Pseudomonas aeruginosa and risk of death and exacerbations in patients with chronic obstructive pulmonary disease

an observational cohort study of 22,053 patients

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Title page

Category: Original article

Pseudomonas aeruginosa and risk of death and exacerbations in patients with Chronic Obstructive Pulmonary Disease: an observational cohort study of 22,053 patients.


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Abstract

Objectives: The role of *Pseudomonas aeruginosa* on long-term prognosis in COPD is unknown. The purpose of this study was to determine whether *P. aeruginosa* is associated with increased risk of exacerbations or death in patients with chronic obstructive pulmonary disease (COPD).

Methods: This is a multiregional epidemiological study based on complete data on COPD outpatients between 1 January 2010 and 31 October 2017 and corresponding microbiology and national register data. Time-dependent Cox proportional hazards models and propensity matching was used to estimate hospitalisation-demanding exacerbations and death after two years, separately and in combination.

Results: A total of 22,053 COPD outpatients were followed for a median of 1,082 days (interquartile-range: 427-1,862). *P. aeruginosa* was present in 905 (4.1%) patients. During 730 days of follow-up, *P. aeruginosa* strongly and independently predicted an increased risk of hospitalisation for exacerbation or all-cause death (HR 2.8, 95% CI 2.2-3.6; p<0.0001) and all-cause death (HR 2.7, 95% CI 2.3-3.4; p<0.0001) in analyses adjusted for known and suspected confounders. The signal remained unchanged in unadjusted analyses as well as propensity-matched subgroup analyses. Among patients “ever-colonized” with *P. aeruginosa*, the incidence of hospital-demanding exacerbations doubled after the time of the first colonization.

Conclusions: COPD patients with *P. aeruginosa* cultured from the airways had a markedly increased risk of exacerbations and death. It is still not clear whether this risk can be reduced by offering patients targeted antipseudomonal antibiotics. A randomised trial is currently recruiting patients to clarify this (ClinicalTrials.gov: NCT03262142).
Introduction

According to the Global Burden of Disease Study, 3.2 million people died from COPD in 2017 [1], and the burden of the disease continues to grow [2]. A large part of the burden of COPD is associated with recurrent events of exacerbations, which impair health status and worsen the prognosis [3].

*Pseudomonas aeruginosa* has been reported to be present in the airways as frequently as 4-20% in patients with acute exacerbation of COPD [4-7]. Although the evidence is sparse and the methodology varying, it has been suggested that *P. aeruginosa* is associated with prolonged hospitalisation, increased exacerbation rate and poor long-term prognosis in COPD patients [8-10]. However, no definitive conclusions regarding the clinical impact of *P. aeruginosa* in COPD patients can be made since the bacterium primarily is seen in advanced disease [6,11], which in itself is a strong predictor for poor prognosis [12].

Thus, the aim of the current study was to evaluate whether *P. aeruginosa* is independently associated with long-term adverse outcomes in COPD patients. To address this, we conducted a multiregional observational study with complete follow-up on the investigated endpoints and performed multivariate regression analyses and propensity score matching.
Methods

Study design

Multiregional cohort study of COPD outpatients with and without P. aeruginosa cultured from the lower respiratory tract.

Data sources

Data from nationwide and regional administrative registries in accordance with current Danish laws (Data Protection Agency: 2012-58-0004; The Danish National Committee on Health Research Ethics: H-15010949). According to these laws, informed consent is not required for registry-based studies. Linkage between registries was done by using unique personal identification numbers, which allows an exact linkage on individual level and ensures complete follow-up [13].

Data was retrieved from the nationwide Danish Register of Chronic Obstructive Pulmonary Disease (DrCOPD). DrCOPD holds individual data on all outpatient visits and hospital admissions due to exacerbation of COPD, in patients aged 30 years or above, at all Danish hospitals since 2008.

Figure 1 displays the inclusion process. All patients who were registered with an outpatient-clinic visit from 1 January 2010 to 31 October 2017 in DrCOPD were included. Patients with malignant neoplasms within five years prior to study entry were excluded since this condition is strongly associated with mortality and may affect the ability to interpret the results of the study exposure (Supplemental Table S1 lists the International Classification of Disease 10th revision (ICD-10) codes used to define malignant neoplasms).

DrCOPD-data was linked with microbiology data from the Clinical Microbiology Departments in Eastern Denmark (Region Zealand and Capital Region), with approximately 2.6 million inhabitants.

We defined the study population as COPD outpatients with any microbiological data. All patients from Eastern Denmark were included, regardless of microbiological status. Patients from the
western part of Denmark were not included, since we could not gain access to microbiological data from these patients. Study entry day was defined as the date for the patients first outpatient-clinic visit in DrCOPD.

Patients characteristics

Patient characteristics were assessed at study entry. Information was obtained from DrCOPD and the Danish National Patient Registry (DNPR). DNPR holds data on all Danish in-hospital and outpatient-clinic contacts since 1977. Supplemental Table S2 lists the ICD-10 codes used to define comorbidities.

Pseudomonas aeruginosa status

Exposure was defined as any \textit{P. aeruginosa}-positive culture sample from the lower respiratory tract (i.e. sputum, tracheal secretion, bronchial secretion and bronchial alveolar lavage) after the study participant’s entry date in DrCOPD until end of follow-up 31 October 2017. Exposure date was defined as the date of the first registered positive sample.

Outcome measures

The primary study outcomes of this study were 1) combined endpoint of hospitalisation for exacerbation or all-cause death and 2) all-cause death separately, respectively, after two years. A secondary analysis was performed to estimate the separate outcome of hospitalisation for exacerbation after two years using a competing risk model (Supplemental Table S5). We chose to assess outcome after two years since we expected a general low long-term survival rate in the study population based on previous national [14] and international literature [15] on mortality in COPD. Data on outcomes were retrieved from DrCOPD and DNPR. ICD-10 codes used to define exacerbation are listed in Supplemental Table S3.
Statistical methods

A time-dependent Cox proportional hazard regression model was developed to assess the risk between *P. aeruginosa* and outcome. *P. aeruginosa* was included as a time-dependent variable, taking time period as exposed within the first two years from study entry into account. Change in status from unexposed to exposed could only occurred once, at the time for exposure date, and remained unchanged during the remaining follow-up time. The Cox model was adjusted for known and suspected cofounders assessed at study entry based on previous literature in the field: Age (continuous), sex (male vs. female), severity of airflow obstruction based on percentage predicted forced expiratory volume in the first second; FEV$_1$ (ordinal: 1-4), medical research council dyspnoea scale; MRC (ordinal: 1-5), body mass index; BMI (continuous), smoking status (active vs. not active), previous hospitalisation for exacerbation of COPD within 12 months prior to study entry (yes vs. no), inhaled corticosteroid; ICS (yes vs. no) and calendar year for entry in DrCOPD (ordinal: 2010-2017). Patients with unknown smoking status were classified as non-active smokers. No forward or backward variable selection was made. The models were tested for linearity of continuous variables, proportion of hazards and interactions and found to be valid.

A greedy-matched propensity score model [16] was applied as a sensitivity analysis, forming a subpopulation of patients with available microbiological data on cultures samples from the lower respiratory tract. The model used algorithms created and maintained by biomedical statisticians at the Mayo Clinic [17]. Patients with *P. aeruginosa*-positive samples were matched (1:5 ratio) with patients who had never-positive-*P. aeruginosa* lower respiratory tract samples using calendar year for study entry and the probability of being exposed to *P. aeruginosa* based on characteristics at study entry (i.e. age, sex, FEV$_1$% predicted, MRC, BMI, ICS, smoking status and previous
hospitalisation for exacerbation). In patients with unknown FEV₁% predicted, MRC and BMI at study entry, measurements from the first following outpatient clinic visit were used. In the propensity matched population, the risk estimates of *P. aeruginosa* was calculated by comparing outcomes between the two matched groups by using a univariate Cox proportional hazard model (i.e. *Pseudomonas*-status: positive vs. negative). Sample date was set as exposure date in both groups. The estimate was re-tested using a robust variance estimator, accounting for the lack of independence in outcomes induced by the matching.

A second propensity score model, inverse probability of treatment weighting (IPTW) of propensity score [16], was additionally performed as a sensitivity analysis (Supplemental Table S7A-C). Cox proportional hazard regression models are presented as hazard ratios (HRs) with 95% confidence intervals (CI). Cumulative incidence-plots are used to illustrate the cumulative probability of exacerbation and death. Continuous variables are presented as median values and interquartile ranges. Group comparisons were performed using nonparametric test (Wilcoxon two-sample test) and t-test when appropriate. Categorical variables are reported as frequencies and proportions and compared between groups using Fisher’s exact test. A P-value less or equal to 0.05 was considered statistically significant. Statistical analyses were performed using SAS statistical software 9.4 (SAS Institute Inc., Cary, NC, USA) and statistical software R (version 3.4.3).

**Results**

A total of 22,053 COPD outpatients were identified in the DrCOPD between 1 January 2010 - 31 October 2017 (Figure 1). Patients were followed for 1,082 days (interquartile-range: 427-1,862). The number of patients with *P. aeruginosa* was 905 (4.1%). Table 1 displays the characteristics of patients who were *P. aeruginosa*-positive respectively *P. aeruginosa*-negative during the study period at the time for inclusion into the study. The two groups were overall similar, except for clinically relevant differences in lung function capacity, frequency of hospital-demanding
exacerbation for COPD prior to study entry, prescribed corticosteroid inhalation medicine and concurrent diagnoses of asthma and bronchiectasis. Follow-up completion for study outcomes were 100% in both groups. Time to initial \textit{P. aeruginosa}-positive sample was 649 days (interquartile-range: 216-1.278).

### Outcomes

Hospitalisation for exacerbation or all-causes death (94% vs. 48%, \(p<0.0001\)) and all-cause death separately (41% vs. 17%, \(p<0.0001\)) after two years occurred more frequently in \textit{P. aeruginosa}-positive patients compared to patients without \textit{P. aeruginosa}. \textit{P. aeruginosa} was associated with a significantly increased risk of both outcomes in unadjusted analyses (HR 3.7, 95% CI 3.0-4.6, \(p<0.0001\) resp. HR 5.1, 95% CI 4.4-5.8, \(p<0.0001\)) and the risk remained large and significant in the multivariable analyses (Table 2). Figure 2 illustrates the cumulative incidence.

Moreover, in the \textit{P. aeruginosa}-positive patients, the incidence rate of hospital-demanding exacerbations substantially increased after the first isolation of \textit{P. aeruginosa} compared to prior to isolation of the bacterium (Figure 3).

### Sensitivity analyses

A subgroup of 4.679 patients with COPD were analysed in the propensity matched model. This group consisted of 798 (17%) \textit{P. aeruginosa}-positive patients matched 1:5 with 3.881 (83%) patients with never-positive-\textit{P. aeruginosa} lower respiratory tract samples (Supplemental Figure S1). Patient characteristics of the two groups are presented in Supplemental Table S4. \textit{P. aeruginosa}-positive patients experienced significantly higher rates of both hospitalisation for exacerbation or all-cause death (89% vs. 77%, \(p<0.0001\)) and all-cause death separately (47% vs. 37 %, \(p<0.0001\)) and they had numerically higher incidence of exacerbations compared to \textit{P. 
aeruginosa-negative patients (Supplemental Table S6). Although the estimates were somewhat reduced, *P. aeruginosa* remained strongly and significantly associated with both study outcomes (HR 1.7, 95% CI 1.5-1.8, p<0.0001 resp. HR 1.4, 95% CI 1.3-1.6, p<0.0001). Supplemental Figure S2 illustrates the cumulative incidence. The outcome signal remained unchanged in the inverse probability of treatment weighting (IPTW) propensity score model (Supplemental S7A-C).

Lastly, the estimates for hospital-demanding exacerbation remained high in the competing risk analysis in the main cohort population (adjusted HR 2.7, 95% CI 2.1-3.5; p<0.0001) and propensity score subgroups (Supplemental Table S5).

**Discussion**

In this multiregional long-term follow-up epidemiological study of COPD outpatients, we found a low prevalence of *P. aeruginosa*. Patients with *P. aeruginosa* were substantially more likely to get hospitalised for exacerbation of COPD or die of all-causes compared to those who never had this bacterial pathogen. Moreover, among the patients who had *P. aeruginosa* isolated anytime, there was a substantial increase in the incidence rate of hospital-demanding exacerbations after the first isolation of *P. aeruginosa* compared to before isolation of this bacterium. The result was robust for adjustment for several known and suspected confounders and was confirmed in propensity score sensitivity analyses, using two different models.

To our knowledge, this study is the largest study ever to investigate the prevalence of *P. aeruginosa* and the clinical implications of colonization with this bacterium in an unselected population of COPD patients. Additionally, as compared with other research in this field, the current study is the first to report complete long-term follow-up on explored outcomes via nationwide registries.
Previous studies have primarily reported on smaller groups of patients, and the risk of selection bias and information bias may have been higher [4-11,18]. The prevalence of *P. aeruginosa* in our study is considerably lower compared to most of these previous studies [4,7,8], with reports of *P. aeruginosa* in up to 20% of the patients [8]. Our study was performed using complete data from the entire Eastern Denmark population during nearly eight years.

Our finding that patients colonized with *P. aeruginosa* had poor outcome is consistent with Almagro et al. [10], who found *P. aeruginosa* to be an independent prognostic marker of three-year mortality in a prospective study of 181 patients hospitalised with COPD exacerbation. On the contrary, Boutou et al. [18] reported no association between *P. aeruginosa* and long-term survival in COPD outpatients in a smaller study (n=132); power issues may, at least in part, explain that negative finding. Colonization with *P. aeruginosa* plays an important role in the course of other chronic lung diseases, in particular cystic fibrosis [19]. *P. aeruginosa* is also associated with poor outcomes in patients with bronchiectasis, in whom colonization with this bacterium has been reported to be three-fold increased [20].

The completeness of data in the current study is a major strength, allowing us to report long-term follow-up with high accuracy in a well-characterised, large and unselected group of patients. Additionally, the diagnoses used for acute COPD exacerbation and several of the comorbidities in the study have been validated with high positive predictive values in the DNPR (>90%) [21]. Moreover, use of robust and multiple acknowledged statistical models, adjusting for important prognostic predictors further strengthens the internal data validity. Nevertheless, no definitive conclusions regarding causality can be drawn based on our data due to the observational design. Moreover, we hold no data on antibiotic utilisation in the population.
Thus, it is impossible to account fully for other possible unknown confounders that could have affected the results. However, the large study size and multiregional design makes the results most likely to be generalisable to other COPD outpatients inside and outside Denmark with similar COPD characteristics.

Patients were included in the study based on the diagnosis of COPD and any available microbiological data. Thus, *P. aeruginosa*-positive patients could possibly be compared to patients where respiratory cultures never were performed. This might could be considered a limitation. However, the patients entered the study at their first outpatient-clinic visit, prior to and independent of possible respiratory samples in both groups. Furthermore, our propensity score sensitivity analyses were based on a subgroup of patients where data on respiratory cultures were available.

We chose not to control for comorbidities since the major diseases were evenly distributed between the groups. The slight difference of concurrent asthma was controlled for by adjusting for ICS in both the main- and the sensitivity analyses. ICS was seen to be strongly correlated to exacerbations. This is not surprising since ICS is associated with increased risk of pneumonia and is reserved for patients with more severe disease [22]. However, ICS use did not appear to effect mortality. Few patients in our population had bronchiectasis. As a sensitivity analysis, we ran the main regression model while excluding these and this did not alter the signal.

The study addresses the prognostic role of *P. aeruginosa* in COPD, and it reveals that this infrequent bacterium has a substantial association to key outcomes, including exacerbations and mortality. Our study was not designed to address the mechanism associated with these unwanted events. However, possible theories include enhancement of airway inflammation, resulting in susceptibility to exacerbations and accelerated loss of lung function by [23–25]. *P. aeruginosa* is
well-established contributor to irreversible decline in lung function in cystic fibrosis and the pathogenesis is closely linked to well-studied and complex virulent mechanisms, including a high genetic flexibility and biofilm formation [19,26,27].

In conclusion, the long-term outcome, both in regard to death and hospital-demanding exacerbations, was substantially worse in those COPD patients colonized with *P. aeruginosa*. Further research is needed to give a deeper understanding of the mechanism behind these adverse clinical outcomes. And most importantly, trial data are needed to determine if targeted *Pseudomonas*-active antibiotic interventions can improve the prognosis in this highly vulnerable group of patients. A randomised trial is currently recruiting patients to clarify this [ClinicalTrials.gov: NCT03262142].
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17. Biomedical Statistics and Informatics Software Packages [Internet]. Available from: http://bioinformaticstools.mayo.edu/research/gmatch/


Transparency declaration:

Conflicts of interest:

RBD reports grants and personal fees from Roche outside the submitted work. TSI reports personal fees from AstraZeneca outside the submitted work. The other authors report no conflicts of interest relevant to this article.

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Contribution:

JE and JSJ contributed to the conception and design of the study, data collection and analysis, data interpretation, and writing the manuscript. RS and TSI contributed to the conception and design of the study, data analysis, data interpretation, and revising the manuscript. PS contributed to the conception and design of the study, data collection and analysis and revising the manuscript. IA contributed to data analysis and revising the manuscript. JBB, JB, CO, RBD and USJ contributed to data collection and revising the manuscript. AB, TSL, JJ, UMW, KA, TW and NS contributed to the conception and design of the study and revising the manuscript. All authors have approved the final manuscript and agreed to be accountable for all aspects of the work.
Table 1. Characteristics of the N= 22,053 patients at study entry and by exposure to Pseudomonas aeruginosa any time during the study period (1 January 2010-31 October 2017).

<table>
<thead>
<tr>
<th>Demographics</th>
<th>All patients</th>
<th>COPD patients with <em>P. aeruginosa</em></th>
<th>COPD patients without <em>P. aeruginosa</em></th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, median (IQR)</td>
<td>69 (62-76)</td>
<td>71 (65-76)</td>
<td>69 (62-76)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Male, n (%)</td>
<td>9.868 (44.8)</td>
<td>407 (45.0)</td>
<td>9.461 (44.7)</td>
<td>0.891</td>
</tr>
<tr>
<td>MRC, median (IQR)</td>
<td>3 (2-4)</td>
<td>3 (3-4)</td>
<td>3 (2-4)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>FEV&lt;sub&gt;1&lt;/sub&gt; % predicted, median (IQR)</td>
<td>4.823 (21.9)</td>
<td>173 (19.1)</td>
<td>4.650 (22.0)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>BMI, median (IQR)</td>
<td>25 (21-29)</td>
<td>23 (20-27)</td>
<td>25 (21-29)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Smoking status, n (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Active</td>
<td>6.729 (30.5)</td>
<td>242 (26.7)</td>
<td>6.487 (30.7)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Former (&lt;6 months)</td>
<td>599 (2.7)</td>
<td>10 (1.1)</td>
<td>589 (2.8)</td>
<td></td>
</tr>
<tr>
<td>Former (&gt;6 months)</td>
<td>9.694 (44.0)</td>
<td>470 (51.9)</td>
<td>9.224 (43.6)</td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>594 (2.7)</td>
<td>21 (2.3)</td>
<td>573 (2.7)</td>
<td></td>
</tr>
<tr>
<td>Unknown, n (%)</td>
<td>4.437 (20.1)</td>
<td>162 (17.9)</td>
<td>4.275 (20.2)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Prescribed inhalation therapy, n (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inhaled corticosteroids</td>
<td>15.851 (71.9)</td>
<td>823 (90.9)</td>
<td>15.028 (71.1)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Inhaled long-acting beta2-agonist or long-acting muscarin-antagonist</td>
<td>18.315 (83.0)</td>
<td>864 (95.5)</td>
<td>17.451 (82.5)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Hospitalisation for exacerbation of COPD 12 months prior to study entry, n (%)</td>
<td>11.492 (52.1)</td>
<td>598 (66.1)</td>
<td>10.894 (51.5)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Comorbidity, n (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immunodeficiency</td>
<td>130 (0.59)</td>
<td>9 (0.99)</td>
<td>121 (0.57)</td>
<td>0.115</td>
</tr>
<tr>
<td>Inflammatory polyarthropathy</td>
<td>674 (3.1)</td>
<td>16 (1.8)</td>
<td>658 (3.1)</td>
<td>0.018</td>
</tr>
<tr>
<td>Systemic connective tissue disorder</td>
<td>566 (2.5)</td>
<td>30 (3.3)</td>
<td>536 (2.6)</td>
<td>0.161</td>
</tr>
<tr>
<td>Myocardial infarction</td>
<td>1.711 (7.7)</td>
<td>77 (8.5)</td>
<td>1.634 (7.8)</td>
<td>0.375</td>
</tr>
<tr>
<td>Atrial fibrillation</td>
<td>3.205 (14.5)</td>
<td>134 (14.8)</td>
<td>3.071 (14.5)</td>
<td>0.810</td>
</tr>
<tr>
<td>Heart failure</td>
<td>3.607 (16.4)</td>
<td>147 (16.2)</td>
<td>3.460 (16.4)</td>
<td>0.963</td>
</tr>
<tr>
<td>Hypertension</td>
<td>6.856 (31.2)</td>
<td>273 (30.2)</td>
<td>6.583 (31.1)</td>
<td>0.558</td>
</tr>
<tr>
<td>Renal failure</td>
<td>960 (4.4)</td>
<td>38 (4.2)</td>
<td>922 (4.4)</td>
<td>0.934</td>
</tr>
<tr>
<td>Peripheral vascular disease</td>
<td>1.754 (8.0)</td>
<td>66 (7.3)</td>
<td>1.688 (8.0)</td>
<td>0.490</td>
</tr>
<tr>
<td>Cerebrovascular disease</td>
<td>2.099 (9.5)</td>
<td>77 (8.5)</td>
<td>2.022 (9.6)</td>
<td>0.325</td>
</tr>
<tr>
<td>Diabetes mellitus, type 2</td>
<td>2.654 (12.1)</td>
<td>106 (11.7)</td>
<td>2.548 (12.0)</td>
<td>0.794</td>
</tr>
<tr>
<td>Asthma</td>
<td>3.216 (14.6)</td>
<td>187 (20.7)</td>
<td>3.029 (14.3)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Bronchiectasis</td>
<td>249 (1.1)</td>
<td>36 (4.0)</td>
<td>213 (1.0)</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Abbreviations: IQR; Interquartile range, MRC; Medical Research Council Dyspnoea Scale, BMI; Body Mass Index (kg/m2), FEV1; Forced Expiratory Volume in the first second.
Table 2. Cox regression hazard estimates for hospital-demanding exacerbation and all-cause death after 2 years with Pseudomonas aeruginosa as time-dependent variable.

<table>
<thead>
<tr>
<th>Hospitalisation for exacerbation of COPD or all-cause death</th>
<th>Unadjusted HR (95% CI)</th>
<th>P-value</th>
<th>Adjusted HR (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>P. aeruginosa-positive</td>
<td>3.7 (3.0-4.6)</td>
<td>&lt; 0.0001</td>
<td>2.8 (2.2-3.6)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Hospitalisation for exacerbation of COPD 12 months prior to study entry</td>
<td>2.1 (2.1-2.2)</td>
<td>&lt; 0.0001</td>
<td>1.7 (1.7-1.8)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>MRC</td>
<td>1.5 (1.5-1.6)</td>
<td>&lt; 0.0001</td>
<td>1.4 (1.3-1.4)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Inhaled corticosteroid</td>
<td>1.6 (1.5-1.7)</td>
<td>&lt; 0.0001</td>
<td>1.2 (1.2-1.3)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Active smoking</td>
<td>1.0 (1.0-1.1)</td>
<td>0.730</td>
<td>1.2 (1.2-1.3)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Age (per year)</td>
<td>1.03 (1.03-1.03)</td>
<td>&lt; 0.0001</td>
<td>1.02 (1.02-1.02)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Male</td>
<td>0.9 (0.9-1.0)</td>
<td>0.002</td>
<td>1.0 (1.0-1.0)</td>
<td>0.702</td>
</tr>
<tr>
<td>BMI (per unit increase)</td>
<td>0.98 (0.98-0.98)</td>
<td>&lt; 0.0001</td>
<td>1.00 (0.99-1.00)</td>
<td>0.074</td>
</tr>
<tr>
<td>FEV, % predicted</td>
<td>0.8 (0.8-0.8)</td>
<td>&lt; 0.0001</td>
<td>0.8 (0.8-0.9)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>All-cause death</td>
<td>Unadjusted HR (95% CI)</td>
<td>P-value</td>
<td>Adjusted HR (95% CI)</td>
<td>P-value</td>
</tr>
<tr>
<td>-------------------------------------------------------------</td>
<td>------------------------</td>
<td>---------</td>
<td>----------------------</td>
<td>---------</td>
</tr>
<tr>
<td>P. aeruginosa-positive</td>
<td>5.1 (4.4-5.8)</td>
<td>&lt; 0.0001</td>
<td>2.7 (2.3-3.4)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Hospitalisation for exacerbation of COPD 12 months prior to study entry</td>
<td>2.3 (2.2-2.5)</td>
<td>&lt; 0.0001</td>
<td>1.6 (1.5-1.8)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>MRC</td>
<td>1.9 (1.9-2.0)</td>
<td>&lt; 0.0001</td>
<td>1.6 (1.6-1.7)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Active smoking</td>
<td>0.9 (0.8-0.9)</td>
<td>&lt; 0.0001</td>
<td>1.4 (1.3-1.5)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Male</td>
<td>1.0 (1.0-1.1)</td>
<td>0.175</td>
<td>1.2 (1.1-1.3)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Age (per year)</td>
<td>1.07 (1.06-1.07)</td>
<td>&lt; 0.0001</td>
<td>1.06 (1.05-1.06)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>BMI (per unit increase)</td>
<td>0.95 (0.94-0.95)</td>
<td>&lt; 0.0001</td>
<td>0.97 (0.97-0.98)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>Inhaled corticosteroid</td>
<td>1.4 (1.3-1.5)</td>
<td>&lt; 0.0001</td>
<td>0.8 (0.7-0.9)</td>
<td>0.0007</td>
</tr>
<tr>
<td>FEV, % predicted</td>
<td>0.7 (0.6-0.7)</td>
<td>&lt; 0.0001</td>
<td>0.7 (0.7-0.8)</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

Sensitivity analysis: Cox regression hazard estimates for hospital-demanding exacerbation and all-cause death after 2 years with Pseudomonas aeruginosa as time-dependent variable after excluding 249 patients with bronchiectasis.
<table>
<thead>
<tr>
<th>Hospitalisation for exacerbation of COPD or all-cause death</th>
<th>Unadjusted HR (95% CI)</th>
<th>P-value</th>
<th>Adjusted HR (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. aeruginosa</em>-positive</td>
<td>3.3 (2.7-4.2)</td>
<td>&lt; 0.0001</td>
<td>2.5 (1.9-3.3)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>All-cause death</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>P. aeruginosa</em>-positive</td>
<td>5.2 (4.5-6.0)</td>
<td>&lt; 0.0001</td>
<td>2.9 (2.4-3.5)</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

Abbreviations: MRC; Medical Research Council Dyspnoea Scale, BMI; Body Mass Index, FEV1: Forced Expiratory Volume in the first second.

1 Increase in severity stage (1-4) of FEV1% predicted defined by the Global Initiative for Chronic Obstructive Lung Disease (GOLD).

The model is adjusted for calendar year for study entry and all variables displayed in the table.

Categorical variables are reported with one decimal place. Continuous variables are reported with two decimals.
Figure 3. Incidence of hospital-demanding exacerbation in 905 P. aeruginosa-positive patients before and after P. aeruginosa-isolation (event/1000 days).
Figure 1. Selection of study population: 22,053 patients registered with chronic obstructive pulmonary disease (COPD) in the Danish Register of Chronic Obstructive Pulmonary Disease (DrCOPD) from 1 January 2010 - 31 October 2017.

**Patients registered with COPD in the nationwide DrCOPD:**
106,560

- **Excluded:**
  - Patients without out-patient-clinic-visits: 48,717
  - Patients with malignant neoplasm*: 6,648

**Patients registered with out-patient-clinic-visits in DrCOPD:**
51,195

- **Excluded:**
  - Patients from Western Denmark (no microbiological data available): 29,142

**Study population: 22,053 COPD out-patients with microbiological data**
(Clinical Microbiology Departments in Eastern Denmark: Region Zealand and Capital Region)

**Patients with culture-positive *P. aeruginosa* in lower respiratory tract sample:**
905

**Patients without culture-positive *P. aeruginosa* in lower respiratory tract sample:**
21,148

**Follow-up completion, both end-points: 100%**

* International Classification of Disease 10th revision (ICD-10): C00-C97.