Inflammatory changes, recovery and recurrence at COPD exacerbation

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Abstract

COPD exacerbations are associated with increased airway and systemic inflammation, though relationships between exacerbation recovery, recurrent exacerbation and inflammation have not been previously reported. We related inflammatory changes at exacerbations to clinical non-recovery and recurrent exacerbations within 50 days.

Serum interleukin (IL)-6 and C-reactive protein (CRP), sputum IL-6 and IL-8 were measured in 73 COPD patients when stable, at exacerbation and at 7, 14 and 35 days post-exacerbation. In 23% of patients, symptoms did not recover to baseline by day 35, these patients had persistently higher levels of serum CRP during the recovery period (p=0.03). 22% of patients who had recurrent exacerbations within 50 days had significantly higher levels of serum CRP at day 14, compared to those without recurrences; 8.8mg/l vs. 3.4mg/l, p<0.01. Frequent exacerbators had a smaller reduction in systemic inflammation between exacerbation onset and day 35 compared to infrequent exacerbators; serum IL-6, CRP, both p<0.05.

Non-recovery of symptoms at COPD exacerbation is associated with persistently heightened systemic inflammation. The time-course of systemic inflammation following exacerbation is different between frequent and infrequent exacerbators. A high serum CRP concentration 14 days after an index exacerbation may be used as a predictor of recurrent exacerbations within 50 days.

Introduction

Chronic obstructive pulmonary disease (COPD) is characterised by episodic increases in respiratory symptoms called exacerbations. These episodes contribute considerably to the increased morbidity, mortality and health-care costs associated with this condition. Some patients are prone to frequent exacerbations and these patients have worse health status [1], greater limitation of their daily activities [2] and faster disease progression [3,4].

COPD exacerbations are associated with increases in airway and systemic inflammation [5-11] that are heterogeneous and probably related to the aetiology of the exacerbation [5, 12]. However, no detailed information is available on the time-course of airway and systemic inflammation during exacerbations and how these changes relate to clinical indices of exacerbation severity, including the length of recovery. We have previously shown that patients with frequent exacerbations have increased airway inflammation in the stable state [5]. We have also demonstrated that non-recovery of symptoms and peak flow at 35 days after the onset of the exacerbation occurs in up to 25% of patients [13].

Exacerbations have a tendency to cluster in an individual as demonstrated by studies of patients hospitalised with severe COPD exacerbations. Such studies have shown that after the index exacerbation, patients are at increased risk of re-admission and one study showed that 34% of patients were re-admitted with a recurrent exacerbation in the three months following their discharge [14-17]. Frequent re-admissions with COPD exacerbations have been highlighted as an independent risk factor for increased mortality [15]. It is currently unknown if these recurrent exacerbations are associated with persistence of a heightened inflammatory status, or are perhaps related to non-recovery of the index exacerbation.

We hypothesised that persistent exacerbation symptoms may be related to a heightened inflammatory status, and that clinical non-recovery and recurrence of exacerbations would relate to non-recovery of the inflammatory response. We therefore performed a prospective study in a well-characterised cohort in which airway and systemic inflammation were assessed in the stable state, at exacerbation onset prior to treatment, and throughout the recovery period at days seven, 14 and 35. Secondly, we assessed the relationship between these airway and systemic markers at exacerbation with recurrent exacerbations occurring within 50 days of the index event. Finally, we studied the evolution of airway and systemic inflammatory markers in patients stratified according to their exacerbation frequency.

Method

Patient recruitment

Seventy-three patients were recruited from the London COPD study. This is a rolling cohort of patients used to prospectively investigate the mechanisms of COPD exacerbations [1, 2, 4-6, 10-13]. COPD was defined as a post-bronchodilator forced expiratory volume in one second (FEV_1) to forced vital capacity (FVC) ratio below 70% and a β_2-agonist reversibility on predicted FEV_1 of less than 15% or 200ml. Patients with a history of other significant respiratory diseases were excluded, as were those unable to complete daily diary cards. Patients were recruited when stable, at least six weeks away from their last exacerbation. At recruitment, patients' daily respiratory symptoms, smoking history, COPD exacerbation history and drug history were recorded. Height and weight were measured, in addition to baseline lung function using a rolling seal spirometer (Sensor Medic Corp, Yorba Linda, US).

Reversibility to β_2-agonists was measured after inhaling 400μg salbutamol from a metered dose inhaler via spacer. Partial pressures of arterial oxygen and carbon dioxide were measured on arterialised ear lobe blood gases (Model 278 Blood Gas Analyser, Ciba-Corning, Medfield, US). Patients also completed the St. George's Respiratory Questionnaire (SGRQ), which is a disease-specific measure of health-status [18].

The study was approved by the ethics committee of the East London and City Health Authority and all patients gave written informed consent. Twelve of these samples were previously used for an analysis of the upper and lower airway at exacerbation, though time course and exacerbation recurrence was not addressed in that paper [10] and all the current analyses are original.

Patient follow-up and exacerbation

All patients were asked to keep daily diary cards on which they recorded their morning post-medication peak expiratory flow rate (PEFR) with a mini-Wright peak-flow meter (Clement-Clarke International Ltd, Harlow, UK) and any increase in their daily respiratory symptoms. Patients were routinely followed up every three months in the research clinic, their diary cards were reviewed and spirometry was recorded. Patients were asked to contact the study team when they experienced any increase in their daily respiratory symptoms. They were generally seen within 48 hours by a study-physician, their symptoms were reviewed and the exacerbation was confirmed according to our symptomatic definition described below. Patients were sampled before starting any additional treatment for the exacerbation.

Definition of baseline

Baseline was defined as the median symptom score recorded on days 14 to eight, prior to the onset of the exacerbation.

Definition of exacerbation

The diagnosis of an exacerbation was based on symptomatic criteria previously validated by our group against important outcome measures in COPD including sputum inflammatory markers [5], the rate of decline in lung function [4] and quality of life [1]. Increased dyspnoea, increased sputum volume and increased sputum purulence were called major symptoms; nasal congestion or discharge, sore throat, increased chest tightness or wheeze or increased cough were termed minor symptoms. An exacerbation was defined as any two major symptoms or one major and one minor symptom, present for at least two consecutive days, the first of which was called the day of onset of the exacerbation.

Exacerbation severity

Each symptom, major and minor, was binary coded as one (present or increased over baseline) or zero (absent or not increased) and then summed. This symptom score was calculated daily throughout the exacerbation. Exacerbation severity was assessed by using the increase in the total symptom score at onset of exacerbation and the percentage reduction in FEV_1 compared to baseline.

Recovery time

The recovery time for symptoms was defined as the time from onset of the exacerbation to the day on which a three day moving average of the symptom score had returned to baseline. In 12 exacerbations the diary card data was incomplete and a recovery date could not be ascertained.

Non-recovered exacerbation

A non-recovered exacerbation occurs when the total symptom score had not returned towards baseline by day 35 after exacerbation onset.

Recurrent exacerbation

A recurrent exacerbation was defined in this study as a second exacerbation, fulfilling our definition, occurring within 50 days of the index exacerbation. Symptoms from the first exacerbation must have recovered, defined as above.

Time to the next exacerbation

Time to the next exacerbation was defined as the time between the onset of the index exacerbation and the onset of the next exacerbation. These data were collected from clinic visits and the daily diary cards. Data were available for up to ten months after the last sampled exacerbation.

Exacerbation frequency

The number of exacerbations experienced by each patient was determined and the median exacerbation frequency was calculated. The patients were then divided into two groups around the median exacerbation frequency, those above the median were called frequent exacerbators, and those with an exacerbation frequency below the median were called infrequent exacerbators.

Patient sampling

Patients were seen and sampled in the stable state, at exacerbation prior to treatment, and seven days, 14 days and 35 days later. Pre-exacerbation baseline samples were not available in 12 patients and post-exacerbation samples were therefore employed, obtained a minimum of 42 days after the preceding exacerbation. At each visit, the patients' symptoms were documented, lung function was recorded, and sputum and serum were collected. Spontaneous sputum was used, or when no spontaneous sputum was available, patients were induced according to our previously described methodology [19]. 97% of sputum samples were spontaneous. We have previously reported on the equivalence of induced and spontaneous samples for the assessment of airway inflammatory markers [19]. Sputum and blood samples were processed within two hours of collection. Sputum was processed according to our previously described protocol [19], in the absence of dithiothreitol, and the supernatants were stored at -80ºC. 7ml of peripheral venous blood was collected into a vacutainer tube and centrifuged at 2000rpm for 10 minutes at 4ºC. The serum was separated and stored at -80ºC until later analysis. All exacerbations were treated with bronchodilators, antibiotics and or oral steroids depending on the clinical severity of the episode, as judged by the attending physician.

Measurement of inflammatory markers

Sputum and serum interleukin (IL)-6 and sputum IL-8 were quantified using commercial sandwich ELISA (Enzyme-linked immunosorbent assay) kits (R&D Systems, Abingdon, UK). Serum C reactive protein (CRP) was measured in our hospital laboratory using an Olympus luminometric analyser. All the samples from one patient were measured in the same assay to reduce inter-assay variability. The limit of detection for serum and sputum IL-6 was 0.7pg/ml, for sputum IL-8 it was 10 pg/ml, and for serum CRP it was 0.3 mg/l. The reported levels of all the sputum mediators are ten-fold dilutions by weight of the original sputum sample.

Statistical analysis

Data were analysed using STATA-5 software (Stata Corporation, Texas, US) and SAS version 8. The Kolmogorov-Smirnov test of normality was applied. Normally distributed data were expressed as mean and standard deviation (SD), skewed data as median and inter-quartile range (IQR). Skewed data were log transformed to obtain a normal distribution. Comparisons between baseline and exacerbation data, and between the different exacerbation visits for symptoms, lung function parameters, and airway and systemic inflammatory markers, were analysed using paired t-test, unpaired t-test and Mann-Whitney U tests as appropriate. Spearman's correlation was used to assess the relationships between changes in airway inflammation and recovery time, and between serum CRP at day 14 and time to the next exacerbation. Chi-square test was used to compare exacerbation symptoms at the index and recurrent exacerbation. The percentage changes between baseline and the different exacerbation visits for each parameter were calculated.

A mixed linear regression model was used to study the changes in the time-series of cytokine data, as this technique gives more flexibility to analyse within patient changes, it accounts for a good level of correlation among the variables modelled and it accommodates for missing data and does restrict the bias towards zero. The mixed linear models with random effects compared the evolution of the time curve during the recovery phase of the exacerbation in two groups of patients separated by a binary trait (i.e. frequent versus infrequent exacerbators and recovered versus non-recovered). First, we determined individually the best fitting curve shape for the different cytokines separated by a time variable. Then the best fitting curve model (using time, time-squared or time-cubed) was placed in the final model, in interaction with the binary trait to explain the response of the log-transformed cytokine. Finally, we produced a visual output from this modelling using predicted values for each time point, expressed as a percent of the value at exacerbation onset, accompanied by one standard error (SE). This part of the analysis was supported using SAS version 8. A probability of error of less than or equal to 5% was considered statistically significant.

Results

Patient characteristics

Seventy-three exacerbation time-courses were collected from 73 different patients; their baseline characteristics are reported in Table 1 which demonstrates that the cohort have moderately severe disease with a mean FEV_1 of 1.08 l or 45% predicted. The median (IQR) exacerbation frequency was 3.1 (2.0-4.2) per year. There were no significant differences in the baseline characteristics between frequent and infrequent exacerbators, except for the

exacerbation frequency itself. Median(SD) FEV_1 in frequent exacerbators was 1.10(0.53) l versus 1.05(0.41) l in infrequent exacerbators, p=0.62.

Exacerbation characteristics

At exacerbation, patients were seen and sampled a median (IQR) of 2 (1-3) days after the onset of the exacerbation, and before they started treatment. The median (IQR) total symptom score at exacerbation onset was 5 (4-6). The median (IQR) percentage change in FEV_1 at exacerbation compared to baseline was -5.05 (-15.02 to 6.18) %. All exacerbations were treated exacerbations, 63% were treated with antibiotics and oral corticosteroids, 4% with steroids alone and 32% with antibiotics alone.

Time-course of airway and systemic inflammation at exacerbation of COPD

The time-course data for the inflammatory markers at exacerbation are reported in Table 2. There was a significant rise in serum IL-6, serum CRP and sputum IL-6 between baseline and onset of the exacerbation. The concentration of serum IL-6 measured at day seven was significantly lower than that at baseline, reflecting the response to exacerbation therapy. By day 14, both airway and systemic inflammatory markers had returned to concentrations not significantly different from those present at baseline.

Exacerbation recovery

The median (IQR) symptom recovery time was 9 (4-18) days. In 23% of exacerbations, symptoms had not recovered to baseline by day 35. There were no significant differences in

clinical indices of non-recovery between the frequent and infrequent exacerbators: e.g. non-recovery of symptoms was 31% in frequent exacerbators vs. 25% in infrequent exacerbators (χ^2=0.28, p=0.60). There was no significant difference in treatment with oral steroids at onset of the exacerbation between patients with recovered and non-recovered exacerbations, (χ^2=1.02, p=0.31).

In a mixed linear model analysis, patients with non-recovered exacerbations at day 35 had a persistently higher serum CRP (but not IL-6) concentration during the recovery period, compared to those patients who had recovered (p=0.03). This is illustrated in Figure 1.

Although there was no significant rise in sputum IL-8 at exacerbation onset, when each person's exacerbation value is corrected for their own baseline, we observed that the magnitude of rise in IL-8 was correlated with the symptom recovery time: r_s =0.41, p=0.01. Similarly, there were significant relationships between symptom recovery time and changes in sputum IL-6 and IL-8 between baseline and day seven, likely reflecting the completeness of response to therapy. The greater the reduction in inflammatory marker, the shorter the recovery time as illustrated in Figure 2 (sputum IL-6: r_s=0.57, p=0.004; sputum IL-8: r_s=0.48, p=0.01). The symptom recovery time was not related to the changes in sputum IL-6 or serum IL-6 and CRP between baseline and exacerbation onset, or at any other time-points during the recovery.

Exacerbation frequency

The time-trends at exacerbation in serum IL-6 and CRP, between frequent and infrequent exacerbators, are illustrated in Figures 3 and 4. Frequent exacerbators had a smaller reduction

in systemic inflammation between exacerbation and day 35, despite treatment, compared to infrequent exacerbators: the evolution of serum IL-6 and CRP was significantly different between these groups, both p<0.05. However, analysis of the baseline and exacerbation onset samples showed no significant difference in the percentage rise in serum CRP (480% vs. 443%, p=0.42) or serum IL-6 (245% vs. 301%, p=0.75) between patients with frequent and infrequent exacerbations. There was a trend towards frequent exacerbators being treated with oral steroids more often than infrequent exacerbators (χ^2=2.86, p=0.07).

Recurrent exacerbations

22% of patients had a recurrent exacerbation within 50 days of the index exacerbation. These recurrent exacerbations were all treated exacerbations. Patients who presented with symptoms of a common cold at the first exacerbation were likely to present with a cold at the recurrent exacerbation (χ^2=6.11 and p=0.01). This relationship was not statistically significant for any of the other exacerbation symptoms. Patients who had a recurrent exacerbation had a significantly higher serum CRP concentration 14 days after the index exacerbation, compared to those who did not have a recurrent exacerbation: 8.8 (4.4-15.9) mg/l versus 3.4 (2.1-5.5) mg/l, p= 0.007, Figure 5. In a multivariate analysis, the CRP at day 14 was related to a recurrent exacerbation within 50 days independent of the disease severity, the exacerbation frequency and the treatment of the index exacerbation with oral steroids, p=0.004. No such relationships were demonstrated for the other markers and time-points.

There were no significant relationships between recurrent exacerbations and the concentrations of airway or systemic inflammatory markers in the stable state prior to the index exacerbation. The severity of the index exacerbation was also not related to the

occurrence of recurrent exacerbations (total symptom score for non-recurrent vs. recurrent exacerbations were 3 (IQR 2.0-3.7) vs. 3 (IQR 2-4) respectively (p=0.99), and percentage change FEV_1 -4.8% (-11.6 to -1.2) vs. -12.9% (-14.1 to -6.6), p=0.20).

Time to the next exacerbation

The median (IQR) time to the next exacerbation was 93 (50-178) days. 75% of the subsequent exacerbations were treated exacerbations. A higher CRP concentration 14 days after the index exacerbation was associated with a shorter time to the next exacerbation. This is illustrated in Figure 6: r_s =-0.47, p<0.01.

Discussion

This is the first study to evaluate the time-course of recovery in airway and systemic inflammatory markers at exacerbation of COPD, in a cohort of patients with well-characterised disease. The new and principal findings of this study may be summarised thus. 1) Assessment of serum CRP concentration 14 days after an exacerbation of COPD may be clinically important, as we have established a relationship between non-resolution of systemic inflammation and recurrent exacerbations within 50 days. 2) We have identified a direct relationship between symptom recovery time and the response of airway inflammation to exacerbation therapy. 3) We have also demonstrated, for the first time, that frequent exacerbators have a reduced response to therapy which results in persistently higher systemic inflammatory markers and which may explain the greater decline in lung function observed in

these patients. 4) We have also observed that serum inflammatory markers were better predictors of non-recovery and recurrent exacerbations than sputum inflammatory markers.

We have studied the relationships between airway and systemic inflammatory markers, symptom recovery time, recurrent exacerbations and time to the next exacerbation in a prospectively followed cohort. All the patients recruited into this study completed daily diary cards for changes in exacerbation symptoms and reported exacerbations to the study team as soon as possible after the onset. A unique feature of using daily diary cards is that we can collect precise data on the start and end of an exacerbation and accurately determine non-recovery of symptoms and PEFR. In addition, the study design allowed sampling early in the time-course of the exacerbation when the inflammatory markers reached their initial peak. All the initial exacerbation samples were taken before starting additional exacerbation treatment.

We have previously described that the recovery time of an exacerbation is an index of exacerbation severity [13], and that patients with a longer exacerbation recovery are more likely to have had a delay between exacerbation onset and treatment initiation [20]. Our previous work has also described that a significant number of COPD exacerbations do not recover to baseline [13]. In the present study we have established, for the first time, that patients who do not recover to baseline have persistently higher serum CRP levels during the recovery period. Our results thus suggest that the systemic inflammatory response is an important marker of the recovery time and outcome of the exacerbation.

It is now recognised that exacerbation frequency is an important outcome in COPD, as patients prone to frequent exacerbations have impaired health status [1], reduced physical

activity [2], increased lower airway bacterial colonisation [21] and a faster decline in lung function [3, 4, 22]. These frequent exacerbators also have increased airway inflammation in the stable state [5] and we have recently reported that frequent exacerbators have a faster rise in systemic inflammation over time compared to infrequent exacerbators [23]. In the present study we have established that patients who are frequent exacerbators have significantly higher levels of serum IL-6 and CRP during the recovery period of an exacerbation, and this may account for the increased morbidity and mortality seen in this group. We have recently described that the increase in airway and systemic inflammation in stable COPD patients over time is directly linked to disease progression [23] and thus it can be extrapolated that this delayed recovery in inflammatory markers in frequent exacerbators may be one of the mechanisms underlying faster FEV_1 decline in this group [3, 4, 22]. The slower resolution of exacerbation inflammatory markers in the frequent exacerbator group may also explain the increased airway inflammation that we have previously observed in the stable state [5]. In this study we did not see a difference in recovery time between frequent and infrequent exacerbators, consistent with our previous findings [13].

Although most COPD exacerbations are treated in the community, they are an important cause of hospitalisation and are responsible for around 10% of all acute medical admissions [24]. Within the year after admission to hospital with an exacerbation, 63% of patients are re-admitted at least once [14], and this figure increased to 79% in patients admitted with acute hypercapnic respiratory failure [17]. These repeat admissions were mainly due to recurrent exacerbations and known risk factors for re-admission include being a frequent exacerbator and having more severe COPD [14]. A recent study has reported that recurrent hospital admissions with COPD exacerbations are an independent risk factor for increased mortality [15]. Identification of patients likely to have a recurrent exacerbation early in the course of

their index exacerbation may allow early implementation of appropriate preventive strategies and would be an important new approach in the management of this disease.

A further unique finding of this study is the important association between raised serum CRP during the recovery period and recurrent exacerbations. These findings need to be explored further with interventional studies of therapies such as oral anti-inflammatory agents targeted at those patients with a high CRP concentration after exacerbation. Interventions used in this manner may be capable of preventing recurrent exacerbations. Reducing recurrent exacerbations would lead to substantial reductions in morbidity, mortality and health-care costs.

We have previously described that early presentation to health-care professionals can affect the outcome of the exacerbation and reduce hospitalisation [20]. The present study emphasises the importance of following-up COPD exacerbations and this issue could be incorporated into COPD management guidelines. This might comprise, for example, routine follow up at 14 days after the index exacerbation. Serum CRP can be easily and rapidly measured in many health-care settings and can therefore be integrated into the routine follow-up of patients with COPD exacerbations without major additional cost.

A high CRP level despite treatment with antibiotics and steroids could be related to several factors such as failure of the therapy prescribed at exacerbation to eradicate the causative agent, or resistance of the responsible organism to the treatment prescribed. It may also be possible that heightened inflammatory status, or modification of bacterial flora after the index exacerbation could facilitate infection by a different organism [25]. Change in the strain of bacteria colonising the lower airways has also been implicated in COPD exacerbations and

strain changes may therefore be involved in exacerbation recurrence [26]. Comparing the nature of the index and recurrent exacerbation, we found that only coryzal symptoms were significantly associated with both events. This suggest that patients who develop recurrent exacerbations may be more susceptible to respiratory viral infections and this is consistent with a recent study from our group in which frequent exacerbations were more likely to acquire colds than infrequent exacerbators [27]. Further studies are required to evaluate the origin of the inflammatory response at a recurrent exacerbation.

COPD exacerbations are associated with increases in airway inflammation, and IL-8 is one of the markers of the neutrophilic inflammation found in this condition. Airway IL-8 concentration has been related to the degree of airflow obstruction [28], smoking status [29] and bacterial colonisation [30, 31]. An important finding from the present data was that the magnitude of the increase in sputum IL-8 at exacerbation was directly related to the symptom recovery time. We have also demonstrated a prolonged recovery time in those patients who, despite treatment with antibiotics and steroids, had persistently high levels of sputum IL-6 and IL-8 at day seven compared to baseline. This is likely to reflect the failure to respond to the treatment prescribed at exacerbation. We have therefore established, for the first time, relationships between the non-recovery of airway inflammation and symptoms at exacerbation of COPD.

In conclusion, persistence of increased airway inflammation at exacerbation of COPD is associated with a prolonged symptom recovery time. Heightened systemic inflammation is associated with recurrent exacerbations within 50 days. These results could form the basis for new therapeutic strategies to prevent recurrent exacerbations and delayed recovery. We suggest that it is now important to incorporate into integrated COPD care pathways both early

presentation that improves the outcome of the exacerbation, and adequate follow-up after the index event to reduce recurrent exacerbations. With these novel approaches we may considerably reduce the health-burden of this important condition.

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Legend to figures

FIGURE 1: Time-trend of serum CRP in patients whose symptoms had and had not recovered at day 35, p=0.03. Estimates from the mixed linear model analysis on log transformed data were anti-logged, and means and one standard error (SE) expressed as a percentage of the value of the exacerbation onset sample (time =0).
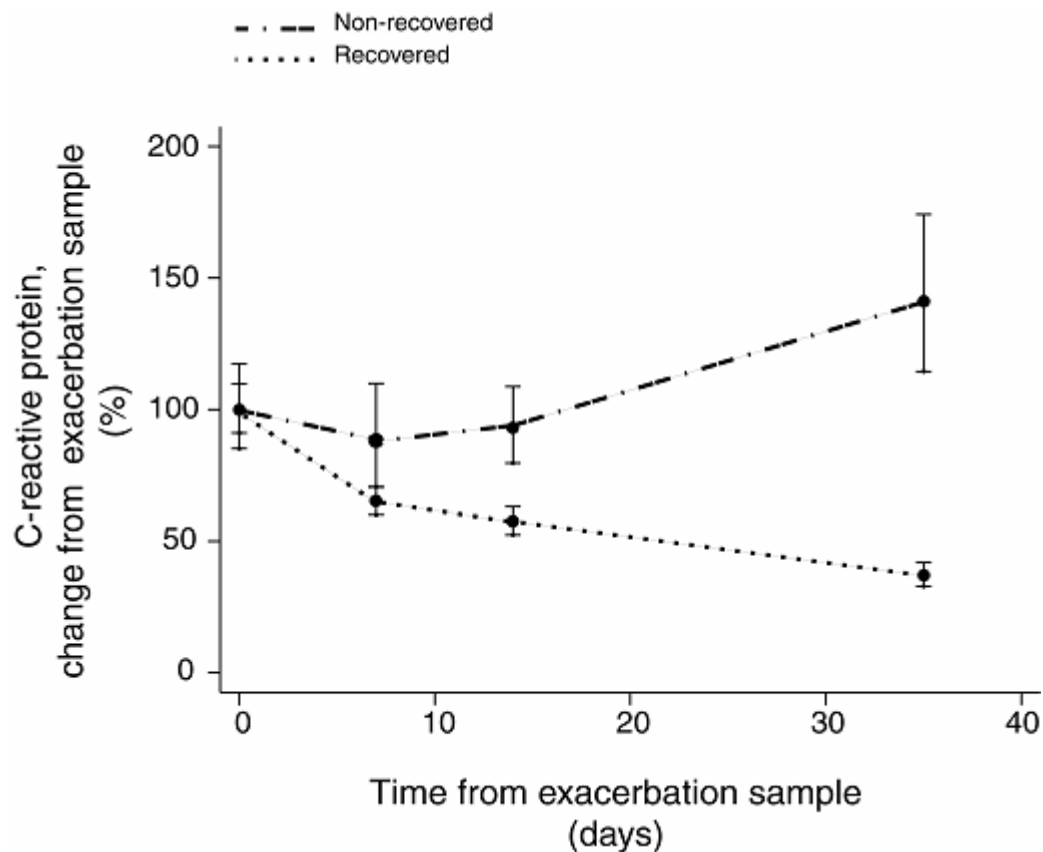

FIGURE 2: Relationship between exacerbation symptom recovery time and changes in sputum IL-6 (r_s=0.57, p=0.004) and sputum IL-8 (r_s =0.48, p=0.01) between baseline and day seven, likely reflecting the completeness of response to therapy.
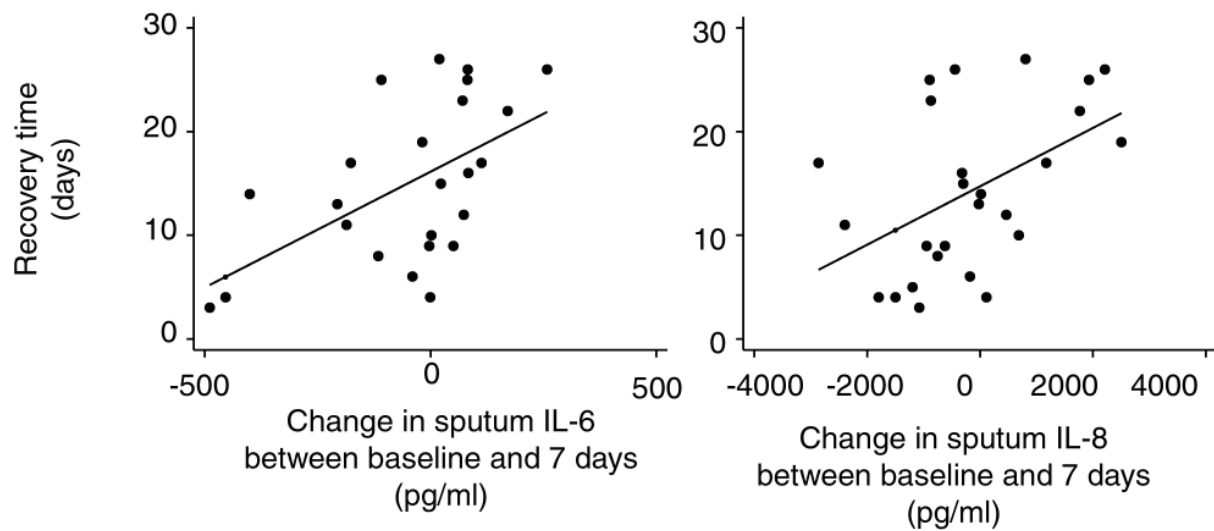
FIGURE 3: Time trend of serum IL-6 at exacerbation of COPD and during recovery, in frequent and infrequent exacerbators, p<0.05. Estimates from the mixed linear model analysis on log transformed data were anti-logged, and means and one standard error (SE) expressed as a percentage of the value of the exacerbation onset sample (time =0).
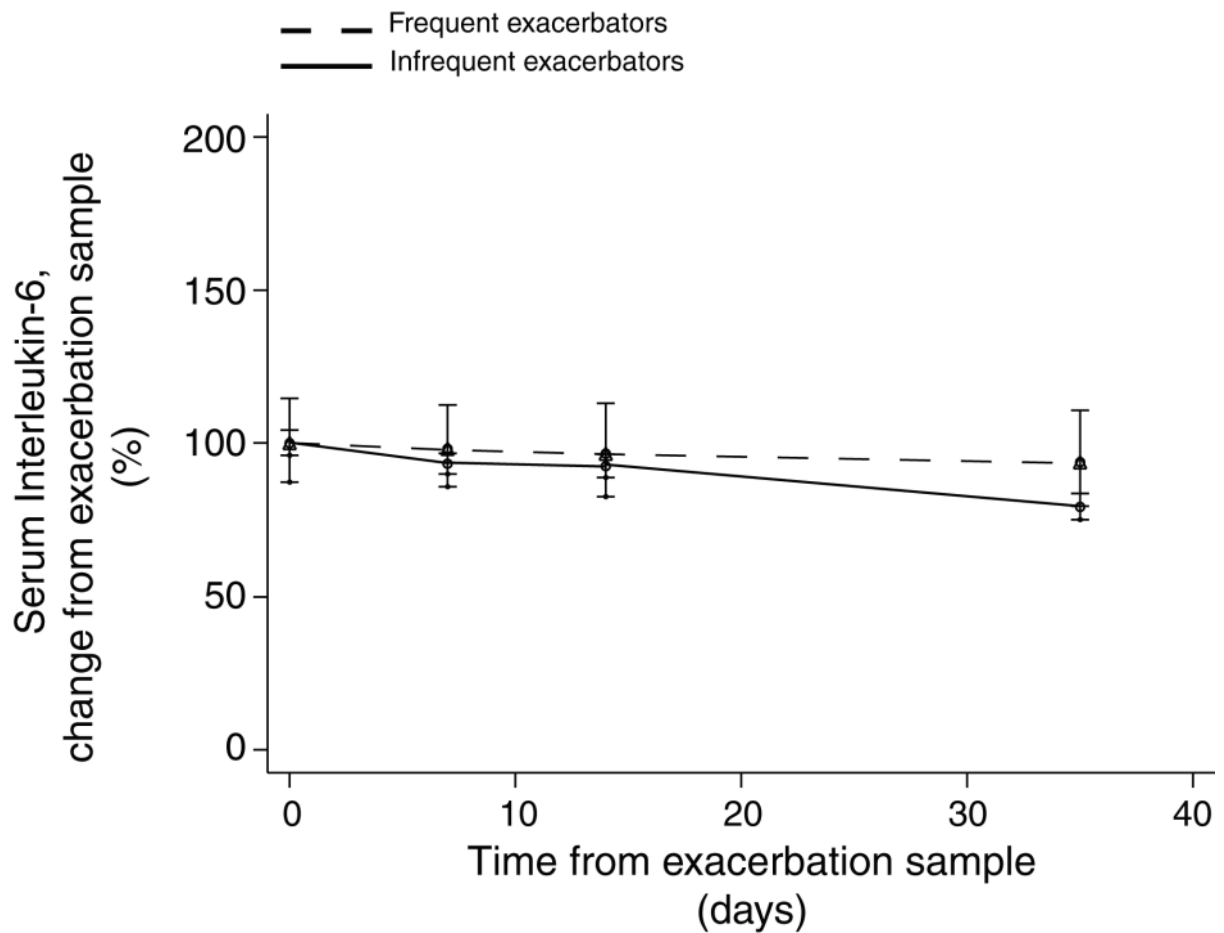
FIGURE 4: Time trend of serum CRP at exacerbation of COPD and during recovery, in frequent and infrequent exacerbators, p<0.05. Estimates from the mixed linear model analysis on log transformed data were anti-logged, and means and one standard error (SE) expressed as a percentage of the value of the exacerbation onset sample (time= 0).
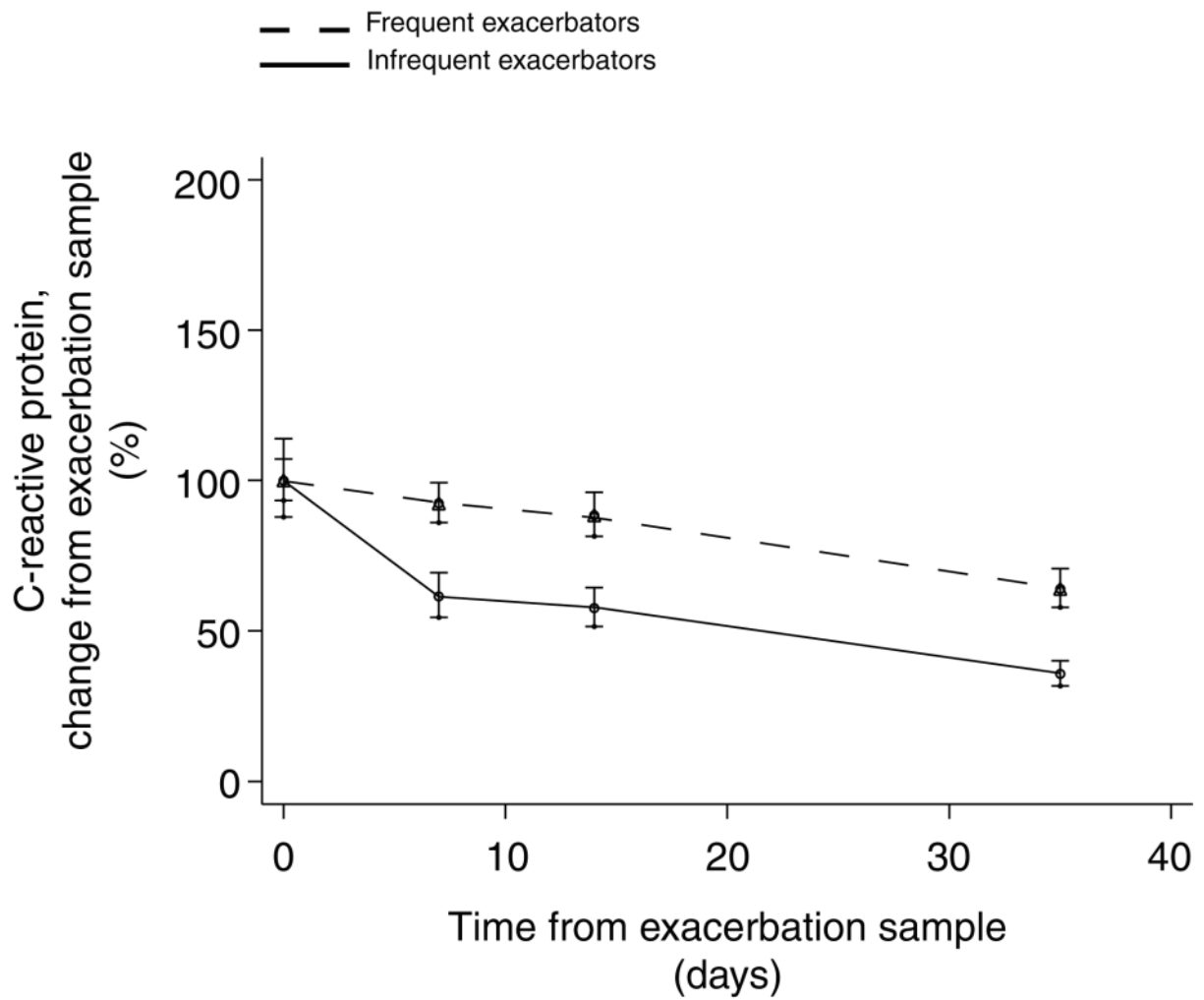
FIGURE 5: Differences in median (IQR) serum CRP concentration at day 14 between patients with and without a recurrent exacerbation within 50 days, 8.8mg/l vs. 3.4mg/l, p=0.007.
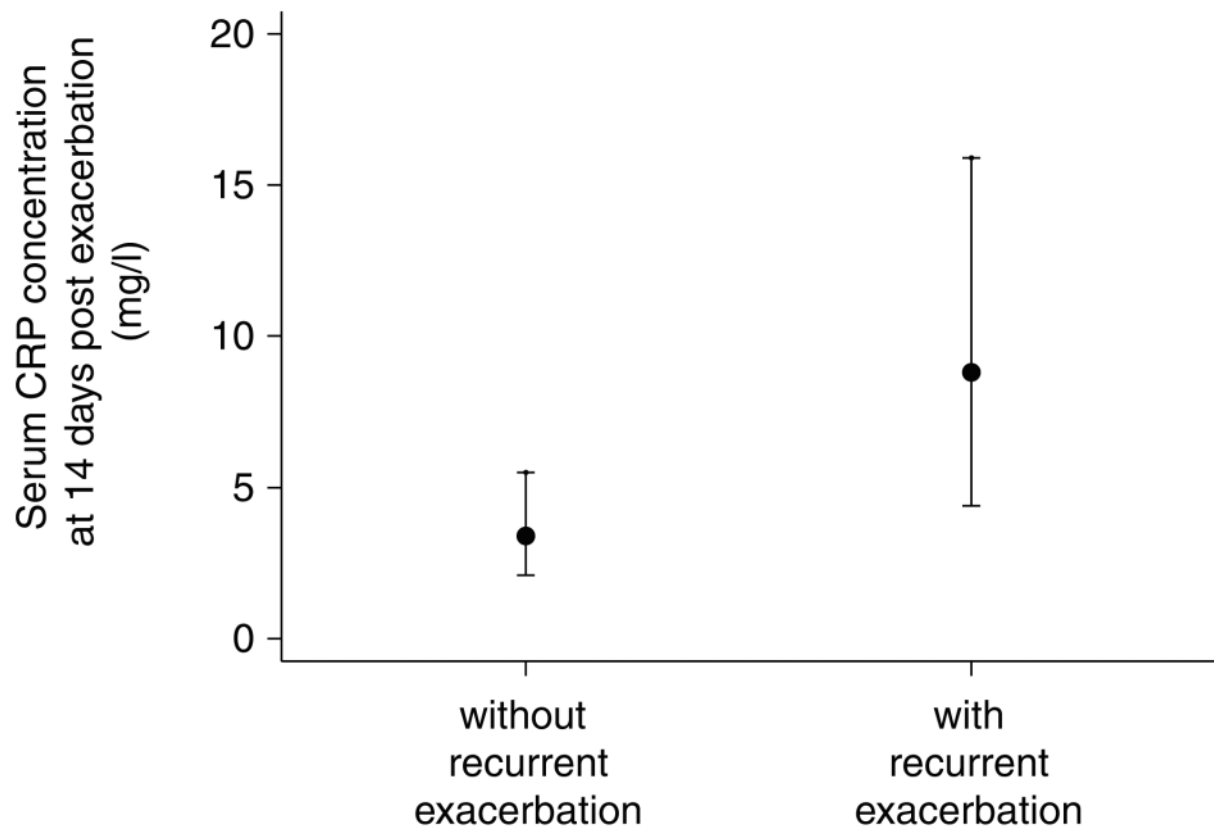
FIGURE 6: Relationship between serum CRP concentration at day 14 of the index exacerbation and time to the next exacerbation (r_s =-0.47, p<0.01).
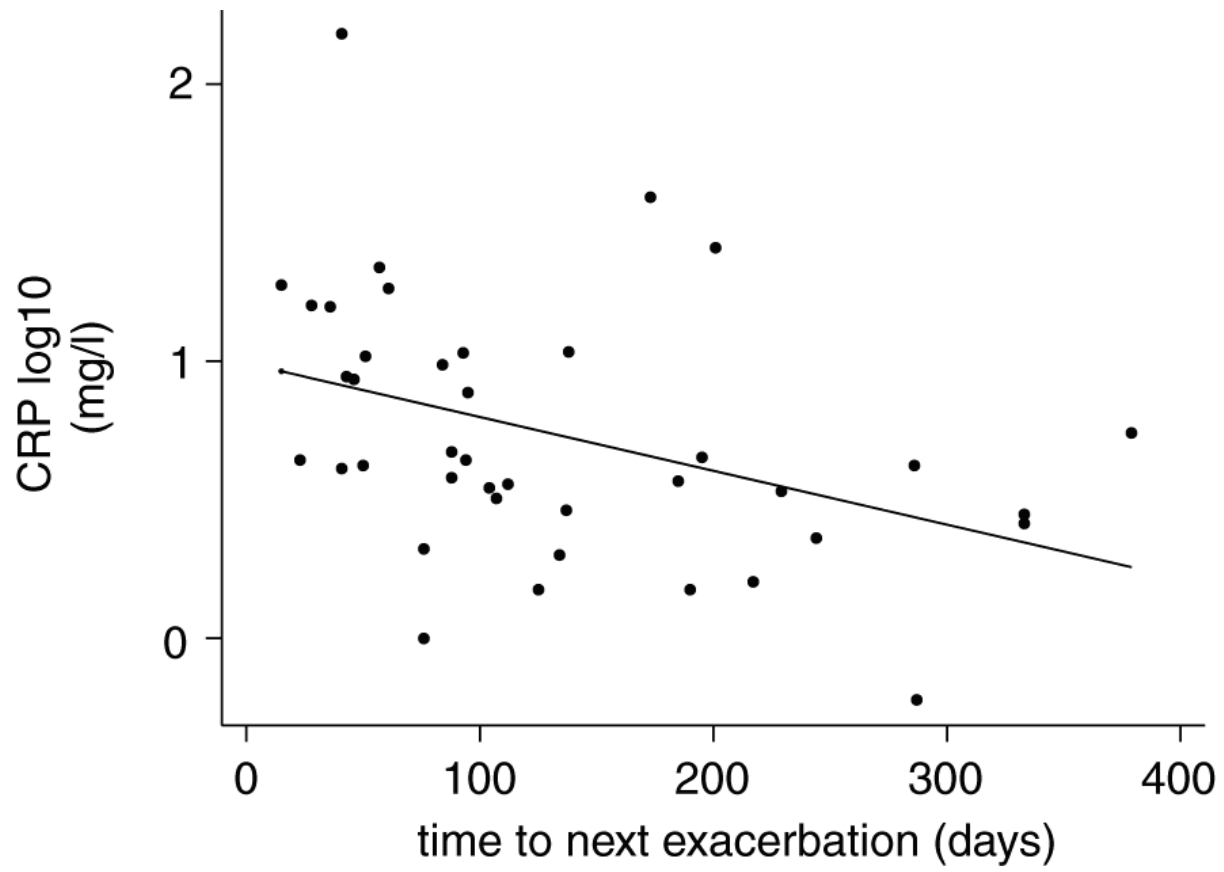
Table 1: Baseline characteristics of the 73 COPD patients (39 male, 20 active smokers).

	MEAN	SD
Age (years)	69.3	7.8
FEV_1 (l)	1.08	0.47
FEV_1 (% predicted)	45	18
FVC (l)	2.36	0.87
BMI (kgm^{-2})	26.2	6.3
PaO_2 (kPa)	8.86	1.08
$PaCO_2$ (kPa)	5.74	0.95
Smoking (pack years)	48.1	34.9
SGRQ (total score)	59.0	18.3

Table 2: Time-course of inflammatory markers at exacerbation of COPD. All concentrations are reported as median (IQR).

Marker	Baseline	Exacerbation Onset	p= Baseline - Onset	Day 7	p= Baseline - Day 7	Day 14	p= Baseline - Day 14	Day 35	p= Baseline - Day 35
SPUTUM									
IL-6 pg/ml	112 (40-219)	161 (86-308)	**0.03**	144 (64-226)	0.31	124 (83-236)	0.95	216 (76-319)	0.63
IL-8 pg/ml	2941 (1913-3808)	3276 (2285-4129)	0.13	2653 (1630-3912)	0.09	2296 (1667-3424)	0.12	3305 (2400-4118)	0.67
SERUM									
IL-6 pg/ml	5.6 (3.1-8.4)	12.1 (4.0-19.9)	**<0.01**	3.9 (1.6-6.4)	**<0.01**	4.9 (2.6-8.7)	0.16	5.8 (3.2-9.0)	0.38
CRP mg/l	6.5 (3.8-11.8)	10.9 (5.5-34.2)	**<0.01**	5.3 (3.5-9.4)	0.95	4.0 (2.3-9.6)	0.10	6.8 (3.6-11.6)	0.42