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22q11.2 deletion syndrome complicated with pulmonary alveolar proteinosis in a child: a case report

Y. ZHENG[1], Y. LI[1], G.-H. MAO[1], H.-C. DAI[1], G.-T. LI[1], C.-L. YANG[2], Y. ZHU[1]

[1]Department of Pediatrics, Fuyang People's Hospital, Fuyang, China
[2]Graduate School of Wannan Medical College, Wuhu, China

Yu Zheng and Yan Li are co-first authors and they contributed equally to the work

Abstract. – BACKGROUND: To analyze the clinical data and next generation sequencing (NGS) results from a child with 22q11.2 deletion syndrome (22q11DS) complicated with pulmonary alveolar proteinosis (PAP) who was admitted to the Department of Pediatrics of Fuyang People's Hospital and to present a review of the literature.

CASE PRESENTATION: A 9-year-old male child, whose face had a small mandible and high-arched palate, but lacked a cleft palate, had repeated respiratory tract infections and bronchiectasis. Clinical examination, computer tomography, and electronic bronchoscopy were performed. Genetic testing via NGS was undertaken.

PAP was confirmed by Periodic Acid Schiff staining of milky white alveolar lavage fluid isolated by electronic bronchoscopy. A deletion of approximately 2.46 Mbp on chromosome 22q11.2 was confirmed by NGS. During hospitalization, anti-infection, nebulization, alveolar lavage, and regular application of thymosin were administered to the patient. The condition of the patient stabilized following treatment.

CONCLUSIONS: 22q11DS and PAP are both rare diseases, and the manifestation of 22q11DS combined with PAP has not been previously reported. The diagnosis and treatment of this case will be a reference for future clinical work.

Key Words:
Children, 22q11.2 deletion syndrome, DiGeorge syndrome, Pulmonary alveolar proteinosis, Case report.

Introduction

22q11.2 deletion syndrome (22q11DS) is a comprehensive disorder of congenital organ dysplasia associated with DiGeorge syndrome, velo-cardio-facial syndrome, Takao syndrome, and conotruncal anomaly face syndrome clinical phenotype[1], which can include a distinctive face together with congenital heart disease, hypocalcemia, immunodeficiency, cleft palate, interstitial pneumonia, and mental disorders. There are many reports[2-6] of this condition in China and globally. However, the manifestation of 22q11DS combined with pulmonary alveolar proteinosis (PAP) is relatively rare and has not been reported. Here, we report the presentation of PAP in a male child with 22q11DS.

Case Presentation

Medical History

A 9-year-old male patient with repeated cough for 2 weeks, aggravation and fever for 1 day was admitted to the Department of Pediatrics of Fuyang People's Hospital on December 28, 2021. At disease onset, the patient was given Amoxicillin Clavulanate Potassium Granules as an oral treatment. However, this treatment was ineffective, and the patient was readmitted in a worse condition. The patient had been diagnosed with congenital laryngomalacia in infancy, had a history of surgical treatment of right thumb syndactyly, and had experienced pneumonia on an average of 3 to 4 times per year. The patient had no history of PAP.

Physical Examination

The patient had a clear mind, average spirit, poor nutritional development, and distinctive facial features with small mandible and high-arched palate (Figure 1). The patient had normal oral mucosa, pharyngeal congestion, and second-degree enlarged tonsils, but did not have a cleft palate.

Corresponding Author: Ying Zhu, MD; e-mail: ahfyww@126.com

Figure 1. Clinical phenotype of patient.

The three concave sign was negative. Respiratory rate was 22 breaths per minute. Breath sounds in both lungs were rough, and moist rales and wheezing were heard. Heart sounds were strong and regular with a heart rate of 110 bpm; pathological murmurs were not detected in the valve area. The abdomen was soft, no mass was observed, liver and spleen were unpalpable, and bowel sounds were also normal. Nervous system examination revealed no abnormality. In addition, old scars were present on the right thumb and forefinger without finger clubbing.

Auxiliary Inspection

After hospitalization, we performed a clinical examination of the patient (Table I).

Chest computer tomography (CT) showed bronchiectasis with infection in the middle and lower lobe of the right lung and right aortic arch (Figure 2).

Electronic Bronchoscopy

After communicating with the parents of the patient and signing the informed consent, electronic bronchoscopy and bronchoalveolar lavage were performed twice, on the 4th and 12th day of admission. The electronic bronchoscopy findings are described as follows. The opening of each branch of the right lung was normal, the mucosa was rough and swollen, and white secretions were attached, which were obvious in the upper right, middle, and lower right basal segments, and the lavage fluid was milky white. Mucus plug formation was seen in the right middle and lower right anterior basal segments, and the ventilation of each branch improved after lavage and suction. The opening of each branch of the other lung was normal. The mucosa was rough and swollen, and there was white secretion attached, which was obvious in the basal segments of the upper left, left tongue, and lower left. Bronchoscopic diagnosis was endobronchial inflammation and mucus plug formation.

Inspection of Bronchoalveolar Lavage Fluid

The appearance of bronchoalveolar lavage fluid was milky white and turbid (Figure 3A). Lavage fluid (10 mL) was collected and sent for inspection. Qualitative detection of nucleic acid of *Haemophilus influenzae* was positive in the lavage fluid, and the periodic acid-Schiff (PAS) was positive, indicating that the precipitated substance was lipoprotein (Figure 3B).

Genetic Testing

Genetic testing was performed after approval was obtained by the hospital Ethics Committee (Ethics Number: 2021-131), and informed consent was given by the parents of the patient. Peripheral blood (5 mL) was collected from the patient and their parents and sent to Fuzhou Furui Medi-

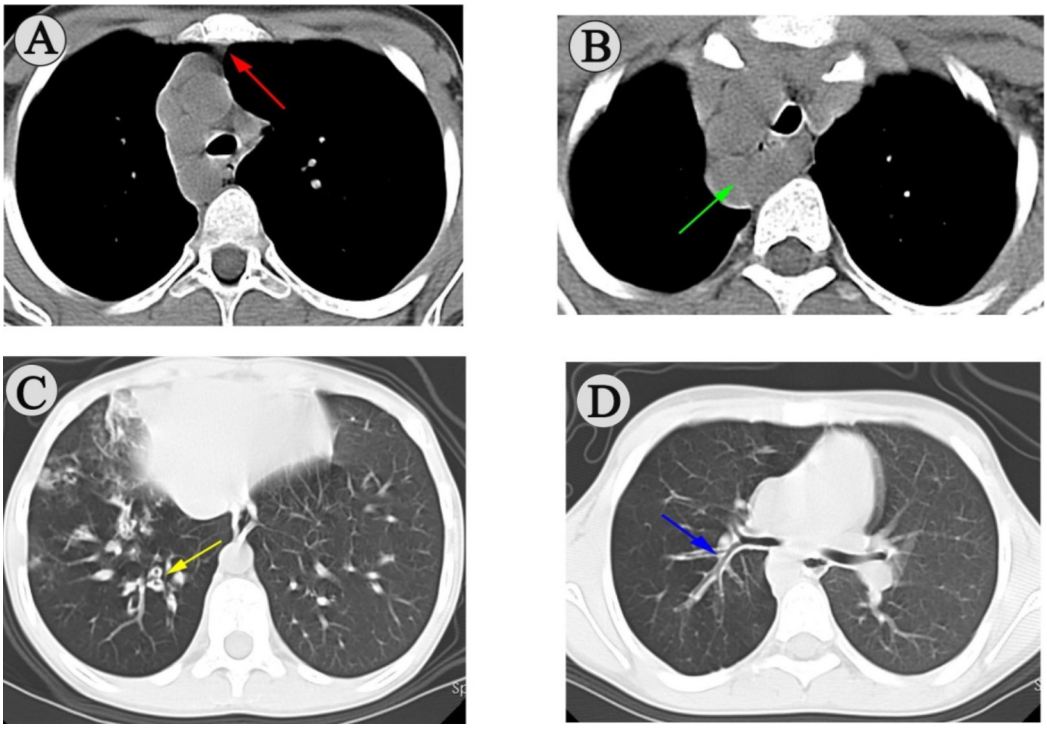

Figure 2. Computed X-ray tomography (CT) imaging scan results. **A**, The patent is thymic hypoplasia (*red arrow*). **B**, Right aortic arch (*green arrow*). **C-D**, Chest CT showed bronchiectasis in the patient: signet ring sign (*yellow arrow*) and orbital sign (*blue arrow*).

Table I. Partial results of workup of the child.

Examination Items	Workup	Reference Ranges
Blood routine		
WBC ($\times 10^9$/L)	10.47	3.50-9.50
NE%	83.4	40.0-75.0
LYMPH%	10.0	20.0-50.0
RBC ($\times 10^{12}$/L)	3.99	4.30-5.80
HGB (g/L)	120	130-175
PLT ($\times 10^9$/L)	163	125-350
Biochemical parameters		
Ca (μmol/L)	1.90	2.11-2.52
P (μmol/L)	1.76	0.85-1.51
CRP (mg/L)	42.71	0.00-2.80
IgA (g/L)	4.45	0.70-4.00
IgM (g/L)	1.42	0.40-2.30
IgG (g/L)	17.55	7.00-16.00
T lymphocyte subsets		
CD+ 3(/μL)	1,412	690-2,540
CD+ 3CD+ 4 (/μL)	592	410-1,590
CD+ 3CD+ 4 (%)	21	31-60
CD+ 3CD+ 8 (/μL)	708	190-1,140
CD+ 3CD+ 8 (%)	26	13-41
CD4/CD8	0.81	0.90-2.50
PTH (pg/mL)	12.20	18.50-88.0
G test (pg/mL)	1.681	0.00-10.00
PPD test	0.4×0.6cm	NA
Echocardiography	Normal	NA

WBC: white blood cells; NE: neutrophil; LYMPH: lymphocyte, RBC: red blood cells; HGB: hemoglobin; PLT: platelet; Ca: calcium; P: phosphorus; CRP: C-reactive protein; IgA: immunoglobulin A; IgM: immunoglobulin M; IgG: immunoglobulin G; PTH: parathyroid hormone.

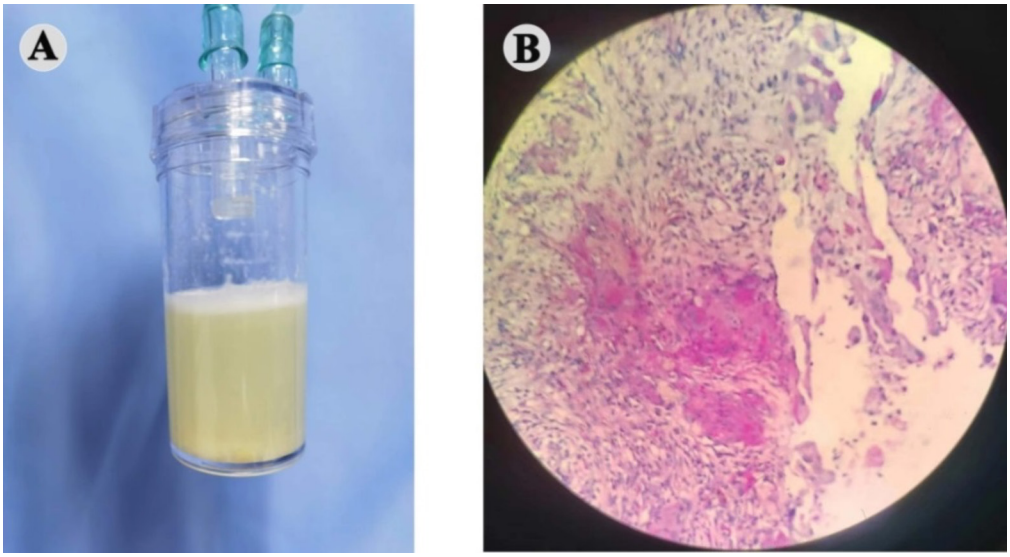

Figure 3. Alveolar lavage fluid results. **A**, The bronchoalveolar lavage fluid appeared milky white and turbid. **B**, Periodic acid-Schiff-stained alveolar lavage fluid sediment (20×).

cal Laboratory. After DNA adapter addition, amplification, and purification, a DNA library was prepared using the hybridization capture method. A high-throughput sequencing platform was then used to detect the exon regions and flanking intron regions (20 bp) of 20,099 genes in the human whole exome. The sequencing data were compared with the reference sequence of human genome hg19 (GRCh37), and the coverage and sequencing quality of the target region were evaluated. A deletion of approximately 2.46 Mbp was detected on chromosome 22q11.21 (chr22:18893715-21535365) in the genomic DNA (Figure 4).

Diagnosis, Treatment, and Follow-Up

According to electronic bronchoscopy and PAS staining of bronchoalveolar lavage fluid, the patient was diagnosed with PAP. The patient had experienced repeated respiratory infections since childhood and had distinctive facial features, congenital laryngomalacia, and syndactylism, and auxiliary examinations showed low calcium and high phosphorus levels, low parathyroid hormone levels, low cellular immune functions, and right aortic arch; the clinical suspicion was 22q11DS, which was confirmed by next generation sequencing (NGS). Consequently, the patient was diagnosed with 22q11.2 deletion syndrome, pulmonary alveolar proteinosis, bronchiectasis with infection. After being admitted to the hospital, the symptoms of the patient improved daily with successive use of cefuroxime, ceftriaxone anti-infective treatment, atomization, and two bronchoalveolar lavage treatments. Afterwards, in addition to anti-infective treatment, intravenous infusion of thymosin was given to enhance immunotherapy (once a week on a regular basis). Ultimately, the condition of the patient was stable, and there was no recurrence of respiratory infection during follow-up.

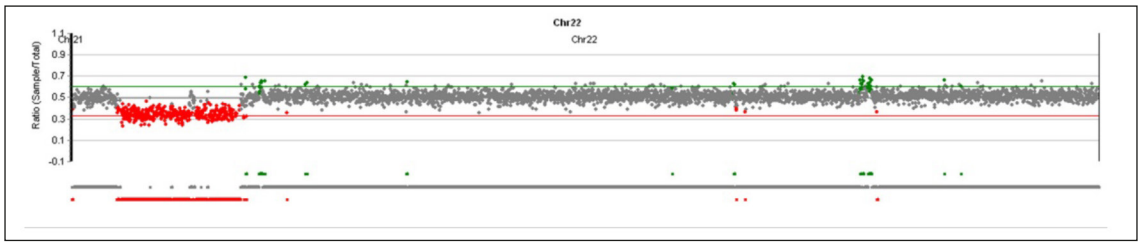

Figure 4. Patient chr22:18893715-21535365 (2.46 Mbp, 22q11 deletion).

Discussion

22q11DS, also known as DiGeorge syndrome, is a polygenic genetic disorder that involves four locus control regions (LCR) *LCR22A*, *LCR22B*, *LCR22C*, and *LCR22D*, and multiple genes, including *PRODH*, *TBX1*, *CRKL*, *SLC25A1*, *GP1BB*, *SCARF2*, *SNAP29*, and *LZTR1*[7]. Studies[8,9] have demonstrated that the *TBX1* is a key gene involved in pathogenesis of 22q11DS and plays a critical role in gene regulation in embryogenesis of the heart, parathyroid gland, thymus, palate, and teeth. The clinical features of the right aortic arch, thymus dysplasia, hypoparathyroidism, and hypocalcemia are consistent with those described in the literature[10]. The *CRKL*, *SCARF2*, and *PI-4KA* genes are modifier genes of this syndrome, but do not directly affect the core clinical phenotype of 22q11DS, although deletion of these genes to varying degrees can result in more severe or complex clinical phenotypes. 22q11DS can occur at all ages, and the clinical phenotypes are various. Accordingly, the morbidity rate varies from high to low, and affected patients have craniofacial deformity, developmental delay, immunodeficiency, congenital heart disease, dysplasia of the palate arch, and hypothyroidism[11]. In this case, the patient had experienced repeated respiratory tract infection since childhood and the combined occurrence of this with PAP indicates immunodeficiency. There are no reports of 22q11DS combined with PAP in the literature.

PAP is a rare clinical disease. The pathogenesis involves impaired clearance or abnormal production of alveolar surfactants that leads to the filling of alveoli with lipid-rich PAS-positive protein substances. The accumulation of surfactant in the alveoli and terminal airways finally affects the pulmonary ventilation function. The clinical manifestations of PAP in children are often nonspecific. The most common symptoms are chronic cough, dyspnea, and chronic bronchitis, and approximately 30% of patients can be asymptomatic. Lung CT in typical PAP children can show characteristic paving stone-like changes. However, in this case, the grid-like interlobular septa in the lung lobe may not have thickened because of the short onset time, and no characteristic imaging changes were shown. There are three clinical forms of PAP: congenital, secondary, and idiopathic; among these, congenital PAP mainly includes autoimmunity and hereditary causes. Furthermore, autoimmunity accounts for 90% of PAP cases in adults. Secondary and congenital cases are more common in children, whereas the idiopathic condition is rare[12]. Congenital PAP is mostly an autosomal recessive hereditary disease that largely occurs in infancy and is mainly caused by the mutation of alveolar surfactant protein-B (*SP-B*) gene. Idiopathic PAP is mainly associated with the function of granulocyte-macrophage colony-stimulating factor (CM-CSF), whereas secondary PAP is predominantly caused by lysinuric protein intolerance, immunodeficiency diseases, and hematological tumors[13]. The patient in this case had repeated respiratory tract infections since childhood, a distinctive face, syndactylism, and congenital laryngomalacia. Auxiliary examinations showed an immunocompromised state, low calcium and high phosphorus levels, hypoparathyroidism, and a right aortic arch. NGS identified the 22q11.2 deletion and showed that the underlying cause was 22q11DS, which further led to secondary PAP.

The cause of 22q11DS complicated with PAP may be relevant to the reduced function of alveolar macrophages[14,15]. The main function of alveolar surfactant is to reduce the surface tension in the alveoli and prevent alveolar collapse. The lipids and proteins in the alveolar surfactant are removed by the uptake and recycling of type II epithelial cells or the uptake and catabolism of alveolar macrophages. The clathrin heavy chain-like 1 (*CLTCL1*) gene is one of the genes deleted in 22q11DS and is a dimer composed of a heavy and a light chain[16]. Current studies[17-19] suggest that clathrin mediates endocytosis, and deletion then impairs clathrin synthesis, which impedes alveolar macrophage scavenging of alveolar surfactant. The obstruction of the catabolic process of alveolar surfactant leads to the deposition of alveolar surfactant.

We performed literature database searches using Chinese “22q11.2 deletion syndrome”, “Di George syndrome”, and “pulmonary alveolar proteinosis” as keywords in China National Knowledge Infrastructure (CNKI), Wan fang Medical Online, and China Science and Technology Journal Database. We also searched PubMed, National Center for Biotechnology Information (NCBI), Springer Link, and Science Direct using English “22q11.2 deletion syndrome”, “Di George syndrome”, and “pulmonary alveolar proteinosis” as keywords. We failed to retrieve any Chinese publications and only one relevant English publication, which described a case of chromosome 10 p microdeletion syndrome with clinical manifestations highly similar to 22q11DS that suggest a

misdiagnoses[20]. Additional cases have not been reported in China or globally so far.

At present, the preferred method for treating PAP remains whole lung lavage[21]. Although lung lavage helps remove abnormal surfactants in the lungs, this cannot solve the etiological problem. Attention should be paid to the treatment of the primary disease. This case achieved curative effect with thymosin treatment after diagnosis, and there was no recurrence of respiratory tract infection during follow-up. Through the diagnosis and treatment of this case, we concluded that: the diagnosis of PAP is often the beginning rather than the end point in the diagnostic process. Children with PAP predominantly have the secondary clinical form and identifying the primary disease and treating the cause is the key to treatment. It is very rare for children with 22q11DS to be complicated with PAP. The presence of immunodeficiency indicates that enhanced immunotherapy is feasible and should be the focus of treatment. Attention should be also paid to treating secondary pulmonary infections caused by common pathogens such as *Haemophilus influenzae* and *Streptococcus* sp. as well as other pathogens such as *Mycobacterium*, *Nocardia*, *Actinomyces*, *Aspergillus*, and *Cryptococcus*[22]. The timely qualitative examination of pathogen nucleic acid in lavage fluid, G test, PPD test, and other tests are helpful for drug selection.

Conclusions

It is necessary to strengthen the understanding of 22q11DS and PAP, both of which are rare diseases with no specific manifestations and as such are easy to be misdiagnosed as pulmonary infection. In clinical work, the cause should be actively searched in children with repeated pneumonia or rare pulmonary clinical manifestations. If necessary, genetic testing should be carried out to exclude immune deficiency-related diseases.

Conflict of Interest

The Authors declare that they have no conflict of interests.

Informed Consent

The informed consent was obtained by the parents of the patient.

Ethics Approval

Ethics approval was obtained by the Ethics Committee of the Fuyang People's Hospital (Ethics Number: 2021-131).

Authors' Contributions

Yu Zheng contributed to the conception and design, data analysis and interpretation, and manuscript writing; Yan Li, Guoshun Mao and Hongchen Dai contributed to the provision of study materials or patients; Guitao Li and Chaolei Yang contributed to the collection and assembly of data; Ying Zhu contributed to the administrative support and final revision; and all authors have read and approved the final manuscript.

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