



Early View

Research letter

Prolidase deficiency: a new genetic cause of combined pulmonary fibrosis and emphysema syndrome in the adult

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Prolidase deficiency: a new genetic cause of combined pulmonary fibrosis and emphysema syndrome in the adult

Letter to the editor

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Pulmonary fibrosis is considered to result from recurrent alveolar epithelial injury coupled with dysfunctional alveolar wound healing mechanisms, some of which have a genetic background. Pulmonary fibrosis in the adult has not been previously associated with prolidase deficiency, an innate deficiency of aminoacids metabolism.

A 22-year old Portuguese man was referred because of digital clubbing present since the age of 12 years. He had no familial history. He smoked 20 cigarettes/day since the age of 13. He was short of breath at exercise, with no cough. Velcro crackles were present at lung auscultation. The chest-CT demonstrated infiltrative lung disease, with mid- and upper lobe predominance, consisting of reticulation, and septal and peribronchovascular thickening (**Figure 1**). Mediastinal and axillar lymphadenopathies were present. Liver and spleen were increased in diameter. Bronchoalveolar lavage demonstrated 10% lymphocytes, 8% neutrophils, and 82% macrophages. No foamy macrophages were reported. Forced vital capacity (FVC) was 93% of predicted value, forced expiratory volume in 1s (FEV1) 94% of predicted, FEV1/FVC 0.83, total lung capacity (TLC) 95%, diffusion capacity for carbon monoxide (DLco) 83% and transfer coefficient 78%. Blood gas was normal. Routine blood tests were normal, except for increased lactic dehydrogenase and gammaglobulins (81 g/L). Serology for HIV was negative.

One year later, lung function had declined. A right-sided videothoroscopic lung biopsy was performed in three lobes, demonstrating diffuse alveolar fibrosis with collagen deposition, architectural distortion and alveolar cysts, which did not fulfil criteria for usual interstitial pneumonia. Dense lymphocytic inflammation was present with bronchiolar and peribronchovascular predominance, together with lymphoid follicles and germinative centers. Bronchiolar metaplasia was present. Fibroblastic foci were not found. Cholesterol clefts were abundant. In addition, airspaces were filled by patchy, granular, eosinophilic proteinaceous

exudate, stained with Periodic Acid Schiff, suggesting secondary pulmonary alveolar proteinosis accompanying pulmonary fibrosis. Immunohistochemistry demonstrated strong staining of alveolar exudate with anti-surfactant protein-C antibody. Birefringent particles were not found under polarized light. Staining of elastic fibers was normal. The pleura was normal except for mild lymphocytic inflammation; a mediastinal lymph node was normal. Anti-GM-CSF antibodies were negative. The patient was advised to quit smoking and corticosteroids were initiated, together with treatment for osteoporosis.

Examination further demonstrated small body size, normal body weight, kyphosis, dysmorphic face, grey hair, flat feet, some cognitive retardation, and numerous scars from dermal cysts. The patient was diagnosed with leg ulcer dermatitis, axonal neuropathy, and non-erosive, bilateral symmetric synovitis, osteoporosis, mixed glomerular and tubular proteinuria (0.5 g/L), and mild-severity autoimmune hepatitis. Rheumatoid factor, homogeneous antinuclear antibodies, and mixed type-II cryoglobulinemia were present. Overlapping rheumatoid arthritis and systemic lupus were diagnosed.

Sequencing of *TERT*, *TERC*, *SFTPC*, *SFTPB*, and *ABCA3* genes did not show pathogenic variants. Analysis of blood and urine aminoacids demonstrated massive iminodipeptiduria. The level of glycine-proline dipeptide was 30 $\mu\text{mol/L}$ in the plasma (normal: absent) and 8480 $\mu\text{mol/L}$ in urine (normal <20). The biochemical diagnosis of prolidase deficiency was established. *PEPD* gene sequencing identified the homozygous variant NM_000285.4:c.692_694del (p.Tyr231del) in exon 10, previously reported in two unrelated Portuguese patients [1], but never observed in GnomAD populations. This in-frame 3bp-deletion impacts the secondary structure of the protein and strongly reduces the prolidase enzymatic activity [2]. Oral supplementation with vitamin C, proline, and essential amino acids was initiated.

On follow-up, he continued smoking 40 cigarettes/day, and 2 joints/day of cannabis for 11 years. The patient was admitted on multiple times for lower airway infections. Oral corticosteroid treatment was maintained despite limited patient compliance, with some transient benefit observed on clinical and functional outcome. Six years after presentation, he developed atrophic polychondritis with typical relapsing inflammation and deformity of ear cartilage. Over the years, lung function deteriorated, with severe airflow obstruction and air trapping. Ten years after the lung biopsy, FVC was 63% of predicted, FEV1 38%, FEV1/FVC 0.39, residual volume 5.6 L (233%), RV/TLC 0.67 (248%), TLC 139%, DLco 48%, and Kco 48%. Hypercarbia was then present, and supplemental oxygen therapy and non-invasive ventilation were initiated, with excellent tolerance. At last follow-up 16 years after lung biopsy, FVC was 49%, FEV1 19%, FEV1/FVC 0.32, TLC 121%, RV 295%, RV/TLC 0.72, DLco 14%, Kco 23%. Chest CT demonstrated progression of interstitial changes over the first 10 years of follow-up, with later apparent decreased severity of interstitial abnormalities at imaging while emphysematous changes markedly progressed.

Prolidase deficiency (OMIM #170100) is a rare autosomal recessive multisystem disorder associated with imidodipeptiduria and reduced prolidase activity, caused by homozygous or compound heterozygous mutation in the *PEPD* gene on chromosome 19q13. The disorder is clinically heterogeneous and may comprise slow healing ulcerations of the legs and feet, various types of eruptions, telangiectases, mental retardation, recurrent respiratory tract infections, and facial dysmorphism [3]. Diagnosis is based on biochemical techniques [4].

Previous reports in prolidase deficiency either were related to children [5] or did not provide detail about the phenotype of lung disease, which may include recurrent pulmonary infections, cystic changes, bronchiectases, diffuse ground glass attenuation, minor linear

atelectasis [6] and autoimmune pulmonary capillaritis [5]. Our adult patient had a non-usual interstitial pneumonia pattern of pulmonary fibrosis at biopsy. Cholesterol clefts were prominent similar to pediatric cases [5]. In addition to collagen deposition and alveolar cyst formation, significant lymphocytic bronchiolitis was observed, which may have contributed to air trapping, development of low attenuation areas, and severe airflow obstruction, which became prominent over time despite long-term oral corticosteroids. Indeed, long-term follow-up suggests that fibrosis may surprisingly have become less pronounced with time, while emphysema and cystic lesions progressed and likely were responsible for chronic respiratory insufficiency, with prolonged efficacy of non-invasive ventilation. Although the imaging features were that of combined pulmonary fibrosis and emphysema, pulmonary function demonstrated progressive airflow obstruction with increased residual volume not characteristic of this syndrome. Interestingly, other genetic causes of combined pulmonary fibrosis and emphysema have been described [7]. Corticosteroids initiated because of presence of inflammatory infiltrates on the biopsy provided some medium-term improvement but were discontinued after 8 years because of severe osteoporosis, and may have contributed to recurrent respiratory infections.

Pathogenesis of fibrosis and emphysema in prolidase deficiency is unclear. Tobacco smoking may have played a role. Cell-death of fibroblasts [8] with interference with the normal recycling of collagen [9], and increased oxidative stress [10] may contribute to both fibrosis and emphysema. This observation suggests that prolidase deficiency is a genetic factor that may contribute to pulmonary fibrosis and potentially emphysema.

Consent. The patient has provided written informed consent for the publication of the present manuscript as per guidelines of the journal.

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Figure legends.

Figure 1.

A-C: Lung window of axial chest CT demonstrating infiltrative lung disease, with mid- and upper lobe predominance, with reticulation, and septal and peribronchovascular thickening, at baseline (A), year +9 (B), and year +17 (C). C: At last follow-up, fibrotic features are less pronounced, whereas cystic changes, emphysematous lesions and air trapping have worsened.

D-F: Microscopic examination of right-sided lung biopsy. D: Homogeneous distribution of alveolar wall thickening and of eosinophilic proteinaceous exudate (H.E.S. x1.5). E: Intense cytoplasmic staining of epithelial alveolar cells and of alveolar exudate with anti-surfactant protein C antibody. (IHC x40). F: Alveolar wall thickening and eosinophilic proteinaceous exudate; two lymphoid follicles are also visible (H.E.S. x10).

G: Deformation of ear lobes on both sides resulting from deformity and chronic inflammation due to relapsing polychondritis.

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