Chronic obstructive pulmonary disease, neutrophils and bacteria: from science to integrated care pathways

RA Stockley

ABSTRACT – The incidence of chronic obstructive pulmonary disease (COPD) and its effect on morbidity and mortality is rising. Extensive research implicates the role of inflammation and particularly the effect of neutrophil proteinases in generating many of the pathological features seen as part of the COPD syndrome.

Exacerbations of COPD are an important cause of morbidity, mortality and progressive deterioration in lung function. These episodes are poorly defined but bacteria in the airways drives neutrophilic inflammation and proteinase release and hence may play a key role. The differentiation between benign colonisation and heightened neutrophilic inflammation can be determined by observation of sputum purulence. This reflects the likelihood of bacteria being present and particularly the number of bacteria. This simple clinical observation enables decision making about the need for antibiotic therapy to be as much a part of patient self-management as physician directed management in the integrated care pathway for COPD.

KEY WORDS: antibiotics, bacteria, chronic obstructive pulmonary disease, exacerbations, inflammation, neutrophils, sputum

The incidence of chronic obstructive pulmonary disease (COPD), unlike many chronic diseases such as cerebrovascular disease and myocardial infarction, is rising. It is predicted to become the fifth leading cause of morbidity and mortality over the next 15–20 years.[1] Despite this, there is still no specific therapy for COPD, and treatment is largely restricted to bronchodilators, and treatment of complications with antibiotics, steroids, diuretics and oxygen.

Pathophysiology

For many years the term COPD was considered synonymous with the pathological changes of emphysema. It was well recognised that cigarette smoke was a major contributing factor, but the physiological processes that led to the development of emphysema, resulting in contact with the healthcare system because of symptoms, remained obscure. However, in 1963 Laurell and Eriksson noted a small number of subjects with a missing or faint protein band seen in the alpha-1 region on paper electrophoresis.[2] Clinical assessment of the patients indicated that three had severe early onset emphysema, suggesting a cause and effect. The missing protein was shown to be the major serum inhibitor of the proteinase trypsin, and hence it was labelled alpha-1 antitrypsin (AAT). The serum deficiency was shown to be inherited in an 'apparently' autosomal recessive fashion and was commonly associated with lung disease.[3] Subsequent studies indicated that the affected individuals had a reduced life expectancy, particularly if they smoked.[4]

At this time, animal studies indicated that installation of proteolytic enzymes into the lungs of experimental animals induced pathological changes typical of emphysema.[5] Because AAT was an inhibitor of proteolytic enzymes it was proposed that an enzyme or enzymes normally inactivated by AAT was directly responsible for lung damage leading to emphysema (the proteinase/antiproteinase hypothesis).

The mechanism of serum deficiency of AAT has been elucidated in recent years. It was known that the protein was retained in the liver as periodic acid Schiff (PAS) positive inclusions and was associated with the development of adult cirrhosis[6] and hepatocellular cancer.[7] The common Z variant of the protein which leads to serum deficiency undergoes spontaneous polymerisation as a result of changes in its tertiary structure. This results in polymerisation

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Key Points

Bacteria are frequently isolated from sputum of patients with chronic obstructive pulmonary disease in the stable clinical state

The bacterial load is often low and as the load increases neutrophilic inflammation is generated

High neutrophilic inflammation is characterised by sputum purulence

The presence of sputum purulence reflects likelihood of isolating potentially pathogenic bacteria and the size of the airway bacterial load

New or increased sputum purulence reflects the likelihood that antibiotic therapy is needed when exacerbations of symptoms occurs.

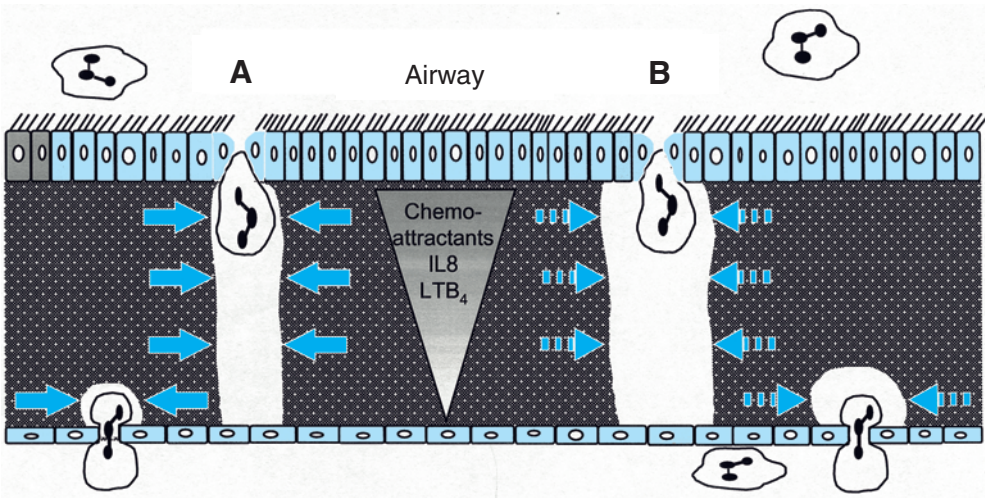

Fig 1. Neutrophil recruitment from the circulation in response to specific chemo-attractants in the airway results in degranulation. Because of the high concentrations of enzyme being released close to the cell, an area of obligate connective tissue destruction takes place **(A)**. This is closely controlled by the natural proteinase inhibitors, especially alpha-1 antitrypsin. In patients with alpha-1 antitrypsin deficiency, the area of obligate proteolysis is greatly increased **(B)**, resulting in more extensive damage and rapid progression of lung disease.

in the hepatocyte, and hence retention rather than secretion.[8] The protein has also been shown to be the major serum inhibitor of neutrophil elastase[9] which is stored in and released from mature neutrophils when activated. This enzyme was shown subsequently to produce emphysema in animal models[10] and became implicated as the cause of emphysema in man.

Direct evidence of the role of neutrophil elastase (NE) in the pathogenesis of emphysema in man awaits clinical trials of specific inhibitors. However, the neutrophil is a major inflammatory cell in the lungs of smokers and those with emphysema, especially in subjects with serum deficiency of AAT.[11] Furthermore, immuno-histochemistry has shown that the amount of NE correlates with the severity of emphysematous change.[12] Finally, the NE knockout mouse is protected against the development of emphysema following smoke exposure.[13]

The exact mechanisms involved are gradually becoming elucidated. Firstly, it is critical to damage the connective tissue framework of the lung, and this must occur during neutrophil activation and migration. The reason for this latter statement relates to the process of degranulation and release of NE by the neutrophil and its control in close proximity to the activated cell.

The enzyme is stored at a concentration of about 5 mmol in the granule. Once released, the enzyme diffuses away and its concentration decreases exponentially. When the concentration equals that of the surrounding inhibitors the enzyme is irreversibly inactivated.[14] Thus, in the presence of normal AAT (with a serum concentration of 30 μmol), the enzyme will only remain active over a short distance close to the granule as the enzyme is released. However, this results in an area of obligate proteolysis even in normal subjects which may be vital to enable the cell to penetrate the tight connective tissue matrix and yet damages the tissue at the same time. However, in subjects with AAT deficiency the area of obligate proteolysis is much wider, leading to greater destruction and (by implication) the increased likelihood of development of emphysema. This is summarised in Fig 1 and explains why subjects with AAT deficiency develop more severe disease at an earlier age than non-deficient subjects. However, the reason this only results in disease in some smokers (even with AAT deficiency) still needs to be resolved, but may relate to other factors modulating the degree of neutrophilic inflammation in man.[15]

In recent years, it has become clearer that not all patients with

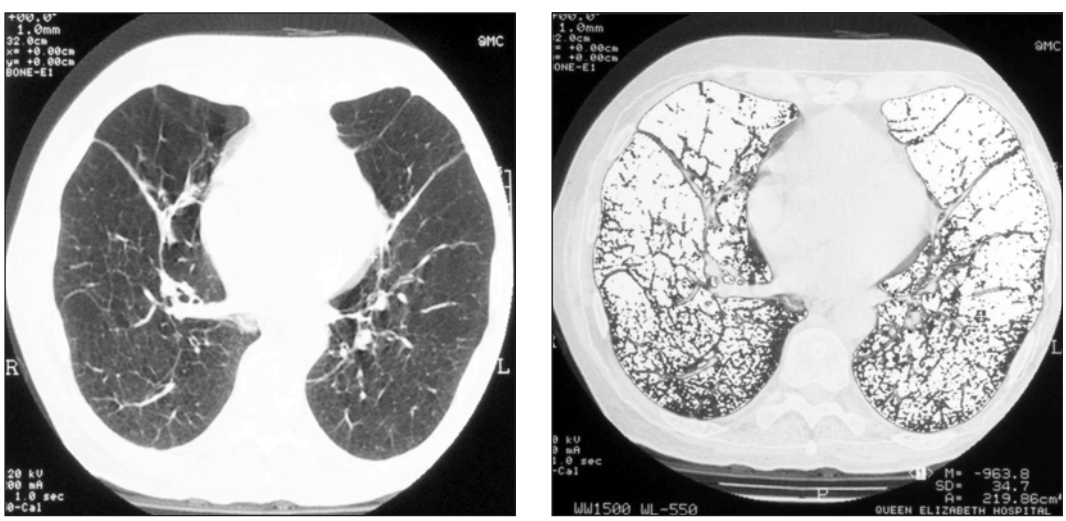

Fig 2. High-resolution CT scan of the chest in a patient with emphysema. Low-density areas (highlighted on the right) indicate the regions of most damage.

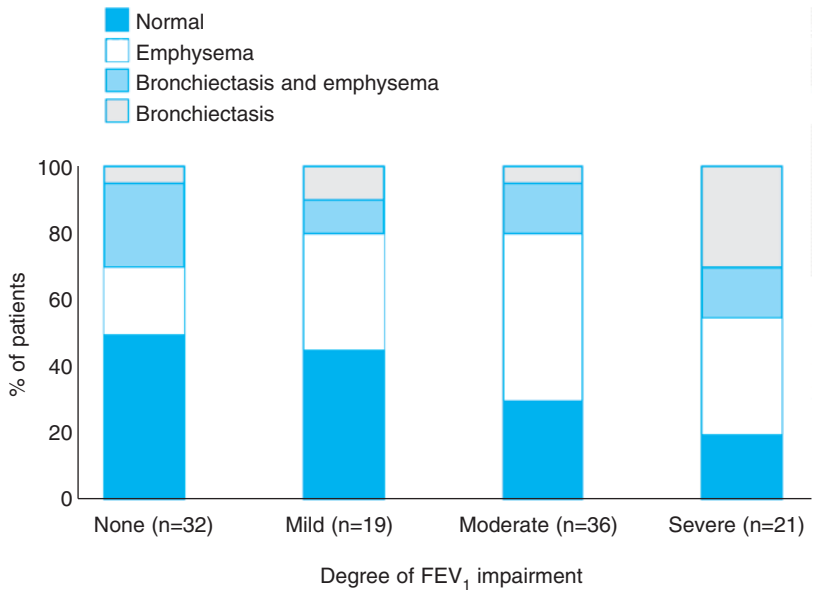

Fig 3. The proportion of patients in whom the CT scan is macroscopically normal, shows areas of emphysema, shows areas of emphysema with bronchiectasis, and shows bronchiectasis alone is indicated. Patients with chronic bronchitis and airflow obstruction are divided into those who have an FEV_1 within the normal range, and those with mild, moderate and severe impairment. FEV_1 = forced expired volume in one second.

COPD have emphysema,[16] even those with AAT deficiency. COPD is characterised by air-flow obstruction and destruction of alveoli is only one cause. High-resolution computed tomography (CT) scan (Fig 2) is now the gold standard for identifying emphysema in life as low-density areas or even bullae. In one study, it became clear that although emphysema may be present in up to 50% of patients with a diagnosis of COPD,[16] up to 20% may have radiologically normal lungs, even when air-flow obstruction is severe (Fig 3) although bronchiectasis becomes more common.

Nevertheless, it is also likely that NE plays a role in the airways of these patients. The enzyme can cause mucous gland hyperplasia,[17] mucus secretion,[18] and both reduce ciliary beat frequency[19] and damage the airway epithelium.[20] These changes would provide a major breach to the most important primary defence system in the lung, the mucociliary escalator. This would facilitate bacterial colonisation and the mucus would provide a potential culture medium for bacterial growth. The effect would be a driving force for further inflammation[21] and exacerbations[22] caused by bacteria.

Exacerbations

Exacerbations of COPD are episodes when the patient notices a deterioration in their symptoms that is unusual and often requires intervention. The importance of these episodes has been highlighted because of their marked effect on patient health status[23] and influence on lung function deterioration,[24,25] especially in continuing smokers.[26] It is not unreasonable to assume that neutrophilic responses to bacterial infection could add to the lung damage by the collateral effects of NE released from the activated cells as they are recruited to the lung. However, there has been some disagreement concerning the role of bacteria in exacerbations of COPD. This controversy relates to several factors:

- Exacerbations are 'poorly defined', patient-led events.
- Many symptoms can deteriorate, including the classical ones of dyspnoea, cough and sputum production, or less specific ones like tiredness and upper respiratory tract symptoms (sore throat, 'cold'), even in the absence of bacteria.
- Symptoms can be worsened by the weather, pollution,[27] viral infections[28] and bronchospasm.
- Antibiotic studies are not convincing although meta-analysis does confirm an effect, albeit small.[29]
- Bacteria often colonise the airway when patients appear to be clinically stable.[30]

Despite these issues, recent studies have helped elucidate the role of bacteria in the airway of COPD patients.

In order to dissect this problem, the relationship between the lung defences and bacteria needs to be understood. Bacteria are inhaled continually and primary host defences, including bacteriostatic proteins such as lysozyme, immunoglobulins (especially IgA) and the mucociliary escalator, are all active in removing bacteria. In addition, airway macrophages continually survey the environment and ingest remaining airway particles, including bacteria.

When this system is breached (immune deficiencies and ciliary dysfunction or an increase in bacterial load), the primary system can no longer retain sterility. As bacterial numbers rise, the airway macrophages and airway epithelium release pro-inflammatory cytokines. These include TNF alpha[31] and IL1B that upregulate endothelial adhesion molecules,[32] and the neutrophil chemoattractants IL-8[33] and particularly LTB4.[34] The net effect is inflammation and particularly neutrophil accumulation.

This concept is supported by animal experiments in which the infusion of bacteria into the airway results in little inflammation and sterilisation when the load is low, but neutrophilic inflammation and bacterial replication when the load is high.[35] Studies in man confirm that bacterial loads $<10^6$ colony-forming units

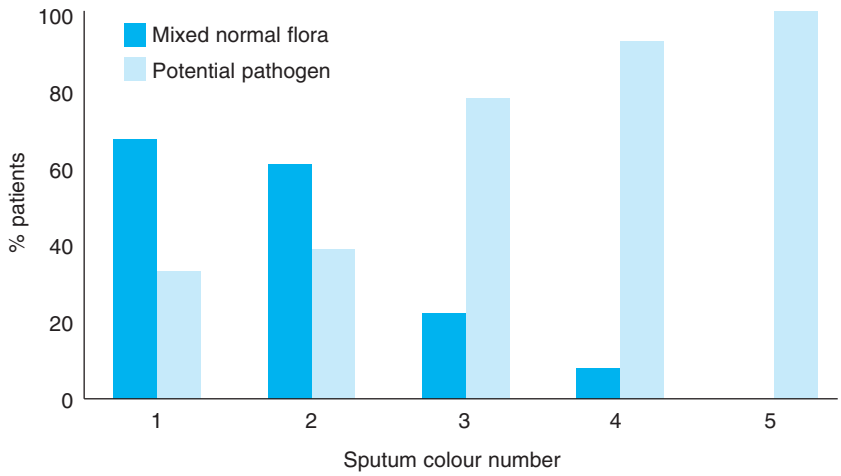

Fig 4. The vertical axis indicates the likelihood of isolating potentially pathogenic bacteria from sputum samples assessed according to sputum colour. Samples with a colour of 3 or greater indicate increasing sputum purulence, whereas those below 3 are essentially mucoid.

(cfu/ml) are not associated with neutrophilic inflammation (a stable colonisation). On the other hand, bacterial loads of $>10^6$ cfu/ml are associated with increasing neutrophil influx as the chemo-attractant cytokines rise.[21]

Studies during exacerbations have shown (using protected brush specimens obtained from the lower airway) that the likelihood of isolation of bacteria is increased and the numbers are greater than in the stable state.[22] Thus antibiotics would seem a logical and reasonable form of therapy during such episodes. However, many controlled trials of antibiotic therapy for exacerbations show no or little advantage for treatment, although meta-analysis suggests a small benefit.[29]

It is easy to understand the minimal benefit of antibiotics since the episodes have been poorly defined in the past and non-bacterial causes of the same symptoms confuse the picture. The best controlled trial in 1987[36] classified the episode according to the presenting symptoms. The three key features were increased breathlessness, increased sputum volume and new or increased sputum purulence (although minor symptoms also occurred and were documented). Although the overall result proved the efficacy of antibiotics, it was only those with all three symptoms who showed clear benefit. The key symptom would be sputum purulence, which has been a subjective description of the yellow/green colour of the expectorated sample.

The neutrophil is a green cell as a result of its myeloperoxidase content. Hence increased neutrophil content can be perceived as a yellow/green colouration reflected in purulence. Indeed, the myeloperoxidase content of sputum is dependent upon bacterial load[21] and when samples are classified according to a graded colour chart,[37] it reflects the likelihood of isolating bacteria (Fig 4). Thus clinical observation is consistent with both animal studies[35] and human studies[21] that relate neutrophilic inflammation to bacterial load and the clear positive benefit of antibiotics when sputum purulence is present.[30,36]

Using sputum purulence (assessed by the graded colour chart) as a defining characteristic enabled us to separate exacerbation into those where the sputum was mucoid or purulent at presentation.[30] The episodes with mucoid sputum did not contain organisms more frequently or in higher numbers than in the stable state.[30] Neither were those episodes associated with evidence of neutrophilic inflammation or increased cytokine release.[37] On the other hand, purulent episodes were associated

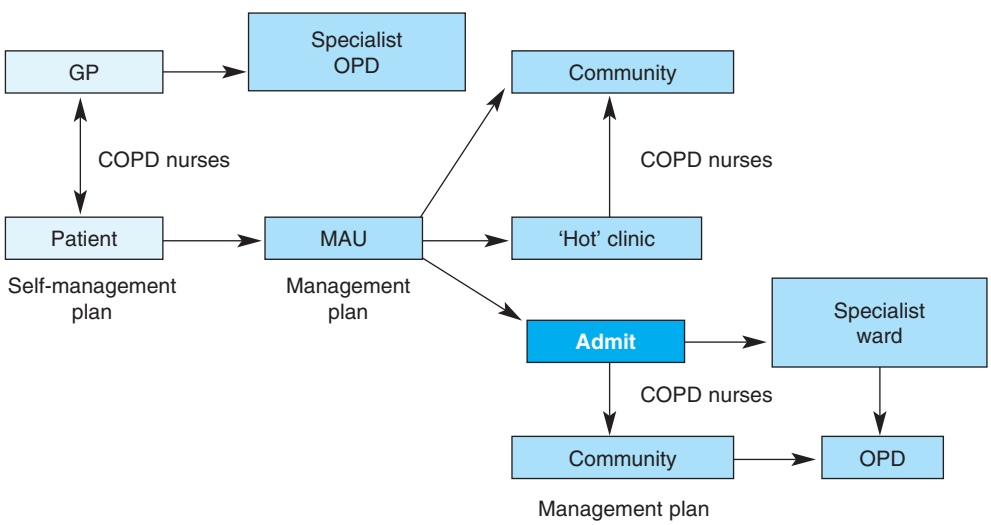

Fig 5. An abbreviated integrated care pathway indicating intervention points as part of clinical and self-management plans, where observations of sputum colour can influence antibiotic prescribing as part of management plans. The overall purpose enables rational antibiotic prescribing and where possible, prevention of admission. OPD = outpatient department; MAU = medical admissions unit; 'Hot' = acute follow-up.

with a high isolation rate of bacteria, marked neutrophilic inflammation and cytokine release including the presence of free NE activity,[30] and high bacterial numbers which disappeared or decreased as the episode resolved.[30,38]

Thus, this simple observation of sputum purulence provides good evidence of bacterial-induced inflammation and subsequent NE release (that potentially causes further lung damage). It is likely that such episodes with NE release are the major reason that declining lung function relates to exacerbation frequency.[24,25]

Clearly, further studies are indicated but they should be facilitated by the agreement on a definition of an exacerbation.[39] The episodes are 'an acute deterioration in symptoms beyond the normal daily variation that requires a change in therapy'. With this definition, the episodes can be identified and at least the purulent episodes characterised by a matching colour chart.[30]

The success of the colour chart in identifying purulent (and hence bacterial episodes) enables patients and doctors to identify the need[30] and response[37] to antibiotics and is currently being used in both self-management plans and hospital inpatient management. The intervention points on an abbreviated integrated care pathway are shown in Fig 5. Whether these episodes and their correct management influences lung function decline remains to be determined.

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