Cobalamin C deficiency presenting with diffuse alveolar hemorrhage and pulmonary microangiopathy: a case series of four patients

Jinrong Liu

Beijing Children's Hospital

Xiaolei Tang

Beijing Children's Hospital

Chunju Zhou

Beijing Children's Hospital

Hui Xu

Beijing Children's Hospital

Haiming Yang

Beijing Children's Hospital

Huimin Li

Beijing Children's Hospital

Shunying Zhao (✉ zhaoshunying2001@126.com)

Beijing Children's Hospital, National Center for Children's Health, Capital Medical University

Case report

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Abstract

Background: The clinical manifestations of combined methylmalonic acidemia (MMA) and homocysteinemia (cobalamin deficiency) vary, but typically include neurologic, developmental and hematologic abnormalities. Only a few patients had diffuse lung disease (DLD). We analyse the clinical features of cobalamin C deficiency (CblC deficiency) who developed late-onset DLD mainly diffuse alveolar hemorrhage (DAH) to strengthen an understanding of it.

Results: This study describes 4 patients aged 4-years-2-months to 7-years-6-months with CblC deficiency who developed late-onset DLD. Of these, the first 3 patients presented predominantly with DAH, and the last patient with pulmonary microangiopathy confirmed by lung biopsy. All patients accompanied by pulmonary arterial hypertension (PAH), and 2 accompanied by moderate megaloblastic anemia. Diffuse ground-glass opacification and poorly defined ground-glass centrilobular nodules were seen on high-resolution computed tomography (HRCT) in 1 patient and 3 patients respectively. All patients were suspected of having idiopathic pulmonary hemosiderosis (IPH) or interstitial lung disease (ILD) at other hospitals. The last 2 patients received treatment with high dose corticosteroid before admission, but the symptoms didn't improve. Moreover, all patients carried compound heterozygous mutations (c.80A>G, c.609G>A) and improved significantly after being treated for CblC deficiency and PAH.

Conclusions: CblC deficiency should be considered in the differential diagnosis of DAH especially with PAH, and pulmonary microangiopathy be the main reason of DLD in these patients.

Background

Hyperhomocysteinemia may damage blood vessels, causing pulmonary arterial hypertension (PAH) and thrombosis [1,2]. Both microangiopathy and thromboembolism can be the underlying mechanisms for PAH in cobalamin deficiency (combined MMA and homocysteinemia) [3–7]. Only a few patients had diffuse lung disease (DLD) in cobalamin C deficiency (CblC deficiency) [8].

Here, we report a series of 4 pediatric patients with CblC deficiency who developed late-onset DLD. Of these, all 4 patients presented with concomitant PAH, 3 presented with diffuse alveolar hemorrhage (DAH), 2 presented with moderate megaloblastic anemia, and 1 presented with pulmonary microangiopathy confirmed by lung biopsy. Interestingly, all patients were compound heterozygous for the c.80A>G variant in combination with the common Chinese variant c.609G>A.

This confirms pulmonary microangiopathy secondary to combined MMA and homocysteinemia should be the primary cause of DLD. The new symptom of DAH extends the pulmonary manifestation spectrum but builds on the well-described pulmonary phenotype in CblC disease. To our knowledge, these are the first such patients reported to date.

Case Presentation

Clinical data, laboratory and auxiliary examinations

From 2017 to 2018, a total of 4 pediatric patients with CblC deficiency were identified at the Beijing Children's Hospital affiliated to Capital Medical University, National Center for Children's Health for DLD. DLD was diagnosed by lung imaging. DAH was confirmed by (1) diffuse alveolar infiltrates on radiological imaging, plus (2) at least one of the following clinical findings: bronchoscopic evidence of bleeding and pathologically by the accumulation of hemosiderin-laden macrophages in bronchoalveolar lavage fluid (BALF). Pulmonary microangiopathy was determined by pulmonary biopsy findings. PAH was confirmed by echocardiography and pulmonary artery (PA) with an enlarged diameter exceeding the aorta in high-resolution computed tomography (HRCT). Laboratory investigations included serum levels of homocysteine and methylmalonic acid in addition to routine biochemical analysis. Bone marrow puncture and genetic analysis was performed in all patients.

The study was approved by the ethical committee Beijing Children's Hospital, National Center for Children's Health. The parents of all of the 4 patients gave informed written consent for publication of the data.

Results

In all patients, metabolic investigations were suggestive of cobalamin deficiency, with elevated blood propionylcarnitine (C3) concentration as well as an elevated propionylcarnitine–acetylcarnitine (C3/C2) ratio. The serum homocysteine and methylmalonic

acid concentrations were elevated. Genetic analysis confirmed a compound heterozygosity in MMACHC, with c.80A>G (p.Q27R) and c.609G>A (p.W203X) sequence variants in all patients, inherited from their mothers and fathers. All patients were finally diagnosed with CblC deficiency.

Patients 1 and 3 had moderate megaloblastic anemia. A large number of hemosiderin-laden alveolar macrophages were observed in BALF in the first 3 patients which suggested DAH. Echocardiography showed PAH in all patients. Diffuse ground-glass opacification and poorly defined ground-glass centrilobular nodules were seen on HRCT in 1 patient and 3 patients respectively (Fig. 1). Thoracoscopic lung biopsy was performed in patient 4, and biopsy findings confirmed microangiopathy (Fig. 2) and negative homocysteine. Autoantibodies associated with connective tissue diseases and anti-glomerular basement membrane antibody were negative in all patients.

Clinical characteristics and biochemical and genetic analyses are summarized in Table 1.

Clinical Investigation

Patient 1 was admitted at 7-years–6-months, because of a 1-year history of pale face and a 2-months history of wet cough and shortness of breath. Laboratory investigations showed a hemoglobin of 6.8 g/dl.

Patient 2 was admitted at 4-years–6-years, because of vomiting for 15 days, and fatigue and shortness of breath for 12 days.

Patient 3 was admitted at 4-years–2-months, because of fatigue and vomiting for 2 months, and shortness of breath for 3 weeks. Laboratory investigations showed a hemoglobin of 7.6 g/dl.

Patient 4 was admitted at 6-years–2-months, because of cough for 2 years, and fatigue and shortness of breath for 1 year.

All patients had no clinical manifestations of renal impairment, but estimated glomerular filtration rate (eGFR) decreased in patients 2 and 4. Patients 1 and 2 had transient proteinuria. Mean corpuscular volume (MCV) increased in all patients. All patients were suspected of having idiopathic pulmonary hemosiderosis (IPH) or interstitial lung disease (ILD), and the last two patients had respiratory failure (Type I) and were treated with pulsed methylprednisolone at other hospitals, but they didn't improve. They significantly improved after being treated for CblC deficiency including hydroxocobalamin, folate, betaine, levocarnitine, and vitamin B6. In addition, all patients were simultaneously treated for PAH such as sildenafil and bosentan.

All patients were asymptomatic at 1 year's follow-up.

Discussion

To our knowledge, this case series is the first description of children with the rare combination of late-onset DAH with CblC deficiency established as the denominator.

In most cases, the clinical symptoms of CblC deficiency tend to appear during infancy or early childhood as a multisystem disease with severe neurologic, hematologic, renal, and gastrointestinal signs. In this case series of children with CblC deficiency presenting predominantly with late-onset DAH and pulmonary microangiopathy. Of these, all 4 patients also had PAH to some extent, 2 had moderate megaloblastic anemia, and 2 had insidious renal injury. In our present reported cases, all patients were late-onset (older than 4 years) and had no obvious sign of neurologic abnormalities, which made diagnosis difficult. However PAH and megaloblastic anemia were the clinical hallmark sign in our patients.

All 4 patients carried compound heterozygous mutations (c.80A>G, c.609G>A) which have been reported in 2 late-onset Chinese patient with PAH (including 1 case of DLD) [8,9] and 8 early-onset Chinese patients [10,11]. The allele frequency of the c.80A>G variant is 9.09% in the Chinese population with CblC deficiency [12]. Our previous published study, 3 CblC deficiency patients with DLD also had c.80A>G mutation [8], which suggested DLD might be highly associated with c.80A>G mutation. The c.609G>A mutation is a hot spot mutation (43.64%) in Chinese patients with CblC deficiency [10–12], which often lead to early-onset cblC disease[12].

Immunohistochemical staining was performed to detect homocysteine of lung tissue and revealed negative in patient 4, which didn't suggest the deposition of homocysteine in the lungs. However high level of plasma homocysteine may damage the blood vessels and

then increase the permeability of pulmonary capillaries. There have been some case reports of pulmonary embolism and PAH in patients with CblC deficiency [3–7,9,13,14]. Severe pulmonary vascular lesions with thromboemboli and massive endothelial proliferation even pulmonary venous occlusive disease (PVOD) were found by lung postmortem in patients with cobalamin deficiency [6, 13–15]. Pathological findings of lung biopsy confirmed pulmonary microangiopathy contributing to DLD in our patient 4. Therefore presence of diffuse ground-glass opacification and poorly defined ground-glass centrilobular nodules on HRCT suggested pulmonary microangiopathy, which might cause DAH. In addition, pulmonary microangiopathy has never been to the best of our knowledge described in living patients with CblC deficiency.

DAH can have a myriad of underlying etiologies including systemic vasculitides, rheumatic disease, anti-glomerular basement membrane disease, IPH, and systemic lupus erythematosus (SLE) [16]; however, DAH secondary to CblC deficiency has not been reported. DAH is considered a life-threatening medical emergency with nonspecific signs, symptoms, and chest imaging that can lead to respiratory failure and death. Consequently, early diagnosis and appropriate management are extremely important. Hemoptysis, anemia, and new lung infiltrates are the most common presentation; however, in some cases, hemoptysis is absent. PAH can be secondary to chronic pulmonary diseases especially ILD. In our present study, all patients had diffuse lung infiltrates and PAH, additionally patients 1 and 3 accompanied by moderate anaemia, therefore all of them were suspected of IPH or ILD at other hospitals. Moreover, patients 3 and 4 had respiratory failure, and they received treatment with pulse methylprednisolone, but no response. All patients improved significantly after being treated for CblC deficiency and PAH. Therefore treatment should be initiated in a timely fashion, as lung disease secondary to this metabolic disorder will respond to appropriate treatment but may not respond to symptomatic treatment.

Conclusions

We have described 4 pediatric patients with DAH and pulmonary microangiopathy caused by CblC deficiency. To our knowledge, this is the first report of CblC deficiency-associated DAH in pediatric patients, suggesting that CblC deficiency should be considered a potential cause of DAH, especially with PAH and megaloblastic anemia. Therefore, pulmonologists should highly pay attention to mean corpuscular volume (MCV) in DAH patients. Diffuse ground-glass opacification and typical poorly defined ground-glass centrilobular nodules on lung HRCT is another important diagnostic clue.

Declarations

Ethics Approval and Consent to Participate

The research was approved by the Ethics Committee of Beijing Children's Hospital. All patient guardians gave informed consent to the study.

Consent for publication

All patient guardians gave informed consent to the publication of this study.

Availability of data and materials

Please contact author for data requests.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

JL supervised the patient care, conceptualized and designed the report, collected the data, and drafted the initial manuscript; XT supervised the patient care and collected the data; Dr Zhou performed histopathology and analyze the data; HX supervised the patient care; HY supervised the patient care; HL supervised the patient care and collected the data; SZ supervised the patient care, conceptualized the report, and reviewed and revised the manuscript. All authors read and approved the final manuscript.

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Author details

¹Department of Respiratory Medicine, Beijing Children's Hospital affiliated to Capital Medical University, National Center for Children's Health, Nanlishi Road 56, Xicheng District, Beijing, P. R. China. ² Department of Pathology, Beijing Children's Hospital affiliated to Capital Medical University, National Center for Children's Health, Nanlishi Road 56, Xicheng District, Beijing, P. R. China.

Abbreviations

BALF: bronchoalveolar lavage fluid; CblC: cobalamin C; DAH: diffuse alveolar hemorrhage; DLD: diffuse lung diseases; HRCT-high-resolution computed tomography; Hcy: homocysteine; ILD: interstitial lung disease; IPH: idiopathic pulmonary hemosiderosis; MMA: Methylmalonic Acidemia; PAH: pulmonary arterial hypertension.

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Tables

Table 1 Clinical features and results of auxiliary examinations of 4 Chinese patients with CblC deficiency

	Patient 1	Patient 2	Patient 3	Patient 4
Gender	Female	Male	Male	Female
Age	7-years-6-months	4-years-6-months	4-years-2-months	6-years-2-months
Presentation	Cough, short of breath	Vomiting, fatigue, short of breath	Vomiting, fatigue, dyspnea	Cough, fatigue, dyspnea
Respiratory failure	-	-	Type I	Type I
Weight(kg)/ PCSAG	18.0/<5th percentile	11.5/3rd percentile	14/<10th percentile	18.8/<25th percentile
MCV (fL)	108.3	105.5	117.5	105.1
Hemoglobin(g/dL)	6.8	12.0	7.6	12.5
Urine protein	2+	-	2+	-
eGFR (80-120ml/min/1.73m ²)	113.9	37.8	93.1	62.9
Plasma Hcy (μmol/L)	108	131	103	136
Metabolic acidosis	-	Mild	-	Moderate
PAH	Mild	Severe	Moderate	Severe
Bone marrow examination	Megaloblastic anemia	Enlargement of some mature red blood cells	Megaloblastic anemia	Enlargement of some mature red blood cells
Treatment for PAH	Captopril	Sildenafil and bosentan	Sildenafil	Sildenafil and bosentan
MMACHC gene	c.80A>G (p.Q27R), c.609G>A (p.W203X)	c.80A>G (p.Q27R), c.609G>A (p.W203X)	c.80A>G (p.Q27R), c.609G>A (p.W203X)	c.80A>G (p.Q27R), c.609G>A (p.W203X)

Abbreviations: eGFR-estimated glomerular filtration rate; Hcy-homocysteine; MCV-Mean corpuscular volume; PCSAG-percentile of the children with same age and gender. PAH-pulmonary arterial hypertension.

Figures

Figure 1

Lung CT showing the presence in both lungs of (A-patient 1, B-patient 2, D-patient 4) diffuse poorly defined ground-glass centrilobular nodules, (C-patient 3) diffuse ground-glass opacification.

Figure 2

Haematoxylin and eosin (H&E) staining of lung tissues from lung biopsy in patient 4 showed (A) thickening of alveolar septa with infiltration of lymphocytes, multinuclear giant cells and cholesterol crystallization in the local alveolar lumen, and a small amount of protein deposition, (B) congestion of small vessel, destruction of vascular wall accompanied by neutrophil infiltration which suggested vasculitis, (C) thickening of small vessel walls and occlusion of the lumen.

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