Heliyon 10 (2024) e31183

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Review article

Assessing the efficacy of Alpha$_1$-Proteinase inhibitor (human) augmentation therapy for Alpha$_1$-Antitrypsin deficiency – Related emphysema: Challenges and opportunities

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ARTICLE INFO

Keywords:
Alpha$_1$-antitrypsin
Alpha$_1$-antitrypsin deficiency
Alpha$_1$-proteinase inhibitor
Emphysema
Systematic review
Lung density
Spirometry
Exacerbations

ABSTRACT

Clinical benefit of Alpha$_1$-Proteinase Inhibitor (Human) (A_1-PI) products for Alpha$_1$-antitrypsin deficiency (AATD) is uncertain, based on a systematic review of observational studies and randomized controlled trials (RCTs) in AATD of Alpha$_1$-Proteinase Inhibitor (Human) (A_1-PI) products. At the recommended dose, A_1-PI products raise its serum concentration but do not normalize levels. Observational studies suggest A_1-PI might modestly slow progression of airflow limitation in patients with intermediate airflow obstruction, a finding not confirmed by three placebo-controlled RCTs of limited power, which showed non-significant rates of forced expiratory volume in 1 s (FEV_1) change favoring placebo. These RCTs found trends favoring A_1-PI in loss of high-resolution computerized tomographic (HRCT) lung density. While two meta-analyses of HRCT lung density change in RCTs achieved significance favoring A_1-PI arms, clinical benefit remains uncertain. HRCT lung density measurements don't distinguish changes in measured density due to fluid shifts into and out of the lungs and changes in lung inflammation from those due to progressive loss of alveolar mass. A meta-analysis of RCTs found exacerbations significantly increased in A_1-PI groups compared to placebo. No RCTs have shown favorable effects of A_1-PI on mortality, FEV_1, 6-min walking distance, quality of life, change in diffusion capacity of carbon monoxide (DL_{CO}), or exacerbation frequency. A fourth RCT comparing two dose regimens of A_1-PI is underway. RCTs have not provided evidence of clinical benefit in terms of how patients feel, function, or survive. Results have implications for the design of future clinical trials of A_1-PI and potentially other products targeting AATD-associated emphysema.

1. Introduction

The objective of this article is to provide a systematic review of observational studies and randomized controlled trials that have attempted to evaluate the efficacy of Alpha$_1$-Proteinase Inhibitor (Human) augmentation Therapy for Alpha$_1$-Antitrypsin Deficiency – Related Emphysema (AATD-RE), and to discuss their implications for the design of future studies. Alpha$_1$-antitrypsin deficiency (AATD) is an autosomal codominant genetic disorder caused by mutations in the gene coding for alpha$_1$-proteinase inhibitor (A_1-PI)/alpha$_1$-antitrypsin (AAT). A_1-PI, among other activities, inhibits lung neutrophil elastase (NE), and helps protect lung parenchyma from proteolytic destruction [1–6]. The most common manifestations of severe AATD are emphysema and liver disease [1,7]. AATD phenotypes, corresponding serum levels and comparative risk of emphysema are listed in Table 1. Normal (Pi*MM) serum AAT ranges

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https://doi.org/10.1016/j.heliyon.2024.e31183
Received 2 October 2023; Received in revised form 4 May 2024; Accepted 12 May 2024
Available online 23 May 2024

from ~20 to 53 μM [8]. Severe AATD is defined by having a serum antigenic A_1-PI level <11 μM. Approximately two-thirds of never-smokers and ~90 % of ever-smokers with severe AATD eventually develop emphysema [7,9].

AATD-RE is characterized by dyspnea, reduced exercise tolerance/capacity, airflow limitation, and acute exacerbations of COPD. In patients with AATD-RE, progressive loss of alveoli can lead to respiratory failure and death or the need for lung transplantation to sustain life.

1.1. Augmentation therapy for AATD-related lung disease

The only therapeutic agents approved for AATD-RE are $Alpha_1$-Proteinase Inhibitor (A_1-PI) (Human) products, manufactured from pools of human plasma [17–20]. Therapy with A_1-PI is referred to as "augmentation therapy," because it augments the blood and lung levels of A_1-PI [2,21]. At the currently recommended weekly dose, the steady-state trough level does not achieve normal levels [22].

The US Food and Drug Administration (FDA) licensed the first A_1-PI product in 1987 based on a demonstration of A_1-PI-induced changes in biochemical biomarker endpoints, prior to the formal introduction in 1992 of an accelerated approval pathway based on a surrogate endpoint. Manufacturers of US-licensed A_1-PI augmentation therapy products have entered into postmarketing commitments (PMCs) [23] to conduct randomized, controlled clinical trials (RCTs) to evaluate potential long-term benefit using clinically-meaningful endpoints [22,24,25]. Data suggest that the current A_1-PI dose regimen may be suboptimal [26–31]. Manufacturers have agreed to study a higher dose to ascertain if it is associated with [improved] efficacy and safety. No PMCs of A_1-PI products have been completely fulfilled, and only one is currently in progress. PMC trials may be informative regarding the relationships between A_1-PI serum levels and emphysema progression, plus exacerbation risk in patients receiving A_1-PI products at currently recommended and higher doses.

The first A_1-PI approval was based on demonstrating (a) a rise from pre-therapy baseline and maintenance of serum A_1-PI trough level to > 11 mM and (b) a rise from baseline in lung epithelial lining fluid (ELF) A_1-PI concentration calculated from broncho-alveolar lavage (BAL) fluid sampling. The history of the hypothetical therapeutic target serum level of 11 mM A_1-PI is discussed in online Supplement A to this article.

Clinical benefit has yet to be demonstrated for A_1-PI products. Their effects, if any, on symptoms, function/exercise tolerance, the incidence, severity, and/or duration of exacerbations, and/or the progression of emphysema or airways obstruction have not been demonstrated in adequate and well-controlled clinical trial(s) [17,32–35]. The current FDA-approved indication for A_1-PI products is listed in Table S1.

1.2. Pharmacokinetics of A_1-PI

Population pharmacokinetic parameters for antigenic serum A_1-PI during chronic dosing with Zemaira brandA_1-PI 60 mg/kg IV weekly, as reported by Tortorici et al., included clearance of 45.2 mL/day, volume of distribution of 10.0 L, half-life of 6.8 days, C_{avg} of 20.9 μM (90 % CI 14.8–26.7), C_{max} of 28.5 μM (90 % CI 13.3–44.7) and C_{trough} of 16.2 μM (90 % CI 11.1–22.6) [36].

There is substantial inter-subject variability in A_1-PI trough levels in persons receiving augmentation therapy at the recommended weekly dose of 60 mg/kg [22]. Published literature and data contained in FDA-approved package inserts of A_1-PI products indicate that most patients receiving regular weekly 60 mg/kg infusions of licensed A_1-PI products do not achieve normal serum levels of A_1-PI and may continue to experience progression of COPD and exacerbations [17,32–34,18,37–39]. Additional information on the pharmacokinetics of A_1-PI is provided in online supplement B.

1.3. Natural history studies of AATD

Several authors have examined the relationship between baseline FEV_1 and subsequent rate of change of FEV_1 in patients with AATD not receiving A_1-PI [40–43]. The fastest deterioration of airflow obstruction tends to occur in patients with intermediate degrees of airflow limitation at baseline, such as those with baseline FEV_1 approximately 30–65 % of predicted [40–43]. Among usual, AAT-replete COPD patients, those with very severe airflow limitation at baseline as a group have slower subsequent decline in FEV_1

Table 1
Normal and Selected AATD Phenotypes and their Serum A_1-PI Levels [1,8,10–13].

Phenotype	Risk of Emphysema	Serum Antigenic A_1-PI Levels (μM)
PI *ZZ	Very High	2–7
PI*SZ	Intermediate[a]	9–23
PI*MZ	Low but greater than normal[b]	17–33
PI*MM (Normal)	Normal (Low)	20–53

The nomenclature for AATD phenotypes relates to the results of isoelectric focusing of patients' serum. The Z, S, and M allelic variants each have a different isoelectric point, giving rise to bands of different mobility. "PI" stands for proteinase inhibitor.

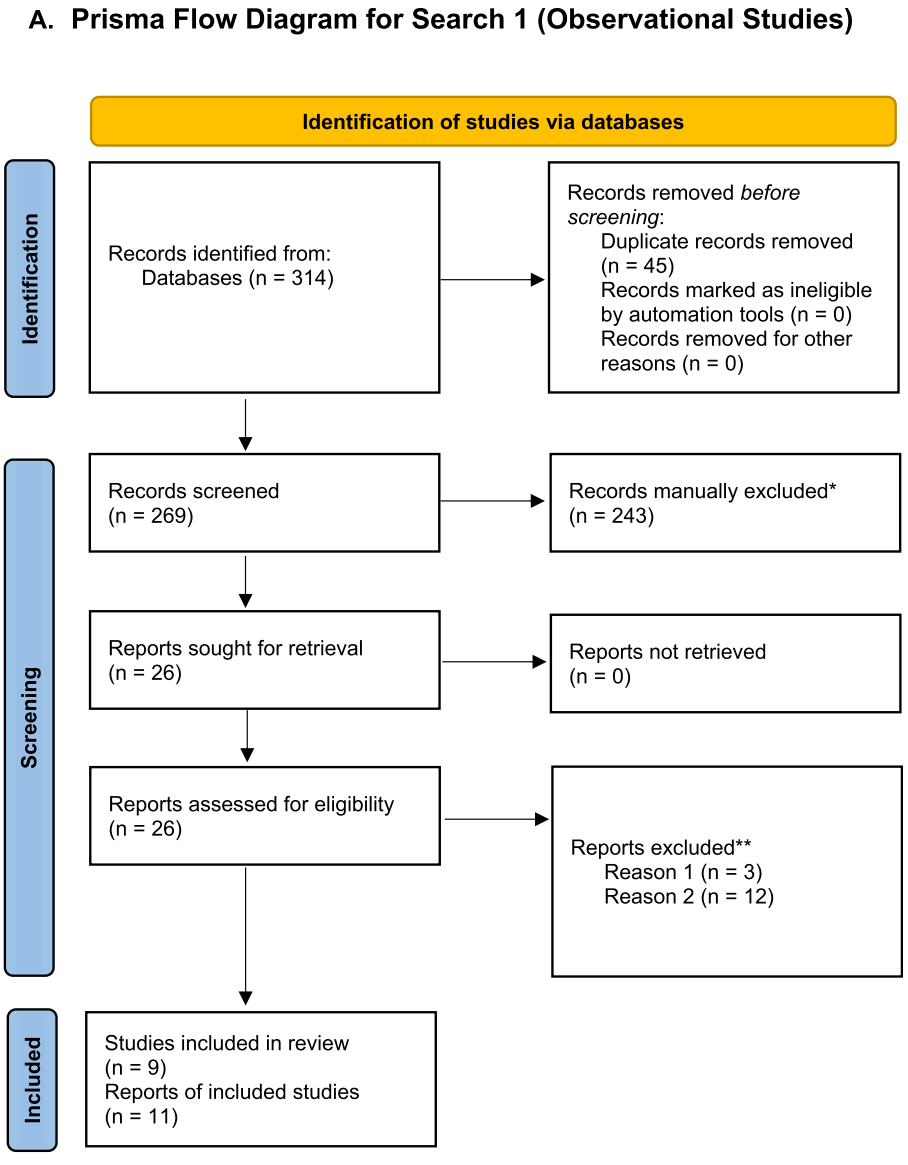
[a] The odds ratio for risk of emphysema for PI*SZ has been estimated in a meta-analysis to be 3.26-fold compared to that of normal PI*MM individuals (95 % CI 1.24–8.57) [14].

[b] The risk of hospitalization for COPD in Denmark for PI*MZ individuals was found to be 2.2-fold greater than that of PI*MM individuals [8,15]. The rate of decline in DL_{CO} among PI*MZ individuals also has been found to be more rapid than for PI*MM individuals [16].

than patients with less severe FEV_1 impairment [44,45]. These data may be relevant when considering possible enrichment strategies when crafting subject inclusion criteria in trials using FEV_1 as a primary or co-primary endpoint.

2. Methods

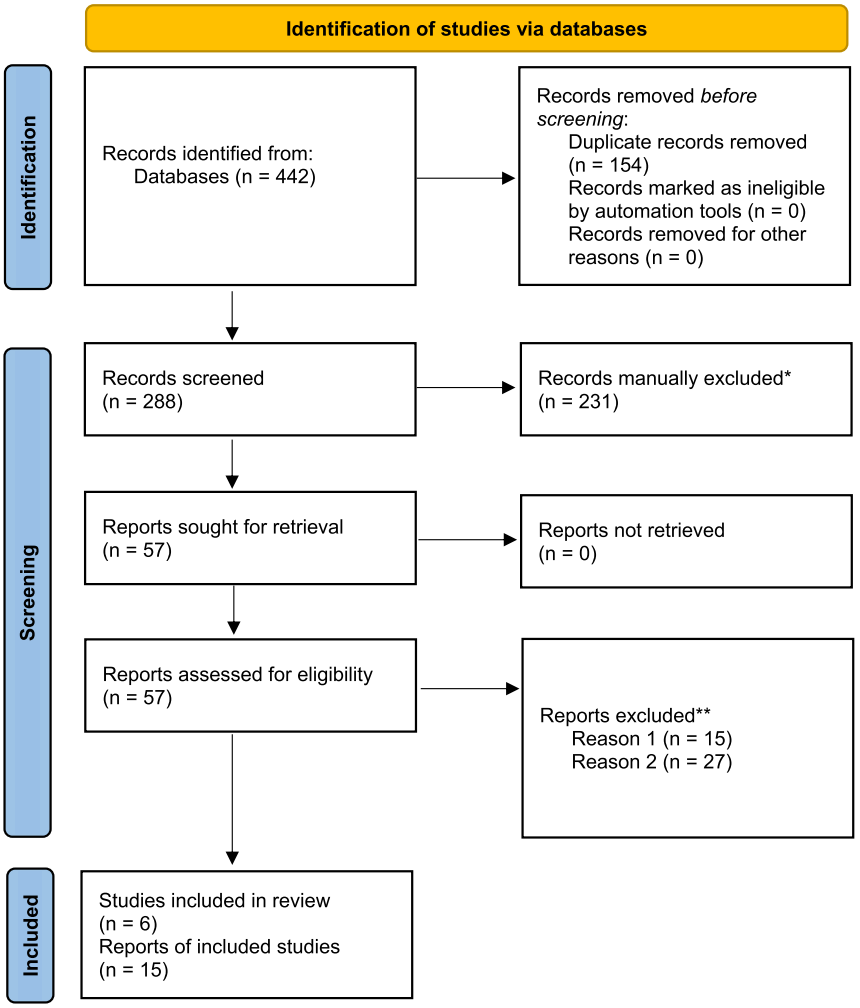
Drs. L. Ross Pierce and Gavin Imperato, with the assistance of Ms. Gwendolun Halford, conducted literature searches in 2022 comprising each of the bullets below using PubMed, Embase, Cochrane Central, and ClinicalTrials.gov and performed a systematic review using the PRISMA 2020 Checklist [46] as follows.



*No automation tools were used for excluding records deemed irrelevant to the screening topic.
**Reason 1 for excluding reports: Study randomized, not observational.
Reason 2 for excluding reports: Otherwise not meeting criteria.

Fig. 1. Prisma Flow Diagram Describing Process for Selection of Studies Included in this Review A. Prisma Flow Diagram for Search 1 (Observational Studies).

B. Prisma Flow Diagram for Search 2 (Randomized Controlled Trials)



*No automation tools were used for excluding records deemed irrelevant to the screening topic.
**Reason 1 for excluding reports: Study randomized, but not placebo – or dose – controlled.
Reason 2 for excluding reports: Study not randomized.

Fig. 1. (*continued*).

- Observational Studies comparing AATD patients who received vs. those who did not receive Alpha$_1$-Proteinase Inhibitor augmentation therapy from the year 1987 to present, English language only. Studies comprising fewer than 50 subjects or over an observation period of <1 year were excluded.
- Randomized controlled trials involving patients with AATD and lung disease/COPD/emphysema and Alpha$_1$-Proteinase Inhibitor augmentation therapy from 1987 to present, all languages. Studies comparing different manufacturers' A$_1$-PI products and not including a placebo group or dose comparison were excluded.

Papers from the searches were screened and relevant articles reviewed independently by Drs. Pierce and Imperato and studies that met the above criteria were included in the review/discussion. All results compatible with each output domain were sought. Relevant data from each included report was directly transcribed by Dr. Pierce into this review. Included studies were assessed for sources of bias, including baseline imbalances in covariates that could affect outcome measures, the potential for regression to the mean, potential differences in stardard of care between comparison groups, and other potential confounding factors. The range of the magnitudes of potential bias among the studies comprising this review is large. The review was not registered.

High Level Specific literature search Strategy Terms and Boolean operators were as follows:

Observational Studies Search:

[efficacy OR effect OR effectiveness OR lung function decline OR lung density decline OR diffusion capacity of carbon monoxide decline OR exercise capacity decline OR FEV_1 decline OR mortality OR survival OR [longitudinal] follow-up] AND [Alpha1-Proteinase Inhibitor OR Alpha1-Antitrypsin OR augmentation therapy] AND alpha1-antitrypsin deficiency AND [COPD OR emphysema or lung disease] NOT cross-sectional, NOT animal.

Randomized Controlled Trials Search:

Randomized Controlled Trial AND [Alpha1-Proteinase Inhibitor (Human) OR Alpha1-Antitrypsin] AND Alpha1-Antitrypsin Deficiency.

The complete list of seach terms and Boolean Operators for the above searches is given in Supplement E.

3. Results

We identified nine observational studies meeting protocol/search criteria comparing AATD patients who received vs. those who did not receive $Alpha_1$-Proteinase Inhibitor augmentation therapy.

We identified six randomized placebo – and/or dose – comparison controlled trials involving patients with AATD and lung disease/COPD/emphysema and $Alpha_1$-Proteinase Inhibitor augmentation therapy. Three randomized placebo – controlled trials were completed and one is ongoing. Two trials used a randomized crossover design to compare two dosage regimens.

Prisma flow diagrams describing the study selection process for searches 1 (observational studies) and 2 (RCTs) are presented in Fig. 1.

4. Non-interventional epidemiology studies of A_1-PI augmentation therapy

4.1. NHLBI registry study

The NHLBI epidemiologic registry study explored the possible effects of augmentation therapy with Prolastin brand A_1-PI over a period of 3.5–7 years on mortality and FEV_1 rate of change in severe AATD [39,47]. The pre-specified primary endpoints of the NHLBI Registry study were comparisons between participants who received augmentation therapy with A_1-PI versus those who did not receive A_1-PI of 1) mortality among registry participants with baseline FEV_1 below 30 %, and (2) FEV_1 rate of change among registry participants whose baseline FEV_1 was greater than 30 % [47]. *The publication reporting the results of the NHLBI registry does not provide the results of either the mortality or the FEV_1 primary endpoints for the prespecified primary analysis subgroups of participants with baseline FEV_1 below or above 30 % of predicted, respectively*. Rather, the results of *post-hoc* analyses using different FEV_1 strata are reported. Inspection of the results for the reported FEV_1 strata suggest that both prespecified primary endpoints likely failed [39].

As shown in Table 2, multivariate survival analysis of NHLBI Registry participants who received versus those who did not receive A_1-PI therapy did not demonstrate a significant survival advantage with A_1PI among patients whose FEV_1 was below 35 % of predicted. However, multivariate survival analysis of the entire registry cohort did suggest a survival advantage with augmentation therapy (RR 0.64, 95 % CI 0.43–0.94, p = 0.02 (unadjusted for multiple *post-hoc* comparisons)). This result was driven by the participants with an intermediate degree of airway obstruction (FEV_1 35 %–49 %) [39].

As shown in Table 3, multivariate analysis of FEV_1 rate of change in the entire NHLBI Registry cohort of participants who received versus those who did not receive A_1-PI therapy did not find a statistically significant advantage with augmentation therapy (difference in rates of change in FEV_1 = 4.2 mL/year, 95 % CI -5.7 – 14.2, p = 0.40, unadjusted). Nor did multivariate analysis of FEV_1 rate of change demonstrate a statistically significant difference with A1-PI among patients whose FEV_1 was 35 %–79 % of predicted (difference = 13.6 mL/year, 95 % CI -4.1 – 31.1, p = 0.13, unadjusted). However, multivariate *post-hoc* analysis of the difference in rates of change in FEV_1 among patients whose FEV_1 was 35 %–49 % in participants who had received versus those who had not received A_1-PI therapy suggested the possibility of benefit with augmentation therapy (difference = 26.8 mL/year, 95 % CI 2.8–50.9, p = 0.03,

Table 2
Multivariate survival analysis[a] showing relative risk of death by mean FEV_1 strata in NHLBI registry prospective epidemiologic study [39].

Mean FEV_1 % Predicted[b]	Number of Subjects	Relative Risk of Death[c] (Therapy vs. Control)	RR 95 % CI[c]	P Value[d]
<35 %	482	0.83	0.52, 1.33	0.44
35–49 %	217	0.21	0.09, 0.50	<0.001
>50 %	349	0.75	0.22, 2.56	0.64
Overall	1048	0.64	0.43, 0.94	0.02

[a] Multivariate Survival Analysis included receipt of a lung transplant treated as a time-varying covariate. The baseline or first available post-bronchodilator measurement of FEV1% predicted was used as a covariate in the survival models, using the following staging strata: FEV1% predicted <35 % [Stage III], 35–49 % [Stage II], 50–79 % [Stage I], and ⩾ 80 % [Normal]). Only subjects who were successfully contacted 6 or more months following enrollment were included in the survival analysis.

[b] Mean FEV_1 predicted over the duration of each subjects' trial participation. Analyses using baseline FEV_1 to define strata yielded similar results, according to the publication.

[c] It is possible that these point estimates for relative risk of death and corresponding confidence intervals overstate, or understate, the true mortality effect (if any) of augmentation therapy at the currently recommended dose because of differences in baseline factors (unaccounted for) between augmented and non-augmented patients other than their augmentation therapy status.

[d] Reported p values are not adjusted for multiple comparisons, so they may be considered to lack validity.

Table 3
Multivariate analysis[a] of FEV_1 decline: Mean FEV_1 decline (ml/yr) by FEV_1% predicted and augmentation therapy status in NHLBI registry prospective epidemiologic study [39].

Mean FEV_1 % Predicted[b]	Difference[c] in FEV_1 Slopes: Receiving vs. Not Receiving A_1-PI[c]	95 % CI[d]	P Value[e]
< 35 %	2.6	−111.3, 16.5	0.71
35–49 %	26.8	2.8, 50.9	0.03
30–64 %	18	2, 34	0.03
50–79 %	7.5	−14.7, 29.6	0.50
≥80 %	−23.8	−50.9, 3.3	0.09
35–79 %	13.6	−4.1, 31.1	0.13
Overall	4.2	−5.7, 14.2	0.40

[a] Covariates in the multivariate analysis linear mixed effects model included mean FEV_1% predicted, calculated from all available visits, bronchodilator responsiveness, coded as whether the subject ever versus never had a bronchodilator response (defined as postbronchodilator increase in FEV_1 of at least 200 mL and 12 % over the prebronchodilator value at any visit, and the cumulative time (since enrollment) for which each subject had received augmentation therapy at each follow-up visit (as a time-dependent covariate).

[b] Mean FEV_1 predicted over the duration of each subjects' trial participation. Analyses using baseline FEV_1 to define strata yielded similar results, according to the publication. For example, the difference in mean FEV_1 rates of decline with and without augmentation therapy for the subgroup with *baseline* FEV_1 35–49 % of predicted was 22 mL/yr, $p = 0.04$).

[c] A positive difference in slopes implies a slower rate of decline for subjects receiving A1-PI augmentation therapy compared with those not receiving augmentation therapy.

[d] It is possible that these point estimates and corresponding confidence intervals overstate, or even understate, the true effect (if any) of augmentation therapy at the currently recommended dose on FEV_1 rate of change because of differences in baseline factors (unaccounted for) between augmented and non-augmented patients other than their augmentation therapy status.

[e] Reported p values are not adjusted for multiple comparisons, so they may be considered to lack validity.

unadjusted for multiple *post-hoc* comparisons) [39]. A similar finding was obtained in an analysis of registry patients whose average FEV_1 was 30–64 % of predicted (difference in rates of change in $FEV_1 = 18$ mL/year, 95 % CI 2–34, $p = 0.03$, unadjusted) [39]. *As post-hoc analyses, these results in subgroups with intermediate degrees of airflow limitation should be considered for hypothesis generation only;*

Table 4
Differences in FEV_1 slopes between Patients Receiving and Not Receiving A_1-PI in Non-Randomized Observational Studies.

Study, Year of Publication/ Reference No./Design	Overall n (A_1-PI)/n (no A_1-PI)/Duration (A_1-PI)/ Duration (no A_1-PI)[a] (years)	Overall Study Δ (FEV_1) mL/year± SD or % predicted	Overall 95 % CI (mL/ year) or p Value[b] for FEV_1 Difference	Subgroup FEV_1 30–65 % Δ (FEV_1) mL/ year ± SD	Subgroup FEV_1 30–65 % 95 % CI (mL/year) or p Value[b]
NHLBI Registry[c] 1998/Ref 28/ Prospective	581/317/3.5–7 yrs	4.2 mL/year	−4.9–13.3	13.6	−3.3–30.5
Seersholm 1995/Ref 31/ Retrospective	198/97/3.2 years/5.8 years (means)	22 mL/year	p = 0.02	21	p = 0.04
Wencker 2001 /Ref 38/Retrospective	96/4.0 years/4.2 years (means)	14.6 ± 61.4 mL/year	p = 0.019	11.6 ± 48.8	p = 0.066
Tonelli 2009/Ref 29 Unstated (Presumably Retrospective)	124/40 + 26[d] /3.5 years (mean, combined)	43.9 mL/year	p = 0.046	54.0	p = 0.07
Schouten 2021/39/ Retrospective	128/246 /8.6/8.6 years (mean)	−0.085 % predicted	−1.144 to 0.717 % predicted p = 0.71	NR[e]	NR
National German Registry 2017/40 Unstated (Presumably Retrospective)	85/15 /4.9 years (mean, combined)	33.8 mL/year	NS[f]	NR	NR
Spanish National Database 2018/41/Retrospective	77/45 /8 years (mean, combined)	NR	p = 0.675	NR	NR

[a] Durations in years are provided for studies in which different cohorts of patients receiving and not receiving A_1-PI augmentation therapy are compared or the same cohort is compared during periods in which the patients were not receiving, then receiving augmentation therapy.

[b] 95 % confidence intervals (CI) for the difference in FEV_1 rate of change between patients receiving and not receiving augmentation therapy (slope of those receiving minus slope of those not receiving A_1-PI) are provided where reported; otherwise, nominal p values as reported, which have not been corrected for multiple comparisons are given. The latter are not considered valid, due to lack of correction for multiple comparisons.

[c] Values for the NHLBI Registry Study are taken from Ref 26 (Chapman meta-analysis) which reports the difference in FEV_1 slopes for the stratum of patients with *baseline* $FEV_1$from 30–65 % of predicted.

[d] Results are reported for the combined original cohort of 124 patients on A_1-PI and 40 patients not on A_1-PI, plus an additional 26 patients who were not included in the original analysis because their smoking status was initially missing (but later located). The breakdown of these additional 26 patients by treatment status was not provided in the publication.

[e] NR = not reported.

[f] NS = not statistically significant ($p \geq 0.05$).

the pre-specified co-primary efficacy endpoint for FEV_1 was unlikely to have been met among participants whose baseline FEV_1 was >30 %, and augmentation therapy did not appear to have any effect in the overall study population.

Among NHLBI Registry patients who received augmentation therapy, only 51 % initially received the currently recommended 60 mg/kg weekly dosage regimen. This dropped to 33 % at last report. Many subjects received a double dose every two weeks or a quadruple dose every four weeks, resulting in higher peak and lower trough A_1-PI levels [39].

As a non-randomized trial, the investigators could not exclude the possibility that any observed differences may have been due to factors other than augmentation therapy. They noted that "a definitive conclusion will require a randomized controlled trial." [39].

Re-analysis of the mortality data from the NHLBI Registry study, published by Rahaghi et al. [48], corrected database errors and included additional mortality status ascertainment. The original analysis plan described in the 1998 NHLBI Registry publication was altered in the Rahaghi analysis, in that patients who did not have contact with the study six months or more following enrollment were included in the Rahaghi analysis, unlike in the 1998 publication. The Rahaghi re-analysis states that the original statistical plan to exclude these participants "will always stand as the primary analysis of mortality since it was *pre-hoc*." The re-analysis found significantly lower mortality among subjects ever on augmentation therapy compared to those never on A_1-PI therapy within each decile of baseline FEV_1% predicted between 10 % and 60 % [48].

FEV_1 results of the NHLBI Registry Study are compared with those of smaller observational studies comparing patients receiving vs. not receiving A_1-PI in Table 4.

4.2. Seersholm 1995 study

A retrospective epidemiological study compared the rates of decline in FEV_1 between 198 ex-smoker PI*ZZ patients in Germany who received Prolastin brand A_1-PI over a mean of 3.2 years and 97 ex-smoker Danish PI*ZZ patients who did not receive A_1-PI and were followed over a mean of 5.8 years [42]. Patients who had at least two FEV_1 measurements obtained at least one year apart were included in the analysis. Attempts were made to adjust for differences between cohorts in the proportions of subjects of each sex, follow-up time, and initial FEV_1. Overall, patients in Germany who received Prolastin had a significantly slower rate of decline in FEV_1 as compared to patients in Denmark who did not receive A_1-PI (means of 53 ± 38 vs. 75 ± 60 mL/year, respectively, difference 22 mL/year, $p = 0.02$). This difference was largely accounted for by patients whose baseline FEV_1 was between 30 and 65 % of predicted, in whom the mean rates of decline in FEV_1 were 62 ± 25 vs. 83 ± 49 mL/year (difference 21 mL/year, $p = 0.04$, unadjusted) for the augmented versus unaugmented patients, respectively. Differences in rates of FEV_1 decline between patients in Germany and Denmark whose FEV_1 baseline values were <30 or >65 % of predicted were not significant ($p = 0.6$ and 0.7, respectively). German patients whose baseline FEV_1 was >65 % of predicted were required to demonstrate a rate of decline in FEV_1 of at least 120 mL/year, but this requirement was not listed for the Danish cohort. Thus, these data are subject to regression to the mean for the German cohort, which could have biased the results. Differences in standard-of-care between the two cohorts in different countries may have also confounded the results.

4.3. Wencker 2001 study

Wencker et al. published a retrospective observational study comparing the rates of change of FEV_1 before and after weekly administration of Prolastin brand A_1-PI at 60 mg/kg in 96 severe AATD subjects in Germany (62 males and 34 females) of mean age 44.3 years and baseline $FEV_1 < 65$ % or, if baseline $FEV_1 > 65$ %, who had a documented rate of decline in FEV_1 of >120 mL/yr [49]. The study included overlapping data of German subjects who received A_1-PI in the Seersholm study [42] and was limited by its retrospective nature, lack of randomization, lack of a concurrent control group, lack of parallel design, lack of blinding, and expected regression to the mean in the subgroup of patients with baseline $FEV_1 > 65$ % of predicted who were selected for analysis based on having a pre-augmentation therapy observed rate change in FEV_1 of >120 mL/yr. Subjects were followed for a mean of 50.7 (SD 34.3) months prior to augmentation therapy and for 35.7 (SD 23.3) months during augmentation therapy.

Table 5
FEV_1 slopes (mL/year) among patients prior to and during A_1-PI augmentation therapy according to baseline FEV_1 strata in the Wencker 2001 study [49].

Initial FEV_1% Predicted[a]	n	Mean Slope FEV_1 (mL/year) Prior to A_1-PI Augmentation	Mean Slope FEV_1 (mL/year) During A_1-PI Augmentation	Difference Between Augmented and Non-Augmented Periods (SD)	P Value[c]
<30 %	25	−15.3	−19.0	−3.7 (48.6)	NS
30–65 %	60	−49.3	−37.8	11.6 (48.8)	0.066
Overall[b]	96[b]	−49.2[b]	−34.3[b]	**14.9**[b] (61.4)	0.019[b]

[a] Initial FEV_1% Predicted refers to the FEV_1% predicted at the start of the period of observation without A_1-PI augmentation therapy. Results for the baseline FEV_1 stratum FEV_1% predicted >65 % are not shown because they are subject to regression to the mean due to the additional requirement for this stratum only that patients be fast decliners (rate of FEV_1 decline of >120 mL/yr).

[b] FDA considers the overall results unreliable due to inclusion of data for 11 subjects with baseline FEV_1% predicted who were also required to be fast decliners (rate of FEV_1 decline of >120 mL/yr) and whose results were therefore subject to regression to the mean.

[c] Reported p values are not adjusted for multiple comparisons, so they may be considered to lack validity.

Wencker et al. observed a mean slowing of 14.9 (SD 61.4) mL/yr in FEV_1 rate of change after initiation of augmentation therapy in the overall cohort (p = 0.019, unadjusted for multiplicity, Table 5). For subjects (n = 60) with an initial FEV_1 30–65 % of predicted at the start of unaugmented observation, the declining slope of FEV_1 was reduced (less negative) by a mean of 11.6 (SD 48.8) mL/yr (p = 0.066, unadjusted) during A_1-PI administration [49].

The above results use the initial FEV_1 to stratify subjects; different results were obtained when FEV_1 at the time of initiation of augmentation therapy was used for stratification [49]. Because of limitations in study design, inconsistent results within FEV_1 strata, depending on whether the initial or final FEV_1 during the unaugmented observation period was used for defining subgroups, and potential regression to the mean to have biased results, this study should be interpreted with great caution. Nevertheless, this study is included in a published meta-analysis of the effects of augmentation therapy in AATD-related COPD [41].

4.4. Tonelli 2009 study

Tonelli et al. published an observational study involving 164 Pi*ZZ AATD patients enrolled in the Alpha-1 Foundation DNA and Tissue Bank study [40]. Change in FEV_1 (ΔFEV_1) was defined as the initial FEV_1 minus the FEV_1 obtained by random effects model. The change in FEV_1 was compared between patients who were receiving A_1-PI augmentation therapy at the time of enrollment (n = 124) and those who were not (n = 40) overall and for subgroups based on initial FEV_1 % of predicted. An analysis of mortality at 5 years used logistic regression with age, gender, baseline FEV_1, presence of COPD and smoking status as covariates.

The mean follow-up time was 41.7 (SEM ± 2.6; range: 6 to 268) months. The augmented patients were older on average, more commonly had COPD with lower baseline FEV_1 (43 % (SEM ± 2 %) versus 77 % (SEM ± 5 %)), and had a higher number of individuals who required oxygen therapy. The study was underpowered to detect any difference in survival between the groups [40].

As shown in Table 6, when adjusted by age at baseline, sex, smoking status and baseline FEV_1 % of predicted, the difference in mean overall change in FEV_1 between these groups was 47.6 mL/year ($p = 0.05$) favoring augmentation therapy. Similar results were obtained when 26 patients in whom the smoking history was initially missing but later located were added to the analysis [40].

Patients with FEV_1 >65 % had higher rates of FEV_1 decline if they received augmentation therapy (ΔFEV_1 augmented: −108.7 ± 17.3 mL/year versus ΔFEV_1 non-augmented: −29.2 ± 15.3 mL/year; for an overall difference of −79.5 mL/year, $P < 0.001$). A trend towards a beneficial effect of A1-PI was observed in the group with an initial FEV_1 30 %–65 % (difference in $\Delta FEV_{1:}$ 54 mL/year, $P = 0.07$) [40].

4.5. Additional studies of FEV_1 rate of change among patients receiving vs. not receiving A_1-PI

The Shouten 2021 retrospective study [49], the National German Registry study [50], and the Spanish National Database study [51] involved 128/246, 85/15, and 77/45 PI*ZZ patients who received/did not receive A_1-PI, respectively. All three studies failed to demonstrate a significant difference in rate of change of FEV_1 between patients who received and did not receive A_1-PI. Further details of these studies are provided in online Supplement C.

4.6. Sandhaus 2020 study

A retrospective analysis of two prospectively followed PI*ZZ (“or worse” genotype) AATD patient cohorts with emphysema in two countries differing in access to A_1.PI augmentation therapy was undertaken [52]. The U.S. cohort comprised patients receiving A_1-PI and followed in AlphaNet’s Disease Management and Prevention program with monthly telephone interviews. Mortality data was derived from the US National Death Index and direct notification by family. The U.K. control group comprised A_1-PI naïve patients from the AATD U.K. Registry who were prospectively followed with annual medical review and physiological measurements. Mortality data for this cohort were derived from a central NHS database, general practitioners or by direct notification by family. Quality of Life was assessed in both groups annually with the St. George’s Respiratory Questionnaire (SGRQ). Subjects (n = 655) were matched based on age, sex, baseline year, and smoking history. Overall 10-year survival rate was longer in the A_1-PI (U.S.) group compared to matched controls from the U.K., with rates of 80 % (95 % CI 76.7 to 83.4) and 68.5 % (95 % CI 64.2 to 73.1), respectively (p < 0.001). Lung transplantation within 5 years of baseline assessment occurred in 13.3 % (95 % CI 7.54 to 18.7) of the U.S. treatment group and 58.5 % (95 % CI 40.3 to 71.2) of the U.K. control group. Annual worsening of SGRQ total was on average 1.3 points worse/year in control group patients compared to those receiving A_1-PI (95 % CI 0.41 to 2.19, p = 0.004). Features of the AlphaNet Program other than A_1-PI augmentation therapy and other differences in care between the U.S. and U.K. may have affected the reported outcomes of this

Table 6
ΔFEV_1 (mL/year) among patients receiving and not receiving A_1-PI augmentation therapy according to baseline FEV_1 strata in the Tonelli study [40].

Initial FEV_1% Predicted	Augmented		Non-Augmented		p Value[a]
	n	ΔFEV_1 (mL/year)	n	ΔFEV_1 (mL/year)	
<30 %	**30**	**0.9**	**3**	**20.1**	**0.59**
30–65 %	**79**	**2.08**	**10**	**−51.9**	**0.07**
>65 %	**15**	**−108.7**	**27**	**−29.2**	**0.0006**
Overall	**124**	**10.6**	**40**	**−37.0**	**0.05**

[a] Reported nominal p values are unadjusted for multiple comparisons, so they may be considered to lack validity.

retrospective cross-country comparison [52].

4.7. Chapman meta-analysis

Chapman et al. published a meta-analysis of four observational studies and one randomized, placebo-controlled trial of A_1-PI augmentation therapy [41]. This meta-analysis of 1509 patients should be interpreted with caution due to the predominant inclusion of observational studies subject to bias from unaccounted covariates. This meta-analysis includes the Wencker 2001 retrospective within-patient comparison [49] and the Seersholm study [42] which compared FEV_1 rates of change between patients in Germany receiving augmentation and patients in Denmark not receiving augmentation therapy. Both studies were subject to regression to the mean among German patients with baseline $FEV_1 > 65$ % at baseline, who were required to be fast decliners. As shown in Table 7 below, among all patients included in the meta-analysis, the mean decline in FEV_1 was slower by 13.4 mL/year (95 % CI 1.5–25.3 mL/year) among patients receiving A1-PI compared to those who were not. This overall effect was said to reflect predominantly the results of patients with baseline FEV_1 of 35–65 % of predicted, whose absolute difference in average rates of FEV_1 decline was 17.9 mL/yr (95 % CI 9.6–26.1 mL/yr). Differences with and without augmentation therapy among patients whose baseline FEV_1 were <30 % or >65 % were not significant. The statistical significance in the overall pooled cohort is lost after removing either the Wencker or Seersholm observational studies, but the statistical significance is retained for the baseline FEV_1 30–65 % of predicted subgroup after removal of the Wencker study. The point estimate for the single randomized, placebo-controlled trial included in the meta-analysis shows a small, non-significant trend in rate of change of FEV_1 favoring the placebo group [32].

4.8. Barros-Tizon 2012 exacerbation study

Barros-Tizon et al. conducted a retrospective, multicenter, observational study of AATD patients in Spain whose records were available for 18 months prior to and 18 months while on A_1-PI augmentation therapy with either Trypsone (Grifols) or Prolastin (Talecris) [53]. Of 127 patients included in the study, only 6 % received A_1-PI weekly, with 17 % receiving A_1-PI biweekly and 76 % receiving A_1-PI every 3 weeks. (Every- 2- and 3- week regimens are associated with higher peak serum levels of A_1-PI). The study's primary objective was to examine whether A_1-PI augmentation therapy was associated with a reduction of both the number of exacerbations and the percentage of patients experiencing exacerbations, defined using GOLD criteria [54] and requiring a change in medical regimen.

Overall, mean exacerbations per patient were reduced from 1.2 ± 1.6 prior to augmentation therapy to 1.0 ± 2.2 ($p < 0.01$) during the 18 months following initiation of A_1-PI treatment. Among patients who had at least one exacerbation during the 18 months prior to initiation of augmentation therapy, mean exacerbations per patient were reduced from 2.0 ± 1.6 to 1.4 ± 2.7 ($p < 0.01$) during augmentation therapy [53].

Augmentation therapy was associated with a reduction in the percentage of patients who experienced one or more exacerbations from 59.1 % to 44.1 % in the overall population ($p < 0.005$). The difference between the two periods in the percentage of patients who experienced one or more *severe* exacerbations was not statistically significant (46.7 % vs. 55.4 %, respectively). Multivariate analysis with imputation of missing values for lung function and laboratory variables suggested that unaugmented patients would be expected to be at 1.4- to 4.2-fold more risk of exacerbation than patients receiving A_1-PI [53]. Courses of systemic antibiotics were significantly reduced during the period of augmentation therapy compared with the prior period (means: 13 vs. 28, respectively, $p < 0.05$) [53].

Limitations of the study include its retrospective nature, lack of randomization and lack of concurrent control, heterogeneity of A_1-PI dosing regimen and agent, and lack of masking which may have contributed to an expectation of benefit and influenced subjects' reporting of exacerbations.

5. Randomized controlled trials of augmentation therapy

The results of three published double-blind, placebo-controlled RCTs of augmentation therapy, each testing a different A_1-PI product, are shown in Table 8.

Table 7
FEV_1 slope (mL/year) among patients receiving and not receiving A_1-PI augmentation therapy according to baseline FEV_1 strata (Chapman meta-analysis) [41].

Initial FEV_1% Predicted	Augmented		Non-Augmented		Slope Difference* (95 % CI)
	n	Mean Slope FEV_1 (mL/year)	n	Mean Slope FEV_1 (mL/year)	
<30 %	454	−30.6	180	−30.9	1.8 (−7, 10.5)
30–65 %	398	−50.8	263	−67.9	17.9 (9.6, 26.1)
>65 %	43	−92.1	175	−97.2	3.5 (−49, 55.9)
Overall	924	−48.0	681	−59.4	13.4 (1.5, 25.3)

[a]Slope differences are as reported in the publication and are noted to not exactly correspond to the differences in mean slopes between augmented and non-augmented subjects as reported in this table and the publication.

Table 8
Randomized, double-blind, placebo-controlled trials of A_1-PI in severe AATD-Associated emphysema.[a].

Reference	A_1PI Product Manufacturer	Number of Subjects Randomized	Adjusted HRCT Lung Density Outcome	FEV_1 Slope Outcome	Exacer-bations Outcome
20 Dirksen A et al., 1999	Laboratoire Biologique du Fractionnement (LBF)	56	PD15[c] p = 0.07	p = 0.20[b] favored placebo	NM[d]
21 Dirksen A et al., 2009 [EXACTLE]	Grifols	77	PD15[b] p = 0.068	NS[c] favored placebo	NS[c] favored placebo
22,23,24 Chapman KR et al., 2015 [RAPID]	CSL Behring	180	PD15[b] p = 0.06	NS[c] favored placebo	NS[c] favored placebo

[a] Lung-Volume-Adjusted High Resolution Computerized Tomographic (HRCT) Lung Density, FEV_1 Slope, and total Exacerbation Incidence Outcomes are given as unadjusted 2-sided p values for the difference between A_1-PI and placebo arms.
[b] Primary Efficacy Endpoint, 2-sided p values.
[c] Secondary Efficacy Endpoint, 2-sided p value.
[d] NM = Not Measured.

5.1. Dirksen 1999 RCT

This RCT randomized 56 Danish and Dutch ex-smokers with PI*ZZ AATD and FEV_1 30 %–80 % of predicted to LBF (French) A_1-PI 250 mg/kg or albumin 625 mg/kg IV every four weeks for at least 3 years [32]. The primary endpoint, FEV_1 rate of decline, was non-significantly slower in the placebo group compared to the A_1-PI group (p = 0.20). The rate of decline in lung density by lung-volume-adjusted CT (assessed by the PD15, the 15th percentile of lung density voxels), a secondary endpoint, was numerically slower in the A_1-PI group (mean 1.5 ± 0.41 g/L/year) compared with the placebo group (mean 2.6 ± 0.41 g/L/year), with a difference of 1.07 g/L/year between treatment groups (p = 0.07, unadjusted). Exacerbation rates and mortality were not reported [32].

5.2. Dirksen 2009 [EXACTLE] RCT

This exploratory RCT, sponsored by Prolastin's manufacturer, randomized 77 PI*ZZ AATD Danish, Swedish, and U.K. subjects with emphysema 1:1 to Prolastin brand A_1-PI or placebo at the weekly IV dose of 60 mg/kg for a treatment period of 2–2.5 years [33]. The primary analysis of the primary endpoint, lung-volume-adjusted rate of change in lung density by CT, was numerically slower in the A_1-PI group (mean −1.384 g/L/year) compared with the placebo group (mean −2.241 g/L/year), with an estimated treatment difference of 0.857 g/L/year (p = 0.068). As in the 1999 Dirksen study, the mean rate of decline in FEV_1 was numerically faster in the A_1-PI group than in the placebo group, but this difference was not statistically significant. The annual incidence of exacerbations trended higher in the A_1-PI group (2.55 ± 2.14) than in the placebo group (2.19 ± 1.33), (difference NS), but, in a *post-hoc* analysis the percentage of severe exacerbations among those with known severity was lower in the A_1-PI group (13/194, 6.7 %) compared to that in the placebo group (21/155, 13.5 %, p value for difference 0.013). A statistically significant weak correlation was observed between changes in lung-volume-adjusted lung density and FEV_1 (Correlation coefficient 0.216, p = 0.007) [33].

5.3. Chapman 2015 (RAPID) RCT

This RCT, sponsored by Zemaira's manufacturer, randomized 180 U S. and EU non-smoker and ex-smoker subjects with AATD and FEV_1 values 35–70 % of predicted 1:1 to Zemaira brand A_1-PI or placebo at 60 mg/kg IV weekly for 2 years [34]. The RAPID trial had a substantial imbalance in baseline lung density that favored the A_1-PI treatment arm. Baseline lung density was 3.2 g/L greater in the placebo group than in the A_1-PI group. Data from the placebo subjects in the trial (and to a lesser extent in the A_1-PI arm subjects) have shown a faster rate of decline of adjusted lung density among subjects with higher baseline lung density values [55]. Eighteen subjects in the placebo group and 9 in the A_1-PI group discontinued the study prematurely; discontinuations due to an adverse event or death numbered 7 versus 2, respectively, in placebo and A_1-PI arms). The primary analysis of the primary endpoint, lung-volume-adjusted rate of change in lung density by CT using scan data obtained both at the two inspiratory states, total lung capacity (TLC) and functional residual capacity (FRC), was numerically slower in the A_1-PI group (mean −1.45 g/L/yr) compared with the placebo group (mean −2.19 g/L/yr), with a treatment difference of 0.62 g/L/yr (2-sided p = 0.06) [34]. Statistically combining CT scan data from scans taken at both TLC and FRC had been proposed by the manufacturer and accepted by FDA. Only one of multiple prespecified supplementary analyses of adjusted lung density rate of change (using only CT data obtained at TLC) achieved statistical significance (without adjustment for multiple analyses). Some have suggested that this particular supplementary analysis should be relied upon instead of the prespecified primary analysis of the primary efficacy endpoint for concluding the product was effective, because the primary analysis of the primary endpoint combined CT data taken at both FRC and TLC and because *unadjusted* lung density measurements made at FRC are more variable than those made at TLC. Such a *post-hoc* approach for drawing inferences regarding efficacy is not statistically justified. Adjusting lung density by lung volume substantially reduces variability [56–59], and the combined FRC/TLC adjusted lung density measurements for the prespecified primary endpoint were slightly *less variable* than those for the supplementary analysis using only adjusted lung density measurements taken at TLC. While changes in air trapping can theoretically

affect CT lung density measurements taken at FRC more than TLC, correction of lung density values for lung volume accounts for changes in lung volume due to air trapping. The RAPID trial's pre-specified primary endpoint combining adjusted TLC and FRC lung density data was an appropriate and acceptable primary analysis approach (which, nevertheless, did not achieve statistical significance).

Subgroup analyses of the between-treatment-group mean annual rates of change in adjusted lung density at TLC from the intent-to-treat population of the RAPID trial indicate a several-fold difference between men and women [60]. While the overall difference between Zemaira and placebo arms in rates of decline in adjusted lung density was 0.74 g/L per year, the difference among females (n = 79) was 1.45 g/L per year. Higher between-treatment-group differences were also seen for subjects with higher baseline A_1-PI serum levels and subjects with BMI >30 kg/m^2 (n = 21, difference in P15 rate of change = 2.21 g/L per year). While these subgroup analyses are exploratory, they merit further study. The observation of a greater between-treatment group difference among obese subjects may relate to expected higher blood A_1-PI levels in subjects dosed on a mg/kg basis who have a lower percent lean body mass.

5.4. RAPID trial extension

In the RAPID trial extension phase non-U.S. subjects were offered open-label Zemaira brand A_1-PI for two years [35]. One hundred forty subjects participated: seventy-six who had been randomized to Zemaira (early-start group) and 64 who had been randomized to placebo (late-start group) during the prior two-year RAPID trial. One hundred thirty-one subjects completed the RAPID trial extension with 121 having complete lung density by HRCT data. In a *post-hoc* subgroup analysis of late-start group subjects who had HRCT lung density data, these subjects experienced a slowing in their mean rate of change of lung volume-adjusted lung density by HRCT at TLC from −2.26 g/L per year during their two years of placebo treatment in the RAPID trial to −1.26 g/L per year during their two years of open-label Zemaira treatment in the RAPID extension [35].

Using a single mixed-effects regression model with combined data from the RAPID trial and its open-label extension, an inflection point in the mean rates of lung density decline was seen when the late-start group began treatment with open-label Zemaira, with a reduction in the mean rate of lung density by HRCT loss during the trial extension of 0.52 g/L per year (95 % CI 0.22–0.83 g/L per year). A slight acceleration in the mean rate of decline in adjusted lung density by HRCT at TLC in early start subjects after entering the RAPID trial extension was not significant (change from −1.51 g/L per year during the RAPID trial to −1.63 g/L per year during the RAPID trial extension) [35]. The less negative slope in the mean rate of lung density decline in the late-start group subjects after starting open-label Zemaira is suggestive of a treatment effect of Zemaira; however, the results are tempered by the post-hoc nature of the analysis, the lack of a concurrent control group continuing placebo treatment during the RAPID trial extension, lack of blinding during the extension, and the modest absolute difference in the delayed start subgroup in the mean rates of lung density decline between the RAPID trial and its extension, which is of uncertain clinical significance.

5.5. Non-HRCT outcomes in the rapid trial

As in the two Dirksen studies, the mean rate of decline in FEV_1 in the RAPID trial was numerically slightly faster in the A_1-PI group compared to the placebo group, but this difference was not statistically significant (NS) [34]. The annual incidence of serious exacerbations was also numerically greater in the A_1-PI group than in the placebo group in RAPID, (differences NS). Non-statistically significant trends in changes in exercise capacity by shuttle walk test, incidence of subjects with dyspnea reported as an adverse event, hospitalizations for exacerbations, duration of hospitalizations, and change in diffusion capacity of carbon monoxide (DL_{CO}) all favored the placebo group [34]. Combining data from the double-blind RAPID trial and its open label extension, a statistically significant weak correlation was observed between rate of change in adjusted lung density by CT and change in FEV_1 (r = 0.286) and change in FEV_1 % predicted (r = 0.0338) [35]. This finding is difficult to interpret, given that in the double-blind RAPID trial the non-statistically significant trend in change in FEV_1 numerically favored the placebo group [34].

6. Exacerbation outcomes in randomized, placebo-controlled trials of A_1-PI augmentation therapy

A meta-analysis of two completed randomized, double-blind, placebo-controlled trials [33,34] of IV A_1-PI products found a statistically significant increase in exacerbation frequency in the A_1-PI arms compared to placebo (difference: 0.29 exacerbations per subject-year, 95 % CI 0.04–0.54, P = 0.02) [61]. As noted, in a post-hoc analysis performed in one of these two trials reporting exacerbation severity data, the frequency of severe exacerbations in the subset of A_1-PI arm exacerbation episodes for which severity data were available was lower compared to the corresponding subgroup of the placebo arm [33]. In the other trial in which serious exacerbation data were analyzed (RAPID), a non-statistically significant numerical increase in exposure-adjusted serious exacerbation rate was observed in the A_1-PI arm (0.146 serious exacerbations per subject-yr) compared to that in the placebo arm (0.115 serious exacerbations per subject year, ratio A_1-PI: placebo 1.256, 95 % CI 0.46–3.45) [17]. In another *post-hoc* analysis of a subset of subjects completing a 2-year open-label extension phase of the RAPID trial, subjects in both randomization arms experienced further increases (60 % and ~100 % in late and early start groups, respectively) in exposure-adjusted serious exacerbation rates after switching from placebo or double-blind A_1-PI to open-label A_1-PI [17]. Only the increase in serious exacerbation rate in the early start group originally randomized to double-blind A_1-PI and then switched to open-label A_1-PI during the extension phase achieved nominal statistical significance (95 % CI for the ratio of rates during the two periods: 1.21–3.67) [17]. The 95 % CI for the ratio of serious exacerbation rates observed in the placebo arm of the RAPID trial after vs. before crossover to open-label Zemaira treatment during the extension study was 0.80–3.03) [17].

7. Meta-analyses of randomized, placebo-controlled trials of A_1-PI augmentation therapy

Gøtzsche and Johansen of the Cochrane Group published a meta-analysis of the three randomized, double-blind, placebo-controlled trials of A_1-PI augmentation therapy described above [62]. The planned primary efficacy endpoint of the meta-analysis, mortality, wasn't possible, as mortality was only reported for the RAPID trial. Combining data from all three trials, the secondary outcome variable in the meta-analysis, lung density by HRCT, deteriorated significantly less in the augmentation therapy group than in the placebo group, (mean difference 0.86 g/L per year, 95 % CI 0.31 to 1.42; p = 0.002). Similar results were obtained by Edgar et al. in a separate meta-analysis of the same three trials (mean difference 0.79 g/L per year, 95 % CI 0.29–1.29; p = 0.002) [61]. Across the three trials, FEV_1 deteriorated more in the treatment group than in the placebo group, but there was no significant between-group difference (SMD -0.19, 95 % CI -0.42 to 0.05; p = 0.12 in Gøtzsche and Johansen [62] difference in mean FEV_1 % predicted −0.56, 95 % CI -1.41 – 0.09, p = 0.20 in Edgar et al. [61]). On average, carbon monoxide diffusion capacity deteriorated more in the treatment group than in the placebo group, but there was no significant between-group difference (SMD -0.11, 95 % CI -0.35 to 0.12; p = 0.34 in Gøtzsche and Johansen [62]; SMD -0.11, 95 % CI -0.33 to 0.11; p = 0.34 in Edgar et al. [61]).

8. Randomized dose comparison studies of A_1-PI augmentation therapy

8.1. Campos 2013 [SPARK] study

The 2013 SPARK study conducted by Campos et al. [63], compared the safety and pharmacokinetics of Prolastin-C brand A_1-PI at weekly 120 mg/kg and 60 mg/kg intravenous doses in 30 subjects with severe AATD who were randomized to receive one of the two weekly doses for 8 weeks before being crossed over to the alternate dose for an additional 8 weeks. Mean trough antigenic A_1-PI levels were 27.7 μM following 120 mg/kg weekly Prolastin-C and 17.3 μM following the 60 mg/kg weekly dose. Twenty-three subjects on the 60 mg/kg dose reported 69 treatment-emergent adverse events (TEAEs) and 18 subjects on the 120 mg/kg dose reported 43 TEAEs. No subjects reported serious AEs and no subject withdrew due to an AE. Seven subjects (23 %) experienced a total of 9 exacerbations while on the 60 mg/kg dose and 5 subjects reported a total of 6 exacerbations while on the 120 mg dose [63].

8.2. Campos 2019 study

The study published in 2019 by Campos et al. [31] was an open-label, 3-period crossover study of ten subjects (nine ZZ and one SZ genotype) with severe AATD-related emphysema who received four weeks of standard 60 mg/kg weekly A_1-PI (Zemaira) therapy, followed by 4 weeks of 120 mg/kg weekly A_1-PI therapy, followed by a return to 4 weeks of 60 mg/kg weekly A_1-PI administration. Bronchoalveolar lavage (BAL) and plasma samples were obtained for A_1-PI levels and other biomarkers at baseline and at the end of each 4-week study period while clinically stable. The study enrolled subjects who had been receiving A_1-PI at the standard weekly intravenous dose of 60 mg/kg, as well as at least one long-acting bronchodilator and an inhaled corticosteroid [31].

The primary outcome was change in pulmonary Inflammatory markers measured in BAL fluid. Secondary endpoints were improvement in serum/plasma inflammatory markers and elastin degradation markers in serum and BAL fluid [31].

Trough antigenic AAT levels increased from 16. 7 ± 2.3 μ M following standard A_1-PI dosing at the end of the first period to 27.2 ± 5.0 μ M by the end of the 2nd (120 mg/kg weekly "double dose") period, and returned to 16.0 ± 2.6 μ M at the end of the 3rd period after resumption of the standard dose. Trough AAT levels were within the normal range on the 120 mg/kg weekly regimen. BAL fluid neutrophil elastase decreased from 3.32 + 0.86 nM on standard dose A_1-PI to 1.61 ± 0.29 nM (p = 0.008) on the double dose regimen, while BAL fluid cathepsin G, another lung protease, fell from 247.0 ± 60.4 nM to 56 ± 15.3 nM (p = 0.005). Mean trough neutrophil elastase and cathepsin G levels were slightly but not statistically significantly higher at the end of period 3 resumption of the standard dose A_1-PI regimen, suggesting a carry-over effect from the prior period of double dose administration. Plasma levels of Aa-Val^{360}, a biomarker of neutrophil elastase activity, fell significantly from 10.15 ± 1.19 nM after SD therapy to 7.89 ± 0.57 nM on double dose therapy (p = 0.016), with a subsequent rise a month after single dose therapy was resumed. The markers of elastin degradation, trough BAL fluid desmosine and isodesmosine, fell from 3.74 ± 3.7 pg/mg protein on SD therapy to 0.65 ± 0.34 pg/mg protein after double dose A_1-PI administration (p = 0.050). The lower BAL fluid levels were maintained after resumption of the standard dose, again suggesting a carry-over effect. Plasma desmosine/isodesmosine levels were unchanged on double dose A_1-PI therapy [31].

Levels of several, but not all markers of airway inflammation (cytokines, chemokines, and growth factors) also fell by the end of period 2 double dosing, including tumor necrosis factor α (TNFα), interleukin-17 (IL-17), granulocyte-macrophage colony-stimulating factor (GM-CSF), macrophage migration inhibitor factor (MIF), macrophage inflammatory protein 1a chemokine (C–C motif) ligand 3 (MIP1aCCL3), macrophage colony-stimulating factor (M-CSF), interferon g (IFNg), IL-2, IL-12p40, IL-10, IL-3, Eotaxin/CCL11, IL-9, IL-4, and basic fibroblast growth factor (bFGF). At the end of period 3, some of these markers returned to levels observed at the end of the initial standard dose period 1, but others remained depressed, suggesting a possible carry-over effect. One month of double-dose therapy appeared to downregulate cytokines that affect the Janus kinase signal transducer and activator of transcription proteins (JAK-STAT), T cell receptor signaling pathways, and those that affect cytokines in macrophage migration, eosinophil recruitment, humoral and adaptive immunity, neutrophil activation, and cachexia [31].

These results suggest that patients receiving standard dose A_1-PI augmentation therapy can have ongoing protease activity, elastin degradation, and lung inflammation that is ameliorated after one month of "double dosing" with 120 mg/kg weekly intravenous dosing. The observation of a possible carry-over effect for several inflammatory biomarkers and protease activities suggests there could be a role for studying alternating periods of higher and standard doses in patients with severe AATD – related lung disease [31].

The authors concluded that "increasing AAT levels into the normal range may provide additional clinical benefits and have a more robust impact on clinical outcomes in subjects with AAT deficiency requiring augmentation therapy." [31].

8.3. SPARTA trial

The **S**tudy of **P**rol**A**stin-C **R**andomized **T**herapy with **A**lpha-1 augmentation (SPARTA, NCT01983241) [25], is a randomized, double-masked, parallel, placebo-controlled, dose comparison phase 3 trial in which 339 subjects with severe AATD-RE are being randomized 1:1:1 to receive placebo, 60 mg/kg weekly Prolastin-C brand A_1-PI, or 120 mg/kg weekly Prolastin C for three years. The primary endpoint is the rate of change of lung volume-adjusted whole lung HRCT lung density (PD15) at TLC. Secondary endpoints include severe exacerbations of COPD, rate of change of PD15 of the basal lung region, changes from baseline in FEV_1, change from baseline in Saint George's Respiratory Questionnaire, and change from baseline in EuroQol (Quality of Life)-5 Demension-5 Level. The study began November 2013 and was projected to be completed June 2025 (total duration 12 years). The trial has enrolled subjects with severe AATD having allelic combinations of ZZ, SZ, Z(null), (null)(null), S(null), or "at-risk" alleles with screening visit $FEV_1 > 30$ % and <80 % of predicted and FEV_1/FVC, <70 %, $DL_{CO} < 60$ % predicted OR evidence of pulmonary emphysema on CT scan. Exclusion criteria include receipt of more than one month of A_1-PI augmentation therapy within the past six months, history of an exacerbation within five weeks prior to screening, and history of lung or liver transplant [25].

9. Discussion

Findings from epidemiologic studies in AATD-RE suggest that the rate of decline in FEV_1 may be greatest among unaugmented patients with intermediate degrees of airflow limitation and that the possible benefit of A_1-PI augmentation therapy, if any, might be concentrated in this subgroup [39–43,64]. A post-hoc analysis of the largest prospective observational study of A_1-PI use in AATD [39] provides weak evidence that it may slow the rate of decline in FEV_1 in the subgroup of subjects with FEV_1 30–65 % of predicted. While the study authors reported statistical significance for FEV_1 change in favor of augmentation therapy among subjects with FEV_1 35–49 % and 30–65 %, these results lack statistical significance after adjustment for multiple comparisons and should be considered for hypothesis generation only. The slowing in mean decline in FEV_1 in this subgroup receiving A_1-PI was less than half that required to slow the decline to that attributable only to aging in AAT-replete individuals. There is no evidence that the study met its prespecified primary endpoints for either FEV_1 change or mortality, although an updated *post-hoc* analysis by different authors [48] suggested that mortality might be improved among patients with baseline FEV_1% predicted between 10 % and 60 %, with the largest mortality reduction in patients whose FEV_1 was 35–49 %. The authors cautioned that this mortality result may have been confounded by differences in important baseline variables that were not captured/accounted for in the analyses.

The data from several of the smaller observational studies included in this review were taken from national registries in AATD whose main objectives were not the evaluation of the efficacy of A1-PI augmentation therapy per se. For this reason, in addition to bias resulting from physician and patient decisions regarding whether to undergo augmentation therapy and possible imbalances in measured and unmeasured covariates, the results of these observational studies results must be interpreted with caution.

Notwithstanding their limitations and variability of results and study power, taken as a whole, epidemiology studies of A_1-PI at the weekly IV dose of 60 mg/kg vs. untreated patients provide weak evidence suggesting potential benefit in slowing FEV_1 rate of decline in subjects with FEV_1 baseline values from ~30 % to ~65 %, with little apparent benefit in subjects with milder or more severe airflow obstruction at baseline [39–43,64]. However, this finding has not been confirmed by three underpowered, randomized, placebo-controlled trials [32–34].

An 11.5 % absolute reduction in mortality with A_1-PI was observed in a *retrospective* cross-country analysis by Sandhaus et al. [52], as well as a substantial reduction in lung transplantation and a slight reduction in the rate of deterioration in SGRQ. However, differences in care between the U.S. and U.K. may have affected the reported outcomes of this retrospective cross-country comparison.

The retrospective crossover study by Barros-Tizon et al. [53] found a 15 % absolute reduction during augmentation therapy in the proportion of patients who had exacerbations, but the A_1-PI regimen consisted of weekly dosing in only 6 % of patients and there was no concurrent control group or masking. The higher peak serum A_1-PI level associated with the higher doses given at less frequent than weekly intervals in that study might have yielded different results from what may occur with standard weekly dosing at 60 mg/kg.

While none of the three randomized, placebo-controlled trials of three different A_1-PI products achieved statistical significance for their prespecified primary efficacy endpoints (FEV_1 in the first trial and PD15 lung-volume-adjusted lung density by HRCT in the second and third trials), two published meta-analyses of the PD15 results of these studies achieved statistical significance favoring the A_1-PI groups [61,62]. Because the first RCT [32] used a markedly different dosage schedule from the other trials (monthly vs. weekly, resulting in a higher peak and lower trough serum A1-PI concentration, I question the appropriateness of including the first study in these meta-analyses. The RAPID trial [34], had a substantial imbalance in baseline lung density that appears to have favored the A_1-PI treatment arm. Baseline lung density was not included in the model used for analysis of the primary endpoint in the RAPID trial [34], so the degree to which the reported p values would have changed by its inclusion in the model remains unknown. I encourage the manufacturer to make public the raw data from the trial, so that the effect of this baseline imbalance could be independently assessed.

Other potential confounders in the RCTs of A_1-PI products include differential dropout rates, potential differences over the course of the trials between randomization groups in subjects experiencing fluid shifts into and out of the lungs and/or changes in inflammatory cell burden as from exacerbations which would affect measured lung density independent of any changes in lung parenchymal mass, and differences in the proportion of men/women between treatment arms, if the underlying rate of lung density loss differs between sexes as is well-established in the case of FEV_1. An analysis of the effect of baseline demographics on HRCT outcomes in the

RAPID trial suggests a large sex difference in the between-treatment-group differences in rate of decline of lung volume-adjusted lung density (PD15) [60].

The between-group differences in mean rates of change in PD15 lung-volume adjusted lung density over the course of the RCTs are of uncertain clinical significance. The heterogeneity of published studies and limited longitudinal data have made it difficult to determine the magnitude of change in adjusted lung density by HRCT rate of change that would constitute the minimum clinically important difference (MCID) [65]. Nevertheless, a MCID for HRCT lung density of −2.89 g/L has been proposed by one group [66,67]. The mean differences observed between A_1-PI and placebo groups in adjusted lung density in RAPID were less than half of this estimated MCID as proposed by Crossley et al., and less than one-fifth of the within-subject standard-deviation of 3.42 g/L per yr. for repeated lung density measurements reported in the ECLIPSE study [45].

There are no data from *interventional* clinical trials to indicate that a difference in HRCT lung density is predictive of any clinical benefit to patients. To date, no RCT has shown both a statistically significant result for HRCT lung density changes and a statistically significant positive result for any other non-HRCT-related clinical endpoint [32–34]. In the largest RCT comparing A_1-PI with placebo (RAPID) [34], trends in non-HRCT lung density secondary clinical endpoints consistently favored placebo, whereas trends in HRCT lung density favored the augmentation therapy group. Although one randomized controlled trial [33] and one subgroup analysis of another randomized controlled trial [34] combined with its open-label extension study [35] have shown weak correlations between change in HRCT lung density and change in FEV_1, in both trials point estimates for change in FEV_1 numerically favored the placebo groups rather than the A_1-PI therapy groups.

For a biomarker such as HRCT lung density to be considered a validated surrogate endpoint suitable for use as a single primary endpoint in Phase 3 clinical trials intended to support marketing, the endpoint should be "supported by a clear mechanistic rationale and clinical data providing strong evidence that an effect on the surrogate endpoint predicts a specific clinical benefit [as measured by how patients feel, function or survive] … Generally, required evidence includes a combination of a clear mechanistic rationale and in most cases, data from multiple randomized clinical trials showing that the effect on the surrogate endpoint predicts the effect on the clinical outcome of primary interest." [68].

As for non-lung-density efficacy endpoints, none of the RCTs provided evidence that the A_1-PI augmentation therapy products at the doses administered produced significant favorable effects on mortality, exacerbation rate, FEV_1 rate of change, DL_{CO} rate of change, exercise capacity, or symptoms such as dyspnea. A meta-analysis of RCTs two and three [33,34] which measured exacerbation frequency found a significant excess of exacerbations in the A_1-PI arms compared to placebo [61]. A post-hoc analysis of the exposure-adjusted serious exacerbation rate found that the rate doubled during open-label A_1-PI administration years 3 and 4 of the RAPID trial extension compared to the rate in the active A_1-PI arm during double-blind years 1 and 2 of the RAPID trial (95 % CI for the ratio rates during the two periods: 1.21–3.67) [17]. The serious exacerbation rate observed in the placebo arm of the RAPID trial increased by 60 % after crossover to open-label Zemaira treatment during the extension study (95 % CI for the ratio of the rates: 0.80–3.03) [17].

After the licensure of the first A_1-PI product, FDA determined that there is no adequate scientific basis to conclude that the historic 11 μM target for serum A_1-PI is an appropriate therapeutic target for augmentation therapy [24]. For this reason, following a unanimous recommendation of the FDA Blood Products Advisor Committee (BPAC) in 2009 [24], FDA has requested manufacturers of new plasma-derived A_1-PI products to conduct pre-licensure a randomized, concurrently controlled, double-blind clinical trial using clinically-meaningful endpoint(s) that reflect how patients feel, function, or survive, or the forced expiratory volume in 1 s (FEV_1) rate of change as the primary efficacy endpoint to provide substantial evidence of effectiveness [28].

10. Conclusion/recommendations

Studies evaluating the clinical efficacy of A_1-PI augmentation therapy in emphysema associated with AATD, taken as a whole, are inconclusive. Further validation of lung volume-adjusted lung density by HRCT in interventional trials should be undertaken before this marker is used as a sole primary endpoint of future clinical trials. This would include demonstrating, in the context of one or more adequate and well-controlled randomized interventional trials, that differences in the rates of change in adjusted lung density predict concordant differences in changes in one or more endpoints directly reflective of how patients feel, function, or survive [68,69] and acceptance of a reasonably-established minimum clinically important difference. For future trials, the author encourages use of adaptive trial designs [70] and enrichment strategies [71,72,] to enroll subjects who are most likely to benefit from augmentation therapy. For example, limiting enrollment to fast decliners in FEV_1 % predicted would improve the likelihood that future studies may be able to determine whether higher doses of A_1-PI than currently recommended are clinically beneficial to patients [72,73].

Funding source

This work was supported by Midnight Sun Technologies, LLC, 1577 Spring Hill RD Ste 320, Vienna, VA 22182 USA. Midnight Sun Technologies, LLC played no role in the collection, analysis and interpretation of data, in the writing of the report, or in the decision to submit the article for publication.

Declarations

Review and/or approval by an ethics committee was not needed for this study because, as a systematic review, no data for this project were collected directly from human subjects.

Informed consent was not required for this study because no data for this project were collected directly from human subjects. All data was taken from published literature in which the respective authors had obtained informed consent.

Data availability

Underlying data for this systematic review may be available from the authors of the literature reports which formed the basis of this review and are not available from the corresponding author.

Disclaimer

This article reflects the views of the author and should not be construed to represent FDA's views or policies. The author's comments are an informal communication and represent his own best judgment. These comments do not bind or obligate FDA.

CRediT authorship contribution statement

L. Ross Pierce: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: L. Ross Pierce, M.D. reports financial support was provided by Midnight Sun Technologies, LLC.

Acknowledgements

The author would like to thank Drs. Prateek Shukla and Million Tegenge for their reviews of the manuscript, and Drs. Gavin Imperato, Lei Xu, Tejashri Purohit-Sheth, and Wilson Bryan for encouragement and support. The author also thanks Dr Gavin Imperato for his assistance in screening literature search output for studies meeting selection criteria for incusion in this review, as well as Ms. Gwwendolyn Halford for assistance with the literature searches.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e31183.

References

[1] J.K. Stoller, L.S. Aboussouan, A review of a1-antitrypsin deficiency, Am. J. Respir. Crit. Care Med. 185 (2012) 246–259.
[2] J.E. Gadek, R.G. Crystal, Alpha$_1$ antitrypsin deficiency, in: F.F.B. Stanbury, F.B. Wyngaarden, D.A.S. Fredrickson, et al. (Eds.), Metabolic Basis of Inherited Disease, fifth ed., McGraw-Hill, NY, 1982, pp. 1450–1567.
[3] R.A. Stockley, Alpha-1 antitrypsin deficiency: have we got the right proteinase? Chron Obstr Pulm Dis 7 (2020) 163–171.
[4] FDA workshop co-sponsored with the alpha one foundation and the dept. Of health and human services, in: Improving Endpoints, Improving Care: Alpha$_1$-Proteinase Inhibiitor (A$_1$-PI) Augmentation Therapy and Clinical Trials. Rockville, MD, USA, 2009.
[5] R.A. Stockley, M. Miravitlles, C. Vogelmeier, Alpha One International Registry, Augmentation therapy for alpha-1 antitrypsin deficiency: toward a personalized approach, Orphanet J of Rare Diseases 8 (2013) 149–157.
[6] K. Siebers, B. Fink, A. Zakrzewicz, et al., Alpha-1 antitrypsin inhibits ATP-mediated release of interleukin-1β via CD36 and nicotinic acetylcholine receptors, Front. Immunol. 9 (2018) 877.
[7] M.J. Tobin, P.J. Cook, D.C. Hutchinson, Alpha 1 antitrypsin deficiency: the clinical and physiological features of pulmonary emphysema in subjects homozygous for PI type Z: a survey by the British thoracic association, Br. J. Dis. Chest 77 (1983) 14–27.
[8] American Thoracic Society/ERS Statement, Standards for the diagnosis and management of individuals with alpha-1 antitrypsin deficiency, Am. J. Respir. Crit. Care Med. 168 (2003) 820–899.
[9] J.K. Stoller, L.S. Aboussouan, Myths and misconceptions about α1-antitrypsin deficiency, Arch. Intern. Med. 169 (2009) 546–550.
[10] R.G. Crystal, α1-Antitrypsin deficiency, emphysema, and liver disease genetic basis and strategies for therapy, J. Clin. Invest. 85 (1990) 1343–1352.
[11] A.G. Sanford, T.D. Weir, J.J. Spinelli, P.D. Paré, Z and S Mutations of the alpha1-antitrypsin gene and the risk of chronic obstructive pulmonary disease, Am. J. Respir. Cell Mol. Biol. 20 (1999) 287–292.
[12] G.M. Turino, A.F. Barker, M.L. Brantley, et al., Clinical features of individuals with PI*SZ phenotype of alpha 1-antitrypsin deficiency, Am. J. Respir. Crit. Care Med. 154 (1996) 1718–1725.
[13] Robert Stockley, Personal Communication to Pierce, L. Ross, 2009.
[14] M. Dahl, C.P. Hersh, N.P. Ly, et al., The protease inhibitor PI*S and COPD: a meta-analysis, Eur. Respir. J. 26 (2005) 67–76.
[15] N. Seersholm, J.T. Wilcke, A. Kok-Jensen, A. Dirksen, Risk of hospital admission for obstructive pulmonary disease in α-1 antitrypsin heterozygotes of phenotype PIMZ, J Respir Crit Care Med 161 (2000) 81–84.
[16] G.E. Silva, S. Guerra, S. Keim, et al., Logitudinal decline of diffusing capacity of the lung for carbon monoxide in community subjects with the PIMZ a1-antitrypsin phenotype, Chest 133 (2008) 1095–1100.
[17] Alpha$_1$-Proteinase inhibitor (human), Zemaira prescribing information. https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=0c3354b5-a1d8-4f98-ad55-2eafe4265c4e. (Accessed 4 November 2020).

[18] Alpha1-Proteinase Inhibitor (Human), Prolastin-C prescriber information, at, https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=91edab72-c889-47S0e-8315-1798b5548dca#section-9.3. (Accessed 6 November 2020).
[19] Alpha1-Proteinase Inhibitor (Human), Aralast NP, Prescriber Information, https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=a9a5b46e-04da-41bd-bb5f-c4936b664fef. (Accessed 7 March 2023).
[20] Alpha1-Proteinase Inhibitor (Human), Glassia. Prescriber information. https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=83473cbb-48e4-42a2-81b6-4c851423da7b. (Accessed 7 March 2023).
[21] J.E. Gadek, H.G. Klein, P.V. Holland, et al., Replacement therapy of alpha 1-antitrypsin deficiency, J. Clin. Invest. 68 (1981) 1158–1165.
[22] FDA Blood Products Advisory Committee Meeting. Bethesda, Maryland. July 20-21, 2009. Transcript and Issue Summary available at, https://wayback.archive-it.org/7993/20170111012330/http://www.fda.gov/AdvisoryCommittees/CommitteesMeetingMaterials/BloodVaccinesandOtherBiologics/BloodProductsAdvisoryCommittee/ucm121612.htm.
[23] FDA website: postmarketing requirements and commitments: introduction, Available at: https://www.fda.gov/drugs/guidance-compliance-regulatory-information/postmarket-requirements-and-commitments. (Accessed 15 April 2021).
[24] FDA Blood Products Advisory Committee Meeting. Bethesda, Maryland. Nov 4, 2005. Transcript available at, https://wayback.archive-it.org/7993/20170404053758/https://www.fda.gov/ohrms/dockets/ac/05/transcripts/2005-4190t2.htm.
[25] S. Sorrells, S. Camprubi, R. Griffin, J. Chen, J. Ayguasanosa, SPARTA clinical trial design: exploring the efficacy and safety of two dose regimens of alpha1-proteinase inhibitor augmentation therapy in alpha1-antitrypsin deficiency, Respir. Med. 109 (2015) 490–499.
[26] F. Rouhani, G. Paone, N.K. Smith, et al., Lung neutrophil burden correlates with increased pro-inflammatory cytokines and decreased lung function in individuals with alpha(1)-antitrypsin deficiency, Chest 117 (5 Suppl 1) (2000), 250S-1S; Presentations at ATS Satellite Conference and Workshops sponsored by Alpha One Foundation.
[27] H.M. Morrison, J.A. Kramps, D. Burnett, et al., Lung lavage fluid from patients with alpha 1-Proteinase Inhibitor Deficiency or Chronic Obstructive Bronchitis: anti-elastase function and cell profile, Clin Sci (Colch) 72 (1987) 373–381.
[28] FDA, Center for Biologics Evaluation and Research (CBER), National institutes of health (NIH), and alpha-one foundation-sponsored public workshop. "Developing alpha-1 antitrypsin therapeutics" September 16, 2019, Bethesda, Maryland. Videocast Available at NIH VideoCast – Developing Alpha-1 Antitrypsin Therapeutics; Public Workshop.
[29] M.A. Tortorici, Quantitative disease progression model of α-1 proteinase inhibitor therapy on computed tomography lung density in patients with α-1 antitrypsin deficiency Brit, J. Clin. Pharmacol. 83 (2017) 2386–2397.
[30] J. Stolk, W. Nieuwenhuizen, High dose intravenous AAT and plasma neutrophil derived fibrinogen fragments, Thorax 60 (2005) 84–85.
[31] M.A. Campos, P. Geraghty, G. Holt, E. Mendes, P.R. Newby, S. Ma, et al., The biological effects of double-dose alpha-1 antitrypsin augmentation therapy: a pilot clinical trial, Am. J. Respir. Crit. Care Med. 200 (2019) 318–326.
[32] A. Dirksen, J.H. Dijkman, F. Madsen, et al., A randomized clinical trial of a1-antitrypsin augmentation therapy, Am. J. Respir. Crit. Care Med. 160 (1999) 1468–1472.
[33] A. Dirksen, E. Piiitulainen, D.G. Parr, et al., Exploring the role of CT densitometry: a randomised study of augmentation therapy in α_1-antitrypsin deficiency, Eur Resp J 33 (2009) 1345–1353.
[34] K.R. Chapman, J.G.W. Burdon, E. Piitulainen, et al., Intravenous augmentation treatment and lung density in severe α_1-antitrypsin deficiency (RAPID): a randomised, double-blind, placebo-controlled trial, Lancet 386 (2015) 360–368 [See also online supplementary material.].
[35] N.G. McElvaney, M. Mark Holmes, A. Glanville, P.A.B. Wark, P.J. Thompson, P. Hernandez, J. Chlumsky, H. Teschler, J.H. Ficker, N. Seersholm, A. Altraja, R. Mäkitaro, J. Chorostowska-Wynimko, M. Sanak, P.I. Stoicescu, E. Piitulainen, O. Vit, M. Wencker, M.A. Tortorici, M.F. Michael Fries, J.M. Edelman, K. R. Chapman, on behalf of the RAPID Extension Trial Group, Long-term efficacy and safety of α1proteinase inhibitor treatment for emphysema caused by severe α1 antitrypsin deficiency: an open-label extension trial (RAPID-OLE), Lancet Respir. Med. 5 (2017) 51–60.
[36] M.A. Tortorici, et al., Population pharmacokinetics of A1-PI in patients with Alpha-1 antitrypsin deficiency, Eur. Respir. J. 46 (suppl 59) (2015). PA 1486.
[37] Prolastin Summary Basis of Approval, Food and Drug Administration (1987).
[38] M.D. Wewers, M.A. Casolaro, S.E. Sellers, et al., Replacement therapy for alpha$_1$-antitrypsin deficiency associated with emphysema, N. Engl. J. Med. 316 (1987) 1055–1062.
[39] The Alpha-1 Antitrypsin Deficiency Registry Study Group, Survival and FEV_1 decline in individuals with severe deficiency of α_1-antitrypsin, Am. J. Respir. Crit. Care Med. 158 (1998) 49–59.
[40] A.R. Tonelli, F. Rouhani, N. Li, P. Schreck, M.L. Brantly, Alpha-1-antitrypsin augmentation therapy in deficient individuals enrolled in the Alpha-1 Foundation DNA and Tissue Bank, Int J Chron Obstruct Pulmon Dis 4 (2009) 443–452.
[41] K.R. Chapman, R.A. Stockley, C. Dawkins, M.M. Wilkes, R.J. Navickis, Augmentation therapy for alpha1 antitrypsin deficiency: a meta-analysis, COPD 6 (3) (2009 Jun) 177–184.
[42] N. Seersholm, M. Wencker, N. Banik, K. Viskum, A. Dirksen, A. Kok-Jensen, N. Konietzko, Does alpha1-antitrypsin augmentation therapy slow the annual decline in FEV1 in patients with severe hereditary alpha1-antitrypsin deficiency? Wissenschaftliche Arbeitsgemeinschaft zur Therapie von Lungenerkrankungen (WATL) alpha1-AT study group, Eur. Respir. J. 10 (10) (1997 Oct) 2260–2263.
[43] P.A. Dawkins, C.L. Dawkins, A.M. Wood, P.G. Nightingale, J.A. Stockley, R.A. Stockley, Rate of progression of lung function impairment in alpha1-antitrypsin deficiency, Eur. Respir. J. 33 (2009) 1338–1344.
[44] C. Tantucci, D. Modina, Lung function decline in COPD, Int J Chron Obstruct Pulmon Dis 7 (2012) 95–99.
[45] A. Agusti, L.D. Edwards, B. Celli, W. Macnee, P.M. Calverley, H. Müllerova, D.A. Lomas, E. Wouters, P. Bakke, S. Rennard, C. Crim, B.E. Miller, H.O. Coxson, J. C. Yates, R. Tal-Singer, J. Vestbo, Investigators Eclipse, Characteristics, stability and outcomes of the 2011 GOLD COPD groups in the ECLIPSE cohort, Eur. Respir. J. 42 (3) (2013 Sep) 636–646.
[46] M.J. Page, J.E. McKenzie, P.M. Bossuyt, et al., The PRISMA 2020 statement: an updated guideline for reporting systematic reviews, BMJ 372 (2021) n71, https://doi.org/10.1136/bmj.n71.
[47] Alpha1-Antitrypsin Deficiency Registry Study Group, A registry of patients with severe deficiency of alpha 1-antitrypsin. Design and methods, Chest 106 (1994) 1223–1232.
[48] F.F. Rahaghi, R. Monk, V. Ramakrishnan, T. Beiko, C. Strange, Alpha-1 antitrypsin augmentation therapy improves survival in severely deficient patients with predicted FEV1 between 10% and 60%: a retrospective analysis of the NHLBI alpha-1 antitrypsin deficiency registry, Int J Chron Obstruct Pulmon Dis 15 (2020 Dec 3) 3193–3199.
[49] M. Wencker, B. Fuhrmann, N. Konietzko, N. Banik, Longitudinal follow-up of patients with α_1-protease inhibitor deficiency before and during therapy with IV α1-protease inhibitor, Chest 119 (2001) 737–744.
[50] I.G.M. Schouten, M.J. Kasteleyn, R. Tsonaka, R. Bals, A.C. Turner, I. Ferrarotti, A.G. Corsico, B. Lara, M. Miravitlles, R.A. Stockley, J. Stolk, Long-term effect of α1-antitrypsin augmentation therapy on the decline of FEV_1 in deficient patients: an analysis of the AIR database, ERJ Open Research 7 (2021) 194–2021.
[51] C. Esquinas, S. Serreri, M. Barrecheguren, et al., Long-term evolution of lung function in individuals with alpha-1 antitrypsin deficiency from the Spanish registry (REDAAT), Int J Chron Obstruct Pulmon Dis 13 (2018) 1001–1007.
[52] R.A. Sandhaus, et al., Augmentation therapy for alpha-1 antitrypsin deficiency: improved survival and quality of life compared to matched augmentation I controls followed for up to 15 years, Am. J. Respir. Crit. Care Med. 201 (1) (2020).
[53] J.C. Barros-Tizon, M.L. Torres, I. Blanco, M.T. Martinez, the investigators of the rEXA study group, Reduction of severe exacerbations and hospitalization-derived costs in alpha-1-antitrypsin-deficient patients treatment with alpha-1-antitrypsin augmentation therapy, Ther. Adv. Respir. Dis. 6 (2012) 67–78.
[54] Global Initiative for Chronic Obstructive Pulmonary Disease – Global Strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease 2019 report. Accessed at www.GoldCOPD.org.
[55] K.R. Chapman, et al., Higher baseline lung density values are associated with an accelerated annual loss of lung density in patients with alpha-1 antitrypsin deficiency (AATD), Am. J. Respir. Crit. Care Med. 199 (9) (2019).

[56] L.J. Rosenblum, R.A. Mauceri, D.E. Wellenstein, F.D. Thomas, D.A. Bassano, B.N. Raasch, C.C. Chamberlain, E.R. Heitzman, Density patterns in the normal lung as determined by computed tomography, Radiology 137 (2) (1980 Nov) 409–416.
[57] Alpha One Foundation sponsored virtual workshop, CT lung density estimates in alpha-1 antitrypsin deficiency: state of the art, in: The 22nd Gordon L. Snider Critical Issues Workshop, 2021.
[58] D.G. Parr, A. Dirksen, E. Piitulainen, C. Deng, M. Wencker, R.A. Stockley, Exploring the optimum approach to the use of CT densitometry in a randomised placebo-controlled study of augmentation therapy in alpha 1-antitryplsin deficiency, Respir. Res. 10 (2009) 1–10.
[59] J. Stolk, B.G. Cooper, B. Stoel, A. Rames, O. Rutman, S. Soliman, R. Stockley, Retinoid treatment of Emphysema in Patients on the Alpha-1 International Registry. The REPAIR study: study design, methodology and quality control of study assessments, Ther. Adv. Respir. Dis. 4 (2010) 319–332.
[60] J. Stocks, R. Sandhaus, K. Chapman, N.G. McElvaney, et al., Baseline demographics and lung density decline in the RAPID trial, Chest 146 (4) (2014) 71A. Meeting Abstracts.
[61] R.G. Edgar, M. Patel, S. Bayliss, D. Crossley, E. Sapey, A.M. Turner, Treatment of lung disease in alpha-1 antitrypsin deficiency: a systematic review, Int. J. COPD 12 (2017) 1295–1308.
[62] P.C. Gøtzsche, H.K. Johansen, Intravenous alpha-1 antitrypsin augmentation therapy for treating patients with alpha-1 antitrypsin deficiency and lung disease, Cochrane Database Syst. Rev. (9) (2016) CD007851, https://doi.org/10.1002/14651858.CD007851.pub3. (Accessed 15 December 2020).
[63] M.A. Campos, F. Kueppers, J.M. Stocks, C. Strange, J. Chen, R. Griffin, et al., Safety and pharmacokinetics of 120 mg/kg versus 60 mg/kg weekly intravenous infusions of alpha-1 proteinase inhibitor in alpha-1 antitrypsin deficiency: a multicenter, randomized, double-blind, crossover study (SPARK), COPD: J Chron Obstr Pulm Dis 10 (2013) 687–695.
[64] M.D. Schluchter, et al., Feasibility of a clinical trial of augmentation therapy for $alpha_1$-antitrypsin deficiency, Am. J. Respir. Crit. Care Med. 161 (2000) 796–801.
[65] D. Crossley, M. Renton, M. Khan, E.V. Low, A.M. Turner, CT Densitometry in Emphysema: a systematic review of its clinical utility, Int J of COPD 13 (2018) 547–563.
[66] D. Crossley, D. Subramanian, R.A. Stockley, A.M. Turner, Proposal and validation of a minimal clinically important difference (MCID) for annual pulmonary CT density decline, Am. J. Respir. Crit. Care Med. 197 (2018) A3095.
[67] Crossley D. The relevance of CT densitometry decline in never and ex-smokers with alpha one antitrypsin deficiency (AATD) and chronic obstructive pulmonary disease (COPD); a systematic review and validation study in untreated patients. A Thesis Submitted to the University of Birmingham for the Degree of Doctor of Medicine. University of Birmingham Research Archive E-Theses Repository. Institute of Applied Health Research, College of Medical and Dental Sciences, University of Birmingham, Edgbaston, Birmingham B15 2 TT, U.K.
[68] Best – biomarkers, endpoints, and other tools resource, Available at: https://www.ncbi.nlm.nih.gov/books/NBK453484/. (Accessed 27 January 2021).
[69] P.W. Jones, A.G.N. Agusti, Outcomes and markers in the assessment of chronic obstructive pulmonary disease, Eur. Respir. J. 27 (2006) 822–832.
[70] Guidance for Industry: Adaptive Designs for Clinical Trials of Drugs and Biologics, Adaptive Design Clinical Trials for Drugs and Biologics Guidance for Industry, FDA, 2019.
[71] Guidance for Industry: Enrichment Strategies for Clinical Trials to Support Approval of Human Drugs and Biological Products. March 1019. Available at https://www.fda.gov/regulatory-information/search-fda-guidance-documents/enrichment-strategies-clinical-trials-support-approval-human-drugs-and-biological-products.
[72] R.A. Stockley, R.G. Edgar, S. Starkey, A.M. Turner, Health status decline in a-1 antitrypsin deficiency: a feasible outcome for disease modifying therapies? Respir. Res. 19 (2018) 137–146.
[73] J.A. Stockley, R.A. Stockley, E. Sapey, There is No fast track to identify fast decliners in alpha-1 antitrypsin deficiency by spirometry: a longitudinal study of repeated measurements, Int J Chron Obstruct Pulmon Dis 16 (2021) 835–840.