Inhaled dose was similar between devices, 28.7% (22.7; 33.5) vs 29.3% of ND (26.3; 33.1) for ISO-NEB® and for RespirgardII® ($p = 0.792$). Residual volume was 0.9313 g (0.9270; 0.9382) and 1.4087 g (1.3845; 1.4416) for ISO-NEB® and for RespirgardII®.

In vivo, all volunteers (23.5 +/− 1.3 years) had spirometric values in the normal range. Cu max was similar between devices with 3.5% (3.1; 3.6) and 3.6% of ND (2.2; 4.2), ($p = 0.893$) for ISO-NEB® and RespirgardII®, respectively. Urine volume was 1.37 L (0.80; 1.72) and 1.30 L (0.75; 2.10) for ISO-NEB® and RespirgardII®, respectively ($p = 0.686$). Elimination constant (K_e) of the drug following nebulization was similar for both devices (0.159 (0.078; 0.208) vs 0.130 (0.085; 0.162) for ISO-NEB® and for RespirgardII®, respectively ($p = 0.225$)). There was no significant difference in RF, Vt and VE. Our results were in the range of previous studies even if the comparison is difficult because there were no data for ISO-NEB® and the previous studies on RespirgardII® presented many differences in protocols and measurements techniques.[6,9,12,13] We used the most frequent dosage reported in the previous studies and in the manufacturer's recommendations (300 mg pentamidine in 6 mL sterile water).[1,2,7,9] The airflow rate (8 L min^{-1}) to produce aerosolized pentamidine was in the range of the rate described in previous studies about RespirgardII® (about 6–10 L min^{-1} for RespirgardII®) or recommended by the manufacturer for ISO-NEB® (from 5 to 9 L min^{-1}). The total amount of drug reaching the lungs was similar and lower than 5% of ND for both devices. Our results are in line with those obtained with pentamidine in prior studies (2–6.74% of ND).[4,6,9,12] Some methodological conditions need to be discussed. In vitro, we used the residual gravimetric technique even though it was not validated for pentamidine. However previous studies validated this process for different drugs.[14] In vivo, we used amikacin sulfate because pentamidine has not been previously considered as a valid pharmacological marker of pulmonary deposition.[11,15] For a methodological consideration, we recruited only male subjects because it is easier for a man to collect his urine without loss and to prevent potential fetal risk ototoxicity in pregnant female subjects. Finally, we did not measure the particle sizes for both nebulizers, but according to previous studies and manufacturer's data we can consider that they were similar (1–2 μm). It is important to notice that the two nebulizers are in the same price range. In conclusion, this in vitro and in vivo study demonstrated that ISO-NEB® and RespirgardII® have similar properties in the conditions study. Further clinical studies are needed to confirm that ISO-NEB® is a valuable alternative to the reference nebulizer recommended by guidelines for pentamidine delivery. Altogether these data suggest that the performance of both devices is similar in the conditions of this in vitro and in vivo study.

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1579-2129/

Mycobacterium xenopi and Squamous Cell Carcinoma of the Lung☆

Mycobacterium xenopi *y carcinoma pulmonar de células escamosas*

Dear Editor,

Non-tuberculous mycobacteria (NTM) or atypical mycobacteria are aerobic bacteria of the genus *Mycobacterium*, the pathogenic potential of which has been known since the 1950s.[1] The AIDS pandemic, the progressive increase in immunosuppressive states, and the improvement of microbiological techniques have made isolation of these microorganisms more common nowadays.[2]

Mycobacterium xenopi (M. xenopi) is a NTM associated with water systems that is found primarily in North America, the south

☆ Please cite this article as: Martín Asenjo M, Martín Guerra JM, López Pedreira MR, Prieto de Paula JM. *Mycobacterium xenopi* y carcinoma pulmonar de células escamosas. Arch Bronconeumol. 2017;53:698–700.

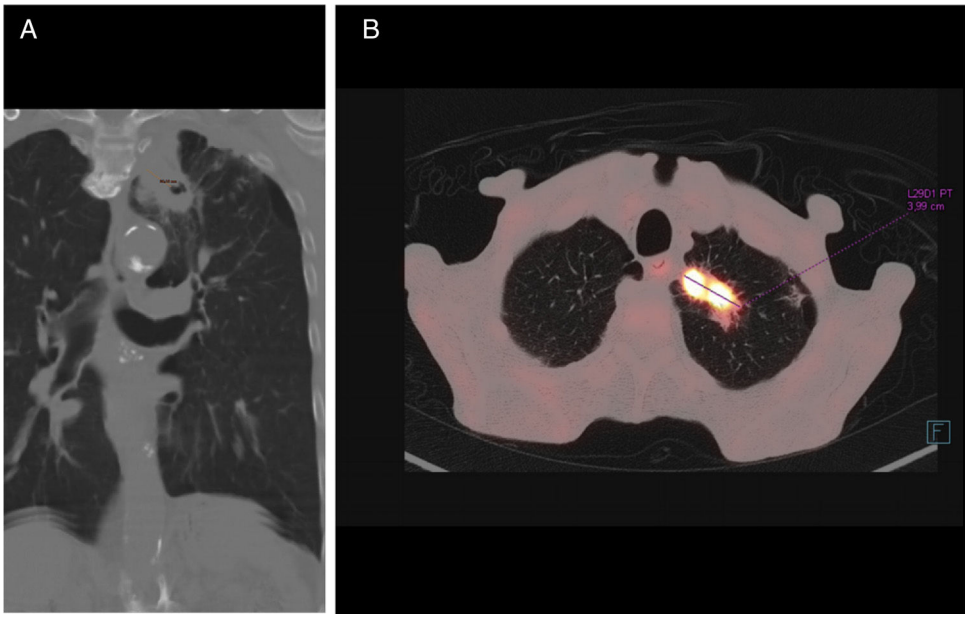

Fig. 1. (A) Coronal CT with lung window: cavitating mass with thick irregular wall, 18 mm in the left upper lobe extending toward the mediastinum. (B) Axial PET-CT: mass showing high uptake of fluorodeoxyglucose, with a standard uptake value of 28.38 units.

east of Great Britain, and the north of France,[3] and was first isolated in immunosuppressed patients. The main risk factors for the disease are chronic lung diseases, during which the organism can colonize the respiratory tract.[4] Cases of mycobacterial infection have been published in cancer patients,[5] but cases involving *M. xenopi* are exceptional. No references to the subject were found in articles retrieved from a literature for articles published in Spanish using the standard search engines, Medline and Pubmed (key words: *Mycobacterium xenopi* and *lung cancer*). We report a case of *M. xenopi* infection in a patient with severe COPD and a diagnosis of squamous cell carcinoma.

This was a 73-year-old man, active smoker, diagnosed with severe COPD in 2006. He consulted for asthenia, epigastric pain, weight loss, and productive cough without fever. The only relevant finding on physical examination was poor nutritional status. Clinical laboratory test results showed normochromic normocytic anemia (hemoglobin 10.8 g/dl), with ESR 46 mm and ferritin 9.4 ng/ml (normal value: 30–400 ng/ml). Chest radiograph revealed an infiltrate in the left upper lobe (LUL), disperse granulomas, and bilateral air trapping. Gastrointestinal endoscopy showed *Candida* esophagitis, erosive duodenitis with a negative urease test, and diverticula in the colon. Acid-fast bacilli were observed in 3 sputum samples, so treatment was initiated with isoniazid, rifampicin, pyrazinamide, and ethambutol, in addition to oral iron and fluconazole. After culture of 3 sputum samples in Löwenstein medium grew *M. xenopi*, the antimicrobial therapy was adjusted, and treatment began with clarithromycin, rifampicin, and ethambutol.

Chest computed tomography (CT) (Fig. 1) confirmed the existence of a solid lesion with a spiculated border and central cavitation in the LUL, measuring 40×35 mm, with an 18 mm-thick wall, contiguous with another paramediastinal lesion measuring 3 cm, signs of emphysema and multiple calcified granulomas. No endoluminal lesions or changes in the mucosa were seen on bronchoscopy. PET-CT confirmed a hypermetabolic mass in the left lung apex, with the appearance of a malignant lung tumor. Transbronchial biopsy yielded a diagnosis of squamous cell carcinoma.

M. xenopi infections usually occur with nonspecific symptoms such as dyspnea, cough, and weight loss,[6] and primarily affect males with COPD.[7,8] Radiological changes are wide-ranging and usually persistent. Cavitating lesions in the upper lobes, masses, miliary nodules, and mediastinal or hilar adenopathies are common.[9] Woodring and Fried[10] found that the majority of cavities larger than 15 mm in diameter were, as in our case, tumor disease. To our knowledge, 3 cases of *M. xenopi* infection associated with lung cancer have been published: adenocarcinoma,[11] large cell carcinoma,[5] and squamous cell carcinoma,[12] none of which presented the 2 entities simultaneously, as observed in our case.

To diagnose these diseases, mycobacteria must be grown in 3 sputum cultures, and clinical and radiological evidence must be consistent.[13] The best therapeutic combination and optimal treatment duration for *M. xenopi* lung infections remain to be determined. According to current criteria for NTM infection (ATS/IDSA 2007),[8] a 12-month course of a combination of rifampicin, ethambutol, and clarithromycin (or moxifloxacin, due to its low inhibitory concentration against mycobacteria[14]) is recommended.

Although *M. xenopi* infection is exceptional, we believe that this case illustrates the importance of ruling out NTM infection in the case of co-existing symptoms or nonspecific signs such as weight loss or anemia in patients with COPD and lung cancer.

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1579-2129/

Alpha-1-Antitrypsin Deficiency Associated With Null Alleles☆

Déficit de alfa-1-antitripsina asociado a alelos nulos

To the Editor,

Alpha-1 antitrypsin deficiency (AATD) is an autosomal codominant disorder that predisposes carriers to early development of chronic obstructive pulmonary disease (COPD) and/or liver disease. It is caused by the inheritance of 2 severe deficiency alleles in the SERPINA1 gene,[1] and a plasma concentration of alpha-1 antitrypsin (AAT) below 50 mg/dl is representative of a significant deficiency. Up to 95% of clinical cases related with AATD are associated with the PI*ZZ genotype, while the other 5% are associated with PI*SZ and PI*MZ genotypes or combinations of PI*S or PI*Z with other extremely rare deficiency or null alleles. These rare alleles account for 4.6% of the deleterious variants recorded in the Spanish Registry of Patients with AAT Deficiency, and null variants are very rare indeed.[2] Although around 25 null variants have been discovered in the last 20 years or so, little information is available on their clinical impact.[3–9] We report 2 cases of patients referred to the respiratory medicine clinic with a diagnosis of AATD associated with the PI*Q0amersfoort and PI*Q0cardiff allele.

The first was a 47-year-old woman, native of the former republic of Yugoslavia, with a smoking history of 15 pack-years, referred to the respiratory medicine clinic for a 1-year history of dyspnea on moderate exertion (mMRC 2). Lung function tests results were as follows: FEV1/FVC 0.5; FEV1 1.80 l (55%); FVC 3.40 l (77%); DLCO 53%; KCO 52%. High-resolution computed tomography revealed centrilobular emphysema, predominantly in both lower lobes. Complete blood count, IgA, IgM, IgG, IgE and transaminases were within normal levels. Plasma AAT determined by nephelometry was 18 mg/dl, so a genetic study to detect deficiency alleles S and Z was performed using real-time PCR (FRET, LightCycler 2.0, and TIB MOLBIOL probes), which detected heterozygosity for the PI*Z allele, while the presence of PI*S alleles was ruled out. In view of the discordance between the genotype obtained and AAT plasma levels, a molecular study was performed of all exonic coding regions and of the intronic sequences flanking the SERPINA1 gene, using Sanger sequencing (BigDye™ Terminator v3.1 Cycle Sequencing, Thermo Fisher Scientific). In this study, in addition to the PI*Z described above, heterozygosity for the PI*Q0amersfoort allele was detected (Fig. 1).

The second case was a 42-year-old man with no clinical history of interest, who was referred to our clinic for a family history of AATD. He had severe AAT deficiency (41 mg/dl), and heterozygosity for the PI*Z allele was detected. Molecular study of the SERPINA1 gene revealed a Z/Q0cardiff genotype (Fig. 1). The patient had no respiratory symptoms or liver disease. Lung function test results were within normal limits: FEV1/FVC 0.79; FEV1 4.82 l (109%); FVC 5.60 l (112%); DLCO 109%; KCO 106%. Chest radiograph and general laboratory tests were unremarkable.

AAT is an antiprotease, produced mainly by the hepatocytes, which inhibits the elastase activity of the neutrophils. Normal plasma levels range between 120 and 200 mg/dl.[1] Although the Z mutation (p.Glu 342 Lys) deficiency allele is the most common and leads to very low AAT levels in plasma (around 10%–15% of the normal level), serum AAT levels in null mutations are extremely low or undetectable[3–9]: a wide range of molecular mechanisms are involved in this outcome, including errors in protein synthesis or post-translational degradation.[6,10–12] For this reason, genotypes consisting of null homozygotes or accompanied by other deficiency alleles of the SERPINA1 gene carry a particularly high risk of very early onset pulmonary emphysema, even earlier than would be expected in the ZZ genotype.[13]

With regard to PI*Q0amersfoort, the limited literature available suggests that both heterozygous and homozygous forms lead to COPD at an early age, as observed in our female patient, with no liver involvement whatsoever.[7,8] This mutation causes a stop codon at position 184 of the protein, resulting in a severe deficiency when associated with other deficiency variants, such as PI*Z. Like other null alleles, the PI*Q0amersfoort variant does not cause liver disease because the protein is not polymerized in the liver, as occurs in mutations caused by amino acid switches, in which deficiencies exist that alter protein structure and folding, giving rise to accumulation in the endoplasmic reticulum of the hepatocytes, resulting ultimately in tissue damage.

In the case of PI*Q0cardiff, the amino acid aspartate is replaced by valine at position 256 of the ATT protein. This substitution causes a severe deficiency when it occurs in homozygosis or in association with other deficiency variants such as PI*Z. Some authors argue that homozygous Q0cardiff patients are not at risk for emphysema, although there may be a risk if it occurs in heterozygosity with the PI*Z allele or other null alleles.[9,14] In our case, the male patient was asymptomatic. In our opinion, PI*Q0cardiff cannot be

☆ Please cite this article as: Figueira Gonçalves JM, Martínez Bugallo F, García-Talavera I, Rodríguez González J. Déficit de alfa-1-antitripsina asociado a alelos nulos. Arch Bronconeumol. 2017;53:700–702.