

Intermediate Alpha-1 Antitrypsin Deficiency Can Play a Role in Pulmonary Exacerbation?

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Abstract: Background: Alpha-1 antitrypsin deficiency is generally suspected in young patients with pulmonary emphysema or chronic obstructive pulmonary disease (COPD). Patients often suffer from diagnostic delays or are misdiagnosed, for example, with COPD, asthma, or airway hyperresponsiveness because of the nonspecific nature of respiratory symptoms recognised with Alpha-1 antitrypsin deficiency (AATD). These pathologies develop in homozygous patients (both compromised alleles) with severely deficient protein; however, they are also frequently observed in heterozygous patients (only one compromised allele) for the gene mutation with a more or less deficient protein and functional anatomical damage of varying severity depending on the type of mutation and the exposure to environmental risk factors and/or professional that can trigger the repeated injurious inflammatory process. **Case Description:** We describe two cases of late diagnosis of alpha-1 antitrypsin deficiency, with many exacerbations and intermediate level of alpha-1 antitrypsin. Due to the peculiar clinical history, and the P_{Lowell} rare mutation, although intermediate AATD, the patients were subjected to replacement therapy and they obtained clinical improvement. **Discussion:** Both the cases carried a heterozygous P_{Lowell} mutation representing two interesting and rare examples of clinical cases with double heterozygosity. The presence in the other AAT allele of the S-mutation in the first case and a concomitant presence of another mutation in the cystic fibrosis gene in the second case contributed to the protease-antiprotease imbalance and, despite intermediate AATD, was the probable cause of the numerous exacerbations. **Conclusion:** Alpha-1 antitrypsin deficiency should always be suspected in patients with respiratory disease and an unclear or complex clinical history. It may be useful to recognize and evaluate treatment even outside the established parameters, in selected cases.

Keywords: AATD, COPD, Respiratory, Deficiency, Antitrypsin.

INTRODUCTION

Alpha-1 antitrypsin deficiency (AATD) causes different respiratory manifestations, such as pulmonary emphysema, chronic obstructive pulmonary disease (COPD), asthma and bronchiectasis. AATD is a progressive lung disease, and early diagnosis allows patients to perform lifestyle changes and begin therapy options that preserve respiratory function and avoid hospitalizations. Unfortunately, AATD is underdiagnosed; evidence from screening programmes in the USA advised that fewer than 10% of patients have been diagnosed, and similar under-diagnosed conditions are also present in Europe¹. Patients often suffer from diagnostic delays or are misdiagnosed, for example, with COPD, asthma, or airway hyperresponsiveness (AHR) due to the nonspecific nature of respiratory symptoms recognised with AATD. It has been suggested that AHR and wheezing may be susceptibility phenotypes for more critical lung outcomes, particularly in the setting of cigarette smoking². Generally, homozygous patients (both compromised alleles) with severely deficient protein develop respiratory disease, however, they are also frequently observed in heterozygous patients (only one compromised allele - intermediate AATD) for the gene mutation with a more or less deficient protein and functional anatomical damage of varying severity depending on the type of mutation and the exposure to environmental and/or professional risk factors that can trigger the repeated injurious inflammatory process. Carefully designed family studies show an increased

risk of emphysema in smokers with intermediate AATD. The risk of asthma in intermediate AATD subjects is less studied, and more literature is needed before firm conclusions can be made.

Recently it was shown that, beyond antiprotease activity, AAT is also characterized by anti-inflammatory and immunoregulatory features³. In this context, the presence of AATD, also in the heterozygous forms, associated with other inflammatory conditions (such as rheumatoid arthritis, diabetes, cystic fibrosis and asthma) should be considered⁴.

CASE 1

We describe a 69-year-old patient, a female, housewife, who had never smoked. The patient reported over the previous 10 years six episodes of exacerbation, documented each year. The exacerbations were characterized by coughing, increased sputum and worsening of dyspnoea. The color of the sputum varied from yellow to green and, in both cases, if antibiotic and anti-inflammatory therapy was not started early, fever and chest pains appeared. In some cases, shortness of breath appears even at rest, so severe that it requires hospitalization. Respiratory symptoms were resolved with steroids and antibiotics. The patient had been hospitalised twice in the last three years for acute respiratory failure.

For the persistence of respiratory symptoms characterised by dyspnea and a dry cough, the patient attended the pulmonary clinic of our department, and a functional assessment was carried out. At a young age,

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she reported occasional wheezing, sporadic episodes of respiratory exacerbation especially during the three pregnancies, intense dyspnea, and asthma-like symptoms. She had no previous documentation about respiratory function. Therapy was already maximized (inhaled corticosteroid/long-acting β_2 -agonist twice daily, long-acting muscarinic antagonist twice daily, oral theophylline 200 mg bis in die, oral montelukast 10 mg/day).

The patient complained of worsening dyspnea with wheezing. Spirometry performed at the first visit showed forced vital capacity (FVC) 2.17 (120%), forced expiratory volume in the 1st second (FEV1) 1.64 (113%) and diffusing capacity of the lungs for carbon monoxide (DLCO) 4.11 mmol/(minKPa), 80%. Spirometry performed showed FVC 1.95 (100%), FEV1 1.44 (100%), Tiffenau index was 75% and DLCO.11 mmol/(minKPa), 80%. She was subjected to a methacholine challenge test, which is positive for severe hyperreactivity. Allergy screening was negative. Blood gas analysis values were normal. Sputum cultures for common bacteria, fungi, Kooch's bacillus, and atypical bacteria were negative. The patient also underwent gastroscopy and an esophageal ph-meter to rule out gastroesophageal pathology.

The blood tests were normal, except for the alpha-1 antitrypsin in serum of 67 mg/L (normal value is from 0.90 to 2.00 g/L). The chest computed tomography (CT) revealed small areas of air trapping and diffuse bronchiectasis. Genetic screening was performed for the AATD congenital and showed compound heterozygosity for deficient alleles PiS (p.Glu288Val c.863A>T) and P_{Lowell} (p.Asp280Val c.839A>T).

After the informed consent of the patient, we started the augmentation treatment with human alpha-1 antitrypsin with a dose of 60 mg/kg every 7 days. At the request of the patient and her family after having monitored serum alpha-1 antitrypsin deficiency, therapy was performed every 14 days. After treatment, the patient showed no exacerbation, dyspnea exertional resolution and stable improvement in respiratory parameters.

The patient, with S mutation and S_{Lowell}, a rare mutation in heterozygous, with an intermediate level of AAT, treated with augmentation therapy, obtained a clinical and functional improvement without exacerbation in 3 years of treatment. Table 1 shows ACT pre and after augmentation therapy, the number of exacerbations per year, MMRC and 6MWT before and after 3 years of treatment.

Table 1: ACT pre and after augmentation therapy for patient 1. In the table we see the number of exacerbations, MMRC and 6MWT before and after 3 years of treatment.

Pt 1	ACT	Exacerbation/y	MMRC	Mt 6MWT
pre	24	6	2	340
post	6	0	1	420

CASE 2

The second case is a 56-year-old Caucasian man, night watchman, former smoker 30 pack/ys (8 years ago, he quit smoking after a severe exacerbation with hospitalization). He was diagnosed with COPD with 8 bronchial exacerbations per year in the last 7 years, treated with therapy in first aid, two hospitalizations, one episode of acute respiratory failure in the course of an exacerbation.

The patient also suffered from type II diabetes, hepatic and pancreatic pseudocysts, and two episodes of acute pancreatitis about a year ago. A heterozygous mutation for F508del was found out in the cystic fibrosis gene, the patient was being treated with pancreatic enzymes. The symptoms complained over the last few months were a worsening of dyspnea during daily activities, cough with mucous expectoration, asthenia and easy fatigue. The patient was treated with inhaled corticosteroids/long-acting beta2-agonists (ICS/LABA) bis in die, long-acting muscarinic antagonists (LAMA) bis in die.

The respiratory function tests showed a moderate obstructive disventilatory syndrome not reversible after salbutamol test (FVC 3.37 L - 77%; FEV1 2.05 L - 60%, FEV1/FVC 63.38%, DLCO 72%, VR (115%). The basal peripheral saturation at rest was 95%, blood gas analysis was normal, COPD assessment test (CAT) 26, MMRC 3. Allergy tests were negative. Serum alpha-1 antitrypsin assay with nephelometry 78 mg/dL (v.n. 90-200), C-reactive protein (CRP) 0,2 mg/dL (v.n. 0.1-0.3). High-resolution chest CT scan was performed showing central lobular emphysema dominant in the lower lobes and small bronchiectasis on both sides.

It was performed a genetic investigation for alpha-1 antitrypsin deficiency and the mutation p.Asp280Val c.839A> T, PI P_{Lowell} in exon 3 of the serpin1 gene was found in heterozygosity.

The patient underwent a 6 months follow-up period during which he presented 3 further exacerbations. After collegial discussion, further clinical monitoring and informed consent, we started augmentation therapy with human alpha-1 antitrypsin with a dose of 60 mg/kg every 7 days.

Currently, the patient has been on augmentation therapy for 3 years, with only one exacerbation reported. The respiratory function tests show a moderate grade obstructive disventilatory syndrome that is not reversible after salbutamol tests (FVC 3.38 L - 77%; FEV1 2.18 L - 65%, FEV1 / FVC 62.29%). The basal peripheral saturation at rest 95%, blood gas analysis normal, CAT 7, MMRC 2 (Table 2)

Table 2: CAT pre and after augmentation therapy for patient 2. The number of exacerbations /years before and after 3 years of treatment.

Pt 2	CAT	Exacerbation/y	MMRC	Mt 6MWT
pre	26	8	3	306
post	7	1	2	390

DISCUSSION

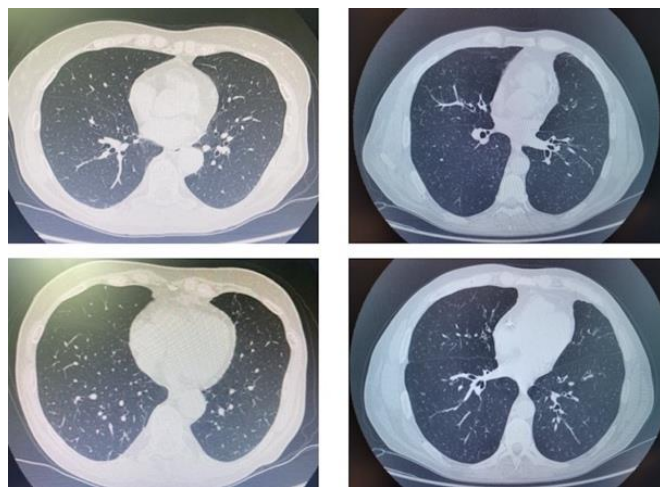


Figure 1: Chest CT scan showed the presence of small bronchiectasis in case 1 (on the left) and case 2 (on the right).

A review by Eden (2010) showed that subjects with AATD are particularly predisposed to AHR, which is associated with the reactivity of bronchodilators, asthma, and allergies⁴. The various asthma phenotypes develop from a synergy between heredity and polygenic environment complex. Atopic asthma is correlated with allergic manifestations, skin test response, eczema, and rhinitis and has triggers for attacks of wheezing and shortness of breath⁵. Cigarette smoke and gastroesophageal reflux can trigger AHR. When AATD contributes to the pathogenesis of asthma, then the lack of airway anti-neutrophil elastase will set up conditions of inflammation alternatives to AHR⁶. Under conditions of neutrophil airway inflammation in asthma and chronic bronchitis, both total and active pro-inflammatory elastase are developed in airways. There is a compensatory development in airway AAT, but the excess of elastase indicates protease anti-protease imbalance⁷. Some studies have revealed that asthma is also a risk factor for an accelerated decline in FEV1^{8,9}. Therefore, the anti-inflammatory and immunomodulatory properties of the AAT should be considered.

For many years, the patients had respiratory symptoms diagnosed as episodes of acute or chronic bronchitis. Although there were no professional and lifestyle risk factors, and patient 1 had never smoked, alpha-1 deficiency was never suspected. Patient 2 was a former smoker (30 packs/ys) but at 46 years he began to suffer from respiratory disorders, and he was never subjected to AAT dosage. Despite the maximisation of drug treatment, the clinical conditions worsened with an increase in the frequency of episodes of bronchial exacerbation. In addition to this, and despite the maximization of treatment, clinical conditions worsened with an increase in the frequency of episodes of bronchial exacerbation; in the last years, exertional dyspnea had arisen, and the patients were subjected to hospitalizations for the exacerbations.

At the time of our first evaluation, the finding of a lower than usual serum alpha-1 antitrypsin dosage required a genetic study documenting the presence in the compound of a PiS and P_{Lowell} mutation for patient 1 and heterozygosity of P_{Lowell} mutation for patient 2.

The P_{Lowell} variant has been found relatively frequently in the European population¹⁰. It has been observed that the condition, even in heterozygosity, is associated with increased degradation of the synthesized levels of alpha-1 antitrypsin, with a reduction in the circulating levels of alpha-1 protease inhibitor. In a work by Bornhorst, it was found that the P_{Lowell} variant is variable and significantly reduces the observed concentrations of AAT. In individuals with the heterozygous M/P_{Lowell} genotype, the concentration was 100 mg/dL, considering that the median reference range of 95% of the native MM genotype is 100-250 mg/dL¹¹.

It is estimated that functional AAT concentrations of 65 mg/dL prevent the progression of lung disorders. However, an intermediate risk range of developing lung disease is recognized at doses above 113 mg/dL. But, in these clinical cases, more than a decreased serum AAT concentration, the influence of several genetic and non-genetic factors seems to be relevant. An increased risk of developing lung disease may be related to genes and gene interactions that we not aware of.

Both the cases carried a heterozygous P_{Lowell} mutation representing two interesting and rare examples of clinical cases with double heterozygosity. The presence in the other AAT allele of the S-mutation in the first case and a concomitant presence of another mutation in the cystic fibrosis gene in the second case underline the involvement of a protease-antiprotease pathological effect which justifies the use of the augmentation therapy.

After therapy, our patients did not show any bronchial exacerbation; they received an increase in the quality of dyspnea assessed with the index of metres covered by the six-minute walking test, MMRC, asthma control test (ACT), CAT test.

The possibility that a deficiency of alpha-1 antitrypsin can act as a factor favouring the onset of pancreatitis finds some supporting cases reported in the literature; in fact, AAT not only inhibits elastase but also pancreatic trypsin and many other proteases, in particular those released from the primary granules of neutrophils¹². However, the possible interaction between the two genes remains controversial in the literature¹³.

The patients both had P_{Lowell} mutation in heterozygosity and a clinical history characterized by frequent exacerbations and hospitalizations. Patient 1 had no known risk factors for developing the respiratory disease while Patient 2 was a former smoker. This could explain the different age of the onset of symptoms and especially the different respiratory function at the time of diagnosis.

The evolution of lung disease in people with AAT deficiency would appear to be aggravated by gene

interactions that are not yet clearly known. Presumably, atopy, modifier genes and extrinsic factors such as environment and cigarette smoke, acting through the development of AHR and asthma accelerate lung function decline in AATD and play a role in exacerbation.

The role of the different mutations of the alpha-1 antitrypsin gene is subjected to discussion and contradictory opinions. Further considerations should then be made on the possibility that the same allelic combination gives rise to different phenotypes, a phenomenon known as variable expressivity and incomplete penetrance. These events occur more typically in autosomal dominant inherited pathologies but may also appear in autosomal recessive, mitochondrial and X-linked pathologies. When there is a very suspicious clinical picture or a young patient, anyway, a mutation for the alpha-1 antitrypsin gene should be excluded. Therefore, a combination of laboratory methods is necessary. In particular, the serum level has only a very limited value in heterozygous carriers and, depending on the state of the inflammatory progression, may change. At a molecular level, rare mutations non-S or non-Z have been traditionally not considered, but the increasing number of novel allelic deficient variants challenges this concept¹⁴

The role of the distinct mutations of the gene for alpha-1 antitrypsin and their combinations is still an ongoing study. Different clinical pictures of different severity are described in the literature in patients with the same mutation or with rare mutations in combination.

Serum alpha-1 antitrypsin dosage should be performed for all asthma and COPD patients at least once in life, as suggested by the WHO¹⁵ nevertheless a large number of patients receive the diagnosis late¹⁶. The latency time for achieving the diagnosis of congenital alpha-1 antitrypsin deficiency is higher than 10 years. The diagnostic delay, from the registry data, is about 8–9 years and is higher than the data known for the United States and Germany (about 6 years)¹⁷. A therapeutic and diagnostic delay can lead to a complex clinical course.

We suggest to always suspect AAT deficiency in the presence of respiratory symptoms at any age and above all to increase greater awareness of this genetic condition, which is still under-recognised over 50 years after its discovery.

Also, we suggest the evaluation of each case for the possible indication and potential advantages of the augmentation treatment. Augmentation therapy has been reported to improve survival in patients with AADT, regardless of FEV1.¹⁸

Alpha-1 Foundation recommends intravenous augmentation therapy for individuals with AATD and an FEV1 in the range of 30%-65%¹⁹. Canadian thoracic society recommends AAT augmentation therapy may be considered in non-smoking or ex-smoking patients

with FEV1 25 - 80% of predicted.²⁰ There are recommendations from the various guidelines often differed. Variation in recommendations may reflect differing prevalence rates of AATD and prevalence of different variants of AAT about which little is known today.²¹ In the last years, evidence is emerging, demonstrating that there may be value in recognising and treating patients outside of the established parameters²². Many testing initiatives are ongoing to improve the discovery of broader genotypes containing new loss alleles currently being identified. Large-scale screenings in general populations, students, newborns and blood donors, as well as new case-finding approaches that target patients with COPD and asthma, will aid in the identification of new patients in need of treatment. Although AAT replacement therapy is the only treatment available that addresses the underlying cause of AATD, symptomatic treatment and lifestyle changes can benefit all patients.

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