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BRIEF REPORT

Disseminated nocardiosis and anti-GM-CSF antibodies

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Abstract

Infections that are unusually severe or caused by opportunistic pathogens are a hallmark of primary immunodeficiency (PID). Anti-cytokine autoantibodies (ACA) are an emerging cause of acquired immunodeficiency mimicking PID. *Nocardia* spp. are Gram-positive bacteria generally inducing disseminated infections in immunocompromised patients, but seldom also occurring in apparently immunocompetent hosts. Anti-GM-CSF autoantibodies are associated with autoimmune pulmonary alveolar proteinosis (PAP). In those patients, an increased incidence of disseminated nocardiosis and cryptococcosis has been observed. It is unclear whether the PAP or the autoantibodies predispose to the infection. We report an apparently immunocompetent woman presenting with disseminated nocardiosis without any evidence of PAP. Clinical data and radiological images were retrospectively collected. Lymphocyte populations were analyzed by flow cytometry. Anti-GM-CSF autoantibodies were measured by ELISA. A 55-year-old otherwise healthy woman presented with cerebral and pulmonary abscesses. Personal and familial history of infections or autoimmunity were negative. After extensive examinations, a final diagnosis of disseminated nocardiosis was made. Immunologic investigations including neutrophilic function and IFN-γ/IL-12 circuitry failed to identify a PID. Whole-exome sequencing did not find pathogenic variants associated with immunodeficiency. Serum anti-GM-CSF autoantibodies were positive. There were no clinical or instrumental signs of PAP. Trimethoprim-sulfamethoxazole and imipenem were administered, with progressive improvement and recovery of the infectious complication. We identified anti-GM-CSF autoantibodies as the cause of disseminated nocardiosis in a previously healthy and apparently immunocompetent adult. This case emphasizes the importance of including ACA in the differential diagnosis of PID, especially in previously healthy adults. Importantly, anti-GM-CSF autoantibodies can present with disseminated nocardiosis without PAP.

Keywords Nocardiosis · Anti-GM-CSF autoantibodies · Anti-cytokine autoantibodies · Immunodeficiency

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Introduction

Infections that are unusually severe or caused by opportunistic pathogens are a hallmark of primary immunodeficiency (PID). Recently, anti-cytokine autoantibodies (ACA) have gathered much attention as the cause of some of those infections. Therefore, ACA are an emerging etiology of acquired immunodeficiency which have been classified as PID phenocopies [1]. *Nocardia* spp. are Gram-positive environmental bacteria generally regarded as opportunistic pathogens [2]. Disseminated *Nocardia* infections have been reported among transplanted patients (solid organ transplantation, allogeneic stem cell transplantation) or patients receiving corticosteroids. However, several cases of nocardiosis have been described in apparently immunocompetent hosts [3]. Increased serum levels of anti-GM-CSF autoantibodies have been reported in patients with pulmonary alveolar proteinosis (PAP), disseminated *Nocardia* and *Cryptococcus gattii* infections, and have been associated to disease pathogenesis [4]. Herein, we report a case of an apparently immunocompetent patient presenting with disseminated nocardiosis.

Materials and methods

Patient history and imaging were retrospectively collected from the clinical records. Flow cytometry was used to analyze lymphocyte populations and intracellular cytokine production after polyclonal stimulation as previously reported [5]. Anti-GM-CSF autoantibodies were detected by an ELISA assay as previously reported [6].

Case presentation

A 55-year-old woman presented to the emergency department with new-onset left hemisoma hypoesthesia and 1-week history of headache. She had arterial hypertension in good control with daily ramipril. Family history was unremarkable. She did not smoke tobacco or use illicit drugs. She reported no exposure to toxic substances. Computed tomography (CT) scan of the head demonstrated two encephalic lesions. Chest X-ray revealed a right pulmonary nodule (11 mm in largest diameter). A contrast-enhanced brain magnetic resonance imaging (MRI) confirmed two rounded polylobed rim-enhanced cortical-subcortical lesions localized in the right basal occipital lobe and right frontal lobe (17 mm and 20 mm in largest diameter, respectively). Perilesional edema was present, and the lesions were deemed compatible with metastasis (Fig. 1). Contrast-enhanced total body CT scan confirmed a pseudo-nodular lesion with

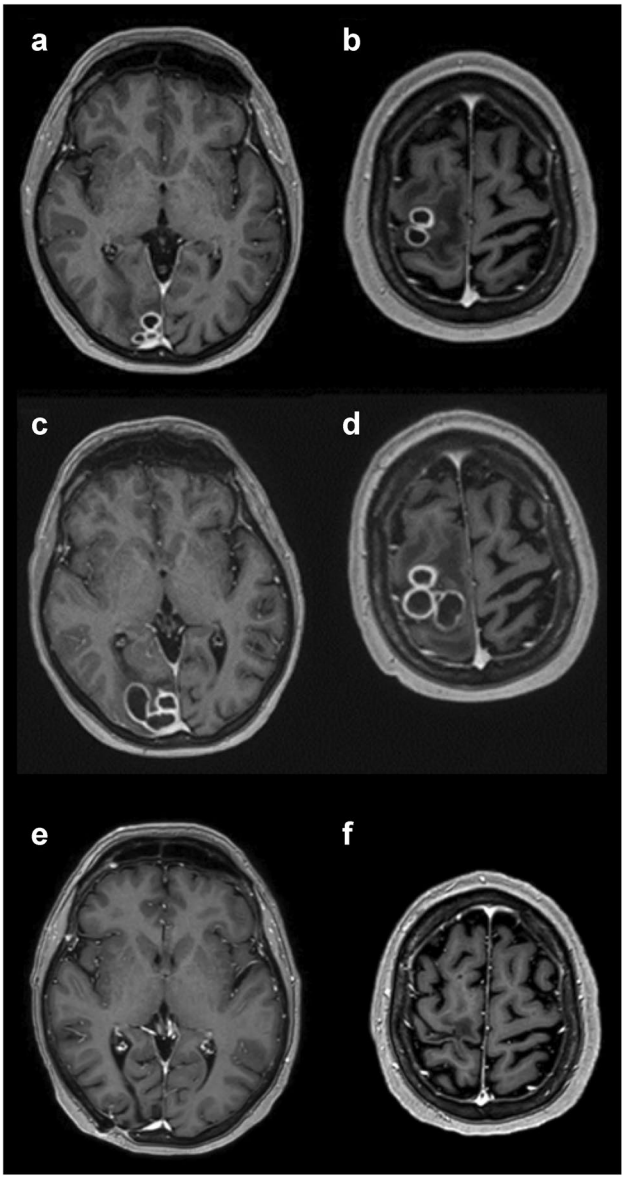

Fig. 1 Brain MRI (1,5T), axial contrast-enhanced T1w turbo field echo (TFE) images. Multiple cortical-subcortical rim-enhancing cerebral lesions located in the occipital lobe parasagittal area (**A**) and in the frontal lobe at the vertex (**B**). Later scans after 37 days showing lesions growth (**C**–**D**). Follow-up imaging after 28 months showing no contrast enhancement in the previously affected areas. Residual mild encephalomalacia in the occipital lobe parasagittal area (**E**) and gliosis in the frontal lobe at the vertex (**F**)

spiculated margins in the posterior segment of the right superior lung lobe (20 × 14 × 14 mm) close to a centrally cavitated nodular formation (12 × 10 mm) (Fig. 2). Pulmonary lesions were intensely positive on ^{18}F-FDG-PET. No pathological findings were found in the abdomen. Routine blood tests were within normal limits. Inflammatory markers were negative. Therapy with dexamethasone and mannitol was started with progressive improvement of neurological

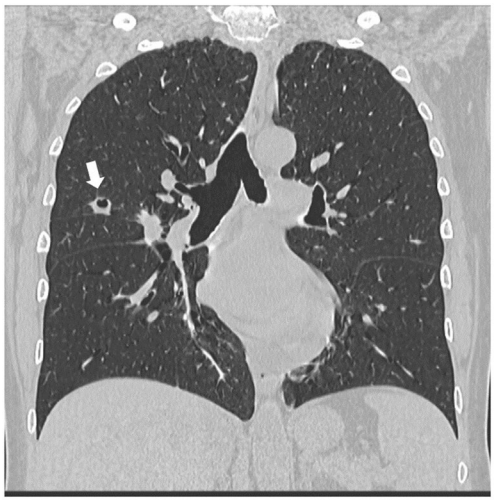

Fig. 2 Coronal chest CT scans showing two nodular lesions with spiculated margins in the posterior segment of the right upper lobe. One lesion is cavitated (white arrow)

symptoms. Lung needle biopsy was performed, and no neoplastic proliferation was found in the specimen, whereas alveolar macrophages mixed with inflammatory cells were detected. Following multidisciplinary discussion, open right superior pulmonary lobectomy and hilar mediastinum lymphadenectomy were performed. Histological analysis of the operative specimens resulted negative for malignancy and showed non-specific exudative and productive necrotizing lung inflammation and reactive lymphadenopathy, respectively. Potential primary foci of infection were excluded by fibro-bronchoscopy, dental CT scan, and transesophageal echocardiogram. A contrast-enhanced brain MRI was repeated 37 days later and demonstrated an increase of the two brain lesions (Fig. 1) as well as a new-onset lesion in the right parietal lobe.

Diagnostic excision of the right frontal encephalic mass was collectively proposed. During intraoperative inspection, the mass appeared to be a brain abscess, but routine cultural examinations were initially negative. The histological examination showed exudative-necrotizing inflammation. Empiric antibiotic therapy with meropenem 1000 mg t.i.d. was started, but a total body CT scan performed 16 days later displayed thickening and cavitation of the pulmonary nodules. Similarly, the right occipital encephalic lesion was increasing as shown on contrast-enhanced brain MRI. Linezolid 600 mg b.i.d. was added, and the patient underwent a second brain surgery to evacuate the occipital lesion.

Five samples (a–e) were microbiologically examined and *Nocardia* spp. grew both on liquid media (modified Middlebrook 7H9 broth—BD BBL MGIT/BD BBL MGIT PANTA/BD BACTEC MGIT Growth Supplement, Becton Dickinson) and on solid media (Lowenstein Jensen agar, Biolife). Only in two samples of five *Nocardia* grew on liquid medium, and in those, time to positivity was shorter in liquid medium versus solid medium (a: 6th day versus 3rd week; b: 19th day versus 9th week; c, d, and e: growth only on Lowenstein Jensen agar in 2nd, 3rd, and 5th week). For susceptibility testing, broth microdilution, according to CLSI, was performed. The MICs (µg/mL) and interpretation according to CLSI were as follows: trimethoprim-sulfamethoxazole ≤ 0.25 (S), doxycycline ≤ 0.12 (S), imipenem ≤ 2 (S), tobramycin ≤ 1 (S), ciprofloxacin > 4 (R), amikacin ≤ 1 (S), clarithromycin 0.25 (S), linezolid ≤ 1 (S), ceftriaxone ≤ 4 (S), amoxicillin-clavulanate > 64/32 (R), and moxifloxacin > 8 (R).

Antibiotic therapy was switched to i.v. trimethoprim/sulfamethoxazole (20 mg/kg/day, equally divided every 8 h) and i.v. imipenem 500 mg q.i.d. for 2 months.

On contrast-enhanced brain MRI performed 11 days after the initiation of specific antibiotic therapy, the right occipital abscess was reduced, the right parietal lesion was stable, and no new findings were found.

Chest CT scan showed reduction of the cavitated nodule in the right inferior lobe. The patient progressively improved: normal gait recovered, but residual hypoesthesia and hyposthenia of the left hemisoma persisted. After 30 days, follow-up imaging was further improved. Chest CT showed complete resolution of the lesions. However, on brain MRI, there was persistent contrast enhancement in some areas. A clinical immunology consult was sought to exclude an immunodeficiency state. Typically, disseminated *Nocardia* infection develops in patients with secondary immunosuppression (i.e., solid-organ or hematopoietic stem cell transplantation, lymphoma, long-term immunosuppressive treatments) or HIV infection [2]. Additionally, disseminated nocardiosis can be observed in patients with PID, mainly in cellular and combined immunodeficiencies, but also in humoral and phagocytic defects [3, 4]. The patient had modest leukopenia and lymphopenia, but B and T lymphocyte subsets were within the normal range excluding CD4+ lymphopenia [7]. Defects in killing of neutrophils and macrophages might also be associated with nocardiosis, such as CGD, which was excluded by a normal dihydrorhodamine test (DHR). Immunodeficiencies affecting the IL-12/IFN-γ axis were considered. The TB-IGRA test was indeterminate, and the production of IFN-γ after polyclonal stimulation of T cell subpopulations was reduced. However, both findings were deemed to be secondary and attributed to the early initiation of steroid therapy during the hospital admission. Moreover, MSMD was deemed unlikely because the patient received BCG vaccination during childhood without any complications. Finally, whole exome sequencing failed to identify pathogenic variants in PID-associated genes. Considering the etiology and severity of the opportunistic infection in this patient, we looked for other causes

of immunodeficiency. Anti-cytokine autoantibodies are an emerging cause of infectious disease in previously healthy adults and are recognized as phenocopies of congenital defects [6]. Anti-GM-CSF autoantibodies were measured and found to be elevated (57 mcg/mL, normal range ≤ 3), using an ELISA assay as previously described by Uchida et al. [6]. Re-evaluation of the previous chest CT scans as well as during follow-up did not detect any sign of PAP.

After 2 months of IV imipenem and trimethoprim/sulfamethoxazole, she was switched to PO trimethoprim/sulfamethoxazole, 2 tablets every 8 h, and doxycycline was initiated but discontinued due to gastric intolerance. The patient continued with trimethoprim/sulfamethoxazole monotherapy for 3 months, until she developed pancytopenia in the setting of SARS-CoV-2 infection. The etiology of the pancytopenia remained unclear; trimethoprim/sulfamethoxazole toxicity and post-infectious or drug-induced SLE were considered. Trimethoprim/sulfamethoxazole was discontinued, and she was hospitalized for management of the pancytopenia and IV antibiotic therapy. She was switched to intravenous ceftriaxone 2 g every b.i.d. and oral minocycline 100 mg b.i.d. for ten additional months. After 1 year of combined antibiotic therapy, ceftriaxone was discontinued, and she continued to take minocycline 100 mg twice a day. Minocycline was discontinued after an additional 12 months. She did not develop any brain or lung complications during this time (supplementary figure with display of the antibiotic therapy timeline).

Follow-up brain MRI after 28 months showed resolution of the brain lesions with leftover scarring (Fig. 1E, F). After 28 months of follow-up, the patient did not develop any respiratory symptoms. Spirometry was within normal limits (FVC 97% predicted, FEV1 102% of predicted) and normal CO diffusion (DLCO 77% of predicted).

Discussion

We identified anti-GM-CSF autoantibodies (AAbs) as the cause of disseminated nocardiosis in a previously healthy and apparently immunocompetent adult. Anti-GM-CSF AAbs were first described in 1999 and are recognized as the leading cause of autoimmune pulmonary alveolar proteinosis (aPAP) [8]. GM-CSF has a primary role as a regulator of maturation and differentiation for many cell types including macrophages, neutrophils, lymphocytes, fibroblasts, endothelial cells, and alveolar epithelial cells [9]. AAbs block GM-CSF binding to its receptor, thereby inhibiting differentiation and function of alveolar macrophages [10]. GM-CSF is critical for surfactant clearance and in the host's defense against pathogens. It regulates the functions of macrophages, increasing their ability as antigen-presenting cells and complement-mediated phagocytosis, as well as the functions of neutrophils, increasing their adherence and killing activity [11]. For these reasons, anti-GM-CSF AAbs may account for disseminated infections by intracellular pathogens. *Nocardia* spp. activates GM-CSF-mediated phosphorylation of STAT5 in monocytes [11]. In addition, GM-CSF is essential against *Cryptococcus* spp. [12] although it is unclear why anti-GM-CSF AAbs increase specifically the risk of central nervous system (CNS) infections by *Cryptococcus gattii* compared to *Cryptococcus neoformans* [13].

In the present case report, disseminated nocardiosis presented with cerebral and pulmonary involvement without PAP. Rosen et al. first described patients with disseminated nocardiosis and anti-GM-CSF autoantibodies but without PAP. PAP may facilitate *Nocardia* dissemination into the blood circulation due to alveolar-capillary barrier damage and consequently extrapulmonary diffusion [14]. Other mechanisms may underlie the diffusion of the pathogen. In fact, clinical cases—as this patient—showing direct development of systemic infection in the absence of PAP could demonstrate that immune dysfunction may be present at a systemic level. The presence of anti-GM-CSF AAbs in this patient can be a sufficient cause of *Nocardia* spp. infection and dissemination in the absence of PAP. So far, only a few cases of disseminated nocardiosis without PAP have been reported, mainly with CNS involvement [15]. PAP can occur several months after manifestation of the infectious phenotype. Consequently, in the case of the presence of anti-GM-CSF AAbs, PAP should be excluded at the diagnosis and thoroughly considered during follow-up. Levels of anti-GM-CSF AAbs seem not to be associated with disease severity. However, patients affected by PAP, cryptococcal meningitis, and disseminated nocardiosis were found to have a tendency towards higher levels of anti-GM-CSF AAbs [16]. The association between AAbs and the development of a particular clinical phenotype remains unclear. Therapeutic approaches with anti-GM-CSF monoclonal antibodies have been used in clinical trials in some inflammatory diseases associated with high levels of GM-CSF, including psoriasis and rheumatoid arthritis. Intriguingly, no increase in opportunistic infections [17] or PAP was reported. Vice versa, anti-GM-CSF AAbs have been detected in the sera of patients with inflammatory diseases such as Crohn's disease and ulcerative colitis. In these cases, levels correlate with severity, complications, and relapses [18]. Anti-GM-CSF AAbs have been reported also in patients with severe endometriosis [19]. It is still unknown whether there is an increased risk of opportunistic infections in these subjects. Currently, there are no specific treatment or prophylaxis recommendations for anti-GM-CSF-related infections. Our patient did not receive any specific treatment, whereas she continued antibiotic prophylaxis after the initial 6-week treatment. For patients who have developed aPAP, inhaled recombinant human GM-CSF is used, with unclear efficacy [20].

In conclusion, we identified anti-GM-CSF autoantibodies as the cause of disseminated nocardiosis in a previously healthy and apparently immunocompetent adult. This case emphasizes the importance of including ACA in the differential diagnosis of immunodeficiencies, especially in previously healthy adults. In addition to severe opportunistic infections, recognition of anti-GM-CSF AAbs warrants careful screening for PAP, but separate clinical presentations are possible.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s10096-024-04785-z.

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Declarations

Consent for publication Informed consent was obtained from the patient for publication of the report and any accompanying images.

Conflict of interest The authors declare no competing interests.

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