. Wiesemann GS, Oshins RA, Flagg TO, Brantly ML. Novel SERPINA1 Alleles Identified through a Large Alpha-1 Antitrypsin Deficiency Screening Program and Review of Known Variants. *Chronic Obstr Pulm Dis*. Jan 25 2023;10(1):7-21. doi:10.15326/jcopdf.2022.0321
73. Ma S, Lin YY, Cantor JO, et al. The Effect of Alpha-1 Proteinase Inhibitor on Biomarkers of Elastin Degradation in Alpha-1 Antitrypsin Deficiency: An Analysis of the RAPID/RAPID Extension Trials. *Chronic Obstr Pulm Dis*. Nov 18 2016;4(1):34-44. doi:10.15326/jcopdf.4.1.2016.0156




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## TABLES

**Table 1. Alpha-1 Clinical Resource Centers with geographic location participating in Alpha-1 Biomarkers Consortium**

| A1F Clinical Resource Centers | Location |
|---|---|
| Columbia University Irving Medical Center | New York, New York |
| Medical University of South Carolina | Charleston, South Carolina |
| University of Alabama at Birmingham | Birmingham, Alabama |
| University of North Carolina at Chapel Hill | Chapel Hill, North Carolina |
| Boston University | Boston, Massachusetts |
| University of Utah | Salt Lake City, Utah |
| National Jewish Health | Denver, Colorado |
| University of California-Los Angeles | Los Angeles, California |
| University of Chicago | Chicago, Illinois |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Table 2. Questionnaires to obtain patient reported outcomes**

| Questionnaires | Clinical Characterization |
|---|---|
| Patient Health Questionnaire (PHQ-9), | Health status |
| Alpha-1 Antitrypsin Deficiency Cohort Exposure Questionnaire (Appendix Table 3) | |
| St George's Respiratory Questionnaire (SGRQ) | Pulmonary symptomatology |
| COPD Assessment Test (CAT) | |
| University of California San Diego, Shortness of Breath Questionnaire (SOBQ) | |
| Modified Medical Research Council Dyspnea scale (mMRC) | |
| Breathlessness, Cough, and Sputum Scale (BCSS) | |
| Baseline and Monthly Exacerbation Questionnaires (Appendix Tables 4 and 5) | Exacerbation frequency |
| University of California San Diego Shortness of Breath Questionnaire (SOBQ) | Dyspnea |
| Chronic Liver Disease Questionnaire (CLDQ) | Liver health |
| Alcohol Use Disorder Identification Test (AUDIT-C) | |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## FIGURES

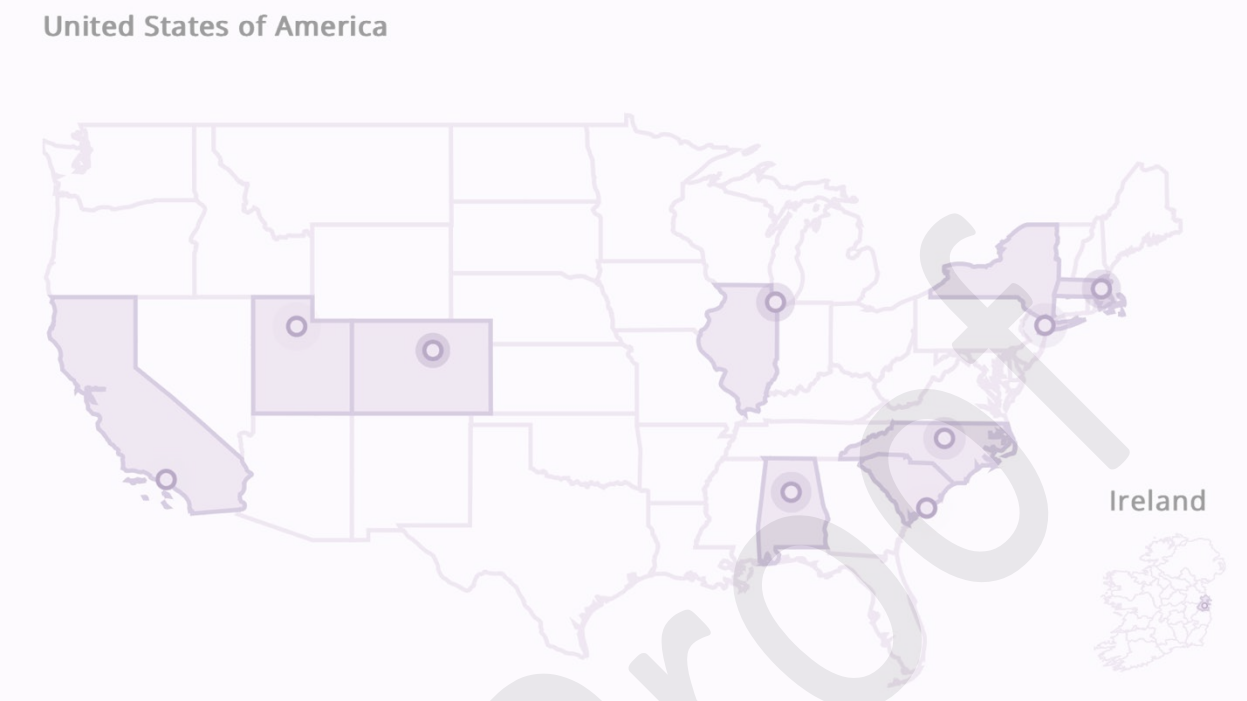


**Figure 1. Alpha-1 Clinical Resource Centers participating in the Alpha-1 Biomarkers Consortium are dispersed around the United States, with a validation cohort in Ireland.**




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## APPENDIX

**Table 1. Primary inclusion and exclusion criteria for enrollment into the Alpha-1 Longitudinal Biomarker Study**

| |
|---|
| ***Inclusion Criteria*** |
| 1. Males and females, age 18 years or older |
| 2. Understand the study procedures, risks, benefits, purpose |
| 3. Able and willing to comply with the study procedures |
| 4. Have PiZZ alpha-1 antitrypsin deficiency |
| 5. Be an existing member of the Alpha-1 Foundation Clinical Cohort (also known as the Alpha-1 Foundation Research Registry) |
| 6. Agree to have the data collected in this study be shared with the Alpha-1 Foundation Research Registry |
| ***Exclusion*** |
| 1. AATD non-PiZZ status, including heterozygous patients |
| 2. Current lung, hematologic, or solid organ malignancy other than skin or cervical Stage 1 cancers within the past 3 years |
| 3. COPD exacerbation or other pulmonary infection within 6 weeks of baseline visit |
| 4. Pregnancy at the time of the screening visit |
| 5. Inability to lie still in a supine position for 15 minutes during CT acquisition |
| 6. Inability to perform quality-controlled lung function testing |
| 7. Allergy to albuterol |
| 8. Currently receiving intravenous or subcutaneous immunoglobulin for any disease state |
| 9. Past or present major surgery on the lungs including pneumonectomy or lobectomy. Wedge resections, past segmentectomy, and pleurodesis surgeries are allowed |
| 10. Previous lung or liver transplantation or currently on the transplant list |
| 11. Current presence of endobronchial coils or valves in the lung |
| 12. Clinically significant bronchiectasis as defined by the investigator. In general, this would exclude patients with chronic infection of the lungs requiring treatment within the past 6 months including non-tuberculous mycobacterial disease, chronic fungal disease, allergic bronchopulmonary aspergillosis, or known colonization of bronchiectasis with pseudomonas or stenotrophomonas species |
| 13. Participation in the active treatment arm of a therapeutic clinical trial at baseline visit unless using one of the Alpha-1 augmentation therapies in alternative doses |
| 14. Patient with Automatic Implantable Cardioverter-Defibrillator and permanent pacemakers |
| 15. Patient receiving biologic immunomodulators that will affect the assessment of the serum biomarkers (as determined by the site PI) |
| 16. Patient with in-dwelling pleural catheters |
| 17. Any condition that in the opinion of the investigator might adversely influence the study outcome |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

## Table 2: *Study schedule of events*

| Procedures | Screen | Visit #1 Baseline | PRO AlphaNet | Phone call | PRO AlphaNet | Phone call | PRO AlphaNet | Visit #2 | PRO AlphaNet | Phone call | PRO AlphaNet | Phone call | PRO AlphaNet | Visit #3 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Months from baseline visit | -1 | 0 | 1-5 | 6 +/- 1 | 7-11 | 12 +/- 1 | 13-17 | 18 +/- 3 | 19-23 | 24 +/- 1 | 25-29 | 30 +/- 1 | 31-35 | 36 +/- 3 |
| **Informed Consent** | X (oral) | X (sign) | | | | | | | | | | | | |
| **Assessment of Eligibility** | X | | | | | | | | | | | | | |
| **Review Medical History** | X | X | | X | | X | | X | | X | | X | | X |
| **Review Medication List** | X | X | | X | | X | | X | | X | | X | | X |
| **Vitals** | | X | | | | | | X | | | | | | X |
| **Blood draw** | | X | | | | | | X | | | | | | X |
| **Dried blood spot card[1]** | | X | | | | | | X | | | | | | X |
| **Urine Pregnancy Test[2]** | | X | | | | | | X | | | | | | X |
| **PFT Spirometry Pre & post** | | X | | | | | | X | | | | | | X |
| **CT scan** | | X | | | | | | X | | | | | | X |
| **Induced Sputum* (varies by site)** | | X | | | | | | X | | | | | | X |
| **Nasal Swab*** | | X | | | | | | X | | | | | | |
| **PROs** | | X | X[3] | X[3] | X[3] | X[3] | X[3] | X[4] | X[3] | X[3] | X[3] | X[3] | X[3] | X[4] |
| **Assessment of Events of Clinical Significance** | | X | | | | | | X | | | | | | X |

Abbreviations: PRO=Patient Reported Outcomes
[1]Dried Blood Spot Card = In addition to study visits, collect DBS cards on onset, 3-day and 7-days post a COPD Exacerbation Event.
[2]Urine Pregnancy Test = Complete Radiation Pregnancy Form when applicable.
[3]X= Monthly AlphaNet COPD Exacerbation questionnaire only.
[4]X= Excludes Exposure Questionnaire (administered at baseline visit only).
*= Optional procedures.


Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Table 3. A1BC Exposure Questionnaire**

| Smoking | |
|---|---|
| Have you ever smoked cigarettes?<br><br>No means less than 20 packs of cigarettes, or 12 ounces of tobacco, or less than 1 cigarette a day for one year at any time in your life. | Yes<br>No |
| 2. How old were you when you first started regular cigarette smoking? | |
| 3. Do you smoke cigarettes (as of one month ago)? | Yes<br>No |
| 4. About how many cigarettes do you smoke per day now? | *Individual cigarettes, not packs* |
| 5. How old were you when you completely stopped smoking? | |
| 6. On average of the entire time you smoked, how many cigarettes did you smoke per day? | |
| 7. Have you ever smoked a pipe regularly?<br><br>YES means more than 12 oz of tobacco in a lifetime | Yes<br>No |
| 8. How old were you when you first started to smoke a pipe regularly? | |
| 9. Do you smoke a pipe (as of one month ago)? | Yes<br>No |
| 10. How many ounces of pipe tobacco do you smoke per day now? | |
| 11. How old were you when you completely stopped smoking a pipe | |
| 12. On average of the entire time you smoked a pipe, how many ounces of tobacco did you smoke per week? | |
| 13. Have you ever smoked cigars regularly?<br>Yes means more than 1 cigar a week for one year at any time in your life | Yes<br>No |
| 14. How old were you when you first started to smoke cigars regularly? | |
| 15. Do you smoke cigars now (as of one month ago)? | Yes<br>No |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

| | |
|---|---|
| 16. How many cigars do you smoke per day? | |
| 17. How old were you when you completely stopped smoking cigars? | |
| 18. On average of the entire time you smoked cigars, how many cigars did you smoke per week? | |
| Vaping | |
| 19. Have you ever used an electronic cigarette or vape product? | Yes<br>No |
| 20. Did your electronic cigarette or vape product contain any of the substances below? | Nicotine<br>Cannabis / marijuana / THC<br>Don't know<br>Other |
| 21. Do you still use e-cigarettes or vape products? | Yes<br>No |
| 22. How often do you use e-cigarettes or vape products? | Everyday<br>Most days<br>4+ days per week<br>1-3 days per week<br>Less than once per week<br>Less than once per month |
| 23. How many years in total have you used electronic cigarettes or vape products? | |
| Second Hand Smoke | |
| 24. Do you currently live in the same household with someone who smokes tobacco products? | Yes<br>No |
| 25. Have you ever lived in the same household with someone who smoked tobacco products? | Yes<br>No |
| 26. Growing up until age 18, were there any adults in your household who smoked at home? | Yes<br>No |
| 27. For how many years in total did you live in the same household with someone else who smoked tobacco products? | |
| 28. Have you been regularly exposed to tobacco smoke in the last 12 months? (Regularly means on most days or nights) | Yes<br>No |
| 29. Do people smoke regularly in the room where you work? | Yes<br>No |
| Occupational | |
| 30. What is your occupation? | |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

| | |
|---|---|
| 31. Does your current job expose you to vapors, gas, dust, or fumes? | Yes<br>Don't know<br>No<br>Not applicable |
| 32. In your longest held job, what kind of work did you do. What was your occupation? | |
| 33. Did your longest job expose you to vapors, gas, dust, or fumes? | Yes<br>Don't know<br>No<br>Not applicable |
| 34. Is an air cleaner/filter used in your residence (stand-alone or central)? | Yes<br>No |
| 35. What type of air filter? | Stand-alone / portable<br>Central<br>Don't know |
| 36. Within the last 12 months have you had wet or damp spots on surfaces inside your home other than in the basement (for example on walls, wall paper, ceilings or carpets)? | Yes<br>No<br>I don’t know |
| 37. Has there ever been any mold or mildew on any surface, other than food, inside the home? | Yes<br>No<br>I don’t know |
| 38. Do you keep a cat inside the house? | Yes<br>No |
| 39. Do you keep a dog inside the house? | Yes<br>No |
| 40. Do you keep any birds inside the house? | Yes<br>No |
| Cleaning Chemicals | |
| 41. Are you responsible for cleaning or washing in your home? | Yes<br>No |
| 42. Have you ever worked as a cleaner? | Yes<br>No |
| 43. How many days per week did you use cleaning products? | Never<br>Less than one day per week<br>1-3 days per week<br>4-7 days per week |
| 44. How many days per week did you use cleaning sprays? | Never<br>Less than one day per week<br>1-3 days per week<br>4-7 days per week |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Table 4. Baseline Exacerbation Questionnaire**

| | |
|---|---|
| 1. Over the past year, how many times have you experienced worsening ("exacerbations" or "flares") of your lung problems? | Every month<br>Every 3 months<br>Every 4 months<br>Every 6 months<br>Once<br>Never |
| 2. Over the past 2 years, have you coughed up sputum/mucus from your lungs on a regular basis for at least three months each year? | Yes<br>No |
| 3. Over the past 12 months, how many times have you experienced the following?<br><br>Please note these are number of events and not days of hospitalization | |
| 3a. Admitted to the hospital? | 0, 1, 2, 3, >3 |
| 3b. Admitted to the intensive care unit? | |
| 3c. Seen in the emergency room? | |
| 3d. Seen by a healthcare practitioner for an unscheduled office visit? | |
| 4. If you were hospitalized over the past year, was the primary reason lung related? | Yes<br>No |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

**Table 5. Monthly Exacerbation Questionnaire**

| | |
|---|---|
| 1. Did you have a new upper respiratory infection, cold, or flu-like illness in the last month? Please note that you should not repeat events reported one month ago. | Yes<br>No |
| 2. How did you manage any lung problems you had this past month?<br><br>Check all that apply | I spoke with my regular primary care physician, nurse practitioner, or physician's assistant.<br>I spoke with my pulmonary specialist.<br>I visited a health care practitioner in his/her office.<br>I went to an emergency room or urgent care center.<br>I treated the problem myself.<br>Not Applicable |
| 3. Over the past month, how many times have you been seen for lung disease and experienced the following?<br><br>Please note these are number of events and not days of hospitalization | |
| 3a. Admitted to the hospital? | 0, 1, 2, 3, >3 |
| 3b. Admitted to the intensive care unit? | |
| 3c. Seen in the emergency room? | |
| 3d. Seen by a healthcare practitioner for an unscheduled office visit? | |
| 4. Have you experienced any worsening of respiratory symptoms (an "exacerbation" or "flare") in the last month?<br>Please note that you should not repeat events reported 1 month ago. | Yes<br>No |
| 5. Have you had any of these symptoms within the past month?<br><br>Check all that apply | a) More shortness of breath<br>b) More cough<br>c) Increased sputum amount<br>d) New wheezing<br>e) Worsening of wheezing<br>f) Sputum changed color<br>g) Fever<br><br>Not Applicable |
| 5.1. Do you have any of these symptoms now?<br><br>Check all that apply | a) More shortness of breath<br>b) More cough<br>c) Increased sputum amount<br>d) New wheezing<br>e) Worsening of wheezing<br>f) Sputum changed color |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603

| | |
|---|---|
| | g) Fever<br><br>Not Applicable |
| 6. Did you have chest imaging (chest X-ray or chest CT) in the past month? | Yes<br>No |
| 7. Did you start oxygen or change your oxygen over the past month? | Yes<br>No |
| If you need to edit a report from one month ago, please report the changes here.<br><br>Examples of an updated report would include that the event from last month had not ended at the time of last report and you would like to add the total duration of the event or that you began new treatments not reported previously. | |




Published online April 24, 2025 https://doi.org/10.15326/jcopdf.2025.0603