Clinical Immunology Communications 6 (2024) 5–8

Contents lists available at ScienceDirect

Clinical Immunology Communications

journal homepage: www.elsevier.com/locate/clicom

First report of Mycobacterium chimaera infection in a patient with chronic granulomatous disease

Nancy E Aguilar Gómez [a], Uriel Pérez Blanco [b], Patricia Saltigeral Simental [b], Sara Espinosa Padilla [a], Jacinta Bustamante [c,d,e], Lizbeth Blancas Galicia [a,*]

[a] Department of Infectious Diseases, National Institute of Pediatrics, 1 IMAN Ave, 04530, Mexico City, Mexico
[b] National Institute of Pediatrics, 1 IMAN Ave, 04530, Mexico City, Mexico
[c] Necker Branch, Necker Hospital for Sick Children, 24 Blvd. Montparnasse, 75015, Paris, France
[d] Imagine Institute, Paris Cité University, 24 Blvd. Montparnasse, 75015, Paris, France
[e] Study Center for Primary Immunodeficiencies, Necker Hospital for Sick Children, AP-HP, 149 Sèvres St, 75743, Paris, France

ARTICLE INFO

Keywords:
Hemophagocytic syndrome
Environmental mycobacteria
Chronic granulomatous disease
Hemophagocytic lymphohistiocytosis

ABSTRACT

Chronic granulomatous disease (CGD) is an inborn error of immunity. NADPH oxidase is an enzyme complex that produces various reactive oxygen species, such as superoxide anions and hydrogen peroxide. Mycobacterial infections in CGD are commonly observed in countries with a high prevalence of these microorganisms, such as those receiving the BCG vaccination at birth or having a high prevalence of tuberculosis. Non-tuberculous mycobacteria (NTM) infections are rare in CGD. The patient also presented with hemophagocytic lymphohistiocytosis, which resolved with gammaglobulin and cyclosporine. Herein, we describe the first case of *M. chimaera* infection in a female patient with autosomal recessive CGD caused by a pathogenic variant in *CYBA*.

Chronic granulomatous disease (CGD) is an inborn error of immunity (IEI). NADPH oxidase is an enzyme complex that produces various reactive oxygen species, such as superoxide anions and hydrogen peroxide. Pathogenic variants in cytochrome B (*CYB*)-*B, CYBA, CYBC1*, neutrophil cytosolic factor (*NCF*)-*1, NCF2*, and *NCF4* genes of the NADPH oxidase enzyme complex are responsible for the clinical phenotype of CGD [1]. CGD has an X-linked or autosomal recessive inheritance pattern [1]. Patients with CGD present with various inflammatory and infectious diseases [1]. They are susceptible to fungal and bacterial infections, including mycobacterial infections [1]. *Mycobacterium tuberculosis* infections have been observed in *Bacillus* Calmette–Guérin (BCG)-vaccinated patients with CGD [2]. Mycobacterial infections in CGD are commonly observed in countries with a high prevalence of these microorganisms, such as those receiving the BCG vaccination at birth or those with a high prevalence of tuberculosis [2]. Nontuberculous mycobacterial (NTM) infections are rare in CGD [2] Here, we described the first *M. chimaera* infection in a female patient with autosomal recessive CGD caused by a pathogenic variant in *CYBA*.

A 16-year-old female from Mexico, the only daughter of consanguineous parents, received the BCG vaccine at birth and exhibited no adverse reactions. At four years of age, the patient developed chronic sinusitis and presented with pneumonia. At five years of age, the patient exhibited cervical lymphadenitis. The biopsy revealed chronic granulomatous inflammation and caseating necrosis with acid-fast bacteria, and a positive tuberculin test result was obtained. Therefore, the patient was diagnosed with tuberculosis, and antituberculosis therapy was initiated with 60 doses of rifampicin (250 mg), isoniazid (33 mg), pyrazinamide (200 mg), and ethambutol (150 mg) in the intensive phase and 45 doses of the same drugs in the maintenance phase. At nine years of age, the patient presented with pneumonia caused by *Aspergillus* spp. However, the patient's condition improved after treatment with voriconazole (9 mg/kg/day) for 180 d. During hospitalization, CGD was diagnosed based on abnormal dihydrorhodamine test results and the absence of $p22^{phox}$ protein expression on flow cytometry. Sanger sequencing revealed a homozygous pathogenic variant of *CYBA*: c.4_24del (p.Gly2_Met8del). The patient was discharged and instructed to continue treatment with trimethoprim/sulfamethoxazole (TMP/SMX), itraconazole, and recombinant interferon-γ. At an outpatient follow-up, the radiological studies revealed pulmonary damage (Figs. 1A and B).

Abbreviations: BAL, bronchoalveolar lavage; CGD, chronic granulomatous disease; NTM, non-tuberculous mycobacteria; SMX, sulfamethoxazole; TMP, trimethoprim.

* Corresponding author at: National Institute of Pediatrics, 1 IMAN Ave, Mexico City, 04530, Mexico.
E-mail address: blancas.lizbeth@gmail.com (L. Blancas Galicia).

https://doi.org/10.1016/j.clicom.2024.06.001
Received 25 April 2024; Received in revised form 14 June 2024; Accepted 20 June 2024
Available online 23 June 2024

At 16 years of age, the patient was admitted to the hospital with a fever (38.6 °C), respiratory distress syndrome, and multiple-focus pneumonia (Fig. 1C). Empirical treatment with voriconazole (8 mg/kg/day), cephalothin (150 mg/kg/day), cefotaxime (150 mg/kg/day), and TMP/SMX (20 mg/kg/day) was initiated. However, the patient experienced hemodynamic and respiratory deterioration and required mechanical ventilation. The patient exhibited secondary hemophagocytic lymphohistiocytosis (HLH), with fever > 38.5 °C, splenomegaly, 7.3 (11.6–15) mg/dL hemoglobin, 15,000/mm^3 (150,000–450,000/mm^3) platelets, 395 (< 150) mg/dL triglycerides, 90 (200–400) mg/dL fibrinogen, and 3489 (24–336) μg/L ferritin. Additionally, the patient's condition was complicated with hepatosplenomegaly, ascites, and acute liver failure (the diagnostic results showed the following: alanine aminotransferase: 78 [17–63] IU/L; aspartate aminotransferase: 274 [15–41] IU/L; total bilirubin: 8.93 [0.30–1.20] mg/dL; indirect bilirubin: 3.26 [0.01–1.10] mg/dL; alkaline phosphatase: 346 [38–126] IU/L; lactate dehydrogenase: 810 [105–233] IU/L; and gamma-glutamyl transpeptidase: 75 [7–50] IU/L). The patient also exhibited other complications, such as disseminated intravascular coagulation, with 16,000/mm^3 (150,000–450,000/mm^3) platelets, prothrombin time > 2 min (11–13.5 s), partial thromboplastin time > 2 min (25–45 s), 38.7 (200–400) mg/dL fibrinogen, and 21 (< 0.5) μg/mL D-dimer. The patient was treated with intravenous gamma globulin (1 g/kg/dose) and cyclosporine (2 mg/kg/day) for secondary HLH, with a favorable response.

As the fever persisted despite the initial antimicrobial regimen, subsequent treatment with azithromycin (10 mg/kg/day) and linezolid (30 mg/kg/day; Fig. 1A) was initiated. *Salmonella* spp. was identified in central and peripheral blood cultures, and meropenem (20 mg/kg/dose) treatment was initiated. Bronchoalveolar lavage (BAL) samples were negative for *M. tuberculosis* but positive for amphotericin B-resistant *Aspergillus niger* (2155 copies/mL). Therefore, the patient was treated with voriconazole and caspofungin (50 mg/m^2 body surface area). Despite treatment, the patient's clinical condition worsened, and chest CT revealed cavitation in the middle and lower lobes of the posterior segment (Fig. 1D). Consequently, sputum smear microscopy was performed. Positive Ziehl–Neelsen staining and negative GeneXpert MTB/RIF PCR results indicated the presence of NTM infection. Treatment with rifampicin (150 mg), isoniazid (75 mg), pyrazinamide (400 mg), ethambutol (300 mg), and clarithromycin (15 mg/kg/day) was initiated (Fig. 1A). Growth of *M. chimaera* was observed in a culture of the BAL fluid, and levofloxacin (10 mg/kg/dose) was subsequently added to the anti-tuberculosis regimen. Sputum cultures revealed multidrug-resistant carbapenemase-producing *Pseudomonas aeruginosa* sensitive to amikacin, *Citrobacter freundii*, and *Klebsiella variicola* strains. Therefore, amoxicillin with clavulanic acid (50 mg/kg/day), amikacin (10 mg/kg/day), and colistin (2.5 mg/kg/dose) were added to the treatment regimen. However, the patient died of a pulmonary hemorrhage.

Here, we report a case of CGD with various infections, including an *M. chimaera* infection. *M. tuberculosis* infections are observed in BCG-vaccinated patients with CGD; however, NTM infections are rare. To date, only 11 patients with CGD and NTM infections caused by *M. avium, M. immunogenum, M. gordonae, M. mucogenicum, M. fortuitum, M. flavensces, M. chelonae, M. abscessus*, and *M. intracellulare* were reported. Interestingly, there have been reports of two or more NTMs in the same patient, *M. mucogenicum* and *M. bovis* or M. *chelonae, M. abscessus, M. intracellulare*, and *M. tuberculosis* [3–10].

Only six patients, including the present case (four *CYBB* and two *CYBA* genes), have been identified with CGD and NTM infections [1,3–5,8]. Patients with both X-linked and autosomal recessive mutations may present with NTM infections. The case reported here (Zielh-Neelsen--positive and GeneXpert MTB/RIF-negative) illustrates the need to incorporate methods such as culture and genetic identification of mycobacteria beyond Zielh–Neelsen staining or GeneXpert MTB/RIF to detect NTM infections in patients with CGD. In these patients, the NTM infection may be localized to the lymph nodes, bones, kidneys, synovial fluid, skin, and lungs [1,3–10]. The lungs were the most affected in the present case (Table 1). Salvator *et al.* studied patients ≥ 16 years with CGD and lung disease and concluded that patients have more pulmonary affection as they age. Furthermore, they concluded that a history of lung disease in childhood favored the recurrence of fungal infections and

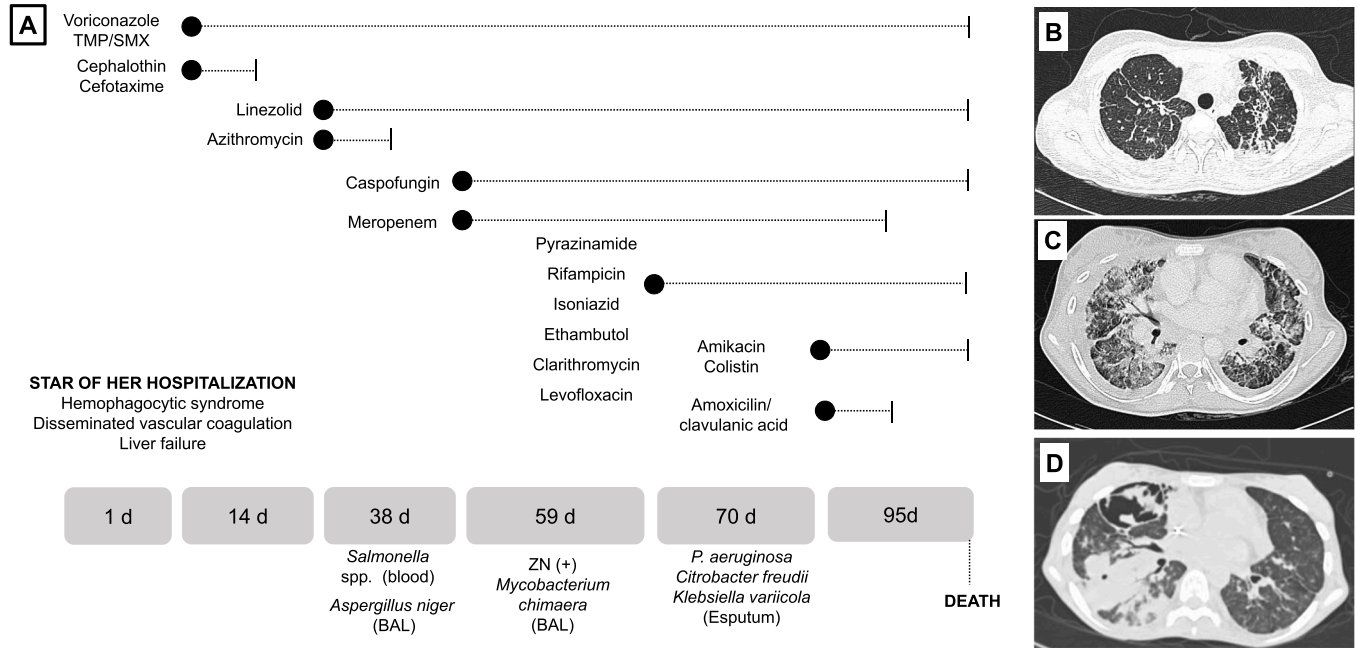

Fig. 1. Radiological and infectious agent test findings. (A) A timeline showing the antimicrobial agents administered and isolates identified in the patient chronologically from days 1 to 59 of hospitalization. (B) Chest computed tomography (CT) scan four years before the reported hospitalization (at 12 years old) revealed bilateral bronchiectasis. (C) Chest CT at the time of reported hospitalization (at 16 years old) revealed multiple hyperdense images, suggesting multiple-focus pneumonia. (D) Chest CT 50 d after hospitalization. In the left lung, hypodense images corresponding to cavitation gradually increased. TMP/SMX, trimethoprim with sulfamethoxazole; BAL, bronchoalveolar lavage; ZN, Ziehl–Neelsen.

Table 1
Genetic and clinical findings of six patients with chronic granulomatous disease (CGD) and non-tuberculosis mycobacterial (NTM) infection. M (male), F (female), yo (years old), mo (months old), DHR (dihydrorhodamine), NBT (nitroblue tetrazolium), ND (not described). [¶]*M. flavescens* resistant to isoniazid. Six patients mentioned within four cohorts of patients with CGD are not included because only the NTM species and the organs involved are mentioned.

Patient (P) Quote	P1 Current case	P2 Labrosse R, 2017 (3)	P3 Weening RS, 2000 (4)	P4 Ohga S, 1997 (5)	P5 Allen DM, 1993 (6)	P6 Chusid MJ, 1975 (7)
Gender	F	M	M	M	M	M
Ethnicity/Origin	Mexican	French Canadian	Unknown	Japanese	Chinese	Unknown
First described manifestation (s)	Cervical lymphadenitis	*S. aureus* bacteremia	Cervical lymphadenitis	Pneumonia	*Salmonella blockley* Pneumonia	Pneumonitis, staphylococcal thigh & facial infection
Age at CGD diagnosis	9 yo	6 mo	18 yo	10 mo	62 yo	27 yo
Diagnosis method	DHR	DHR	ND	DHR	NBT	NBT
Gene & Mutation	*CYBA* (c. 4_24del) p.Gly2_Met8del	*CYBB* (c. 469C>T) p.Arg157*	*CYBB* c. C-52T	*CYBB* (ND)	ND	ND
Mycobacterial Infection Data						
Age at infection	16 yo	5 yo	44 yo	10 mo	62 yo	27 yo
Detected mycobacteria	*M. chimaera*	*M. avium*	*M. avium*	*M. avium*	*M. flavescens*	*M. fortuitum*
Localization	Lung	Lung	Lymph node	Lung & mediastinal lymph node	Lung, synovial fluid, & skin	Lung & bone
Treatment regimen	Rifampicin, ethambutol, pyrazinamide, isoniazid, levofloxacin & clarithromycin	Ethambutol, rifabutin & azithromycin	ND	Drainage & netilmicin	Rifampicin, ethambutol & streptomycin[¶]	Isoniazid, rifampicin, ethionamide & kanamycin
Infection remission	No *Salmonella* spp.	Yes	ND	Yes	No	Yes
NTM & Coinfection (microorganism)	Pulmonary aspergillosis (*A. niger*) Pneumonia (*P. aeruginosa, C. freudii, K. variicola*)	Pneumonia (*Actimomyces* spp.)	No	Skin abscesses (*S. marcescens*)	Pulmonary cryptococcosis	No
Clinical course	Dead	Alive	ND	Alive	Dead	Alive

[¶] Isoniazid-resistant *Mycobacterium flavescens*.

noninfectious pulmonary events in adulthood [9]. Coinfection with NTM occurs in CGD patients with pneumonia due to *Actinomyces* spp. and *Cryptococcus* spp. [3,6]. In the patient reported here, the chronic lung damage associated with an aspergillosis event, together with co-infection and HLH-related changes, may have favored *M. chimaera* infection. The clinical outcomes of NTM infections in patients with CGD remain unclear, as only a few cases have been reported. Only four reported cases provided information on the patient clinical outcomes [3, 5–7] (Table 1; Tables S1 and S2). The patients with CGD and BCG or *M. tuberculosis* infections were mostly from tuberculosis-endemic areas [2]. However, patients with NTM infections can be from any country with a variable prevalence of tuberculosis [1,3–5,8]. Therefore, NTM infections should be investigated in patients with CGD and fever without an infectious focus.

Certain IEIs, different from those that cause primary HLH, have been associated with developing secondary HLH, including CGD [11]. The most common triggering factor of HLH in CGD is infectious, especially *Burkholderia cepacia* and *Leishmania* spp., which have also been described in coinfection with bacteria [11]. In the present case, Salmonella was the infectious agent isolated during the HLH event. The onset of HLH in CGD has been described from one year of age to adulthood [11]; the patient described was 16 years of age at the time of presentation. Patients with CGD and HLH *versus* patients with CGD and primary HLH respond to less aggressive therapies such as steroids and gammaglobulin [11], as in the case reported here. HLH is an inflammatory manifestation of CGD that should be considered in the presence of severe infectious events for early detection and treatment. Overall, this case illustrates the susceptibility of patients with CGD to NTM infection. Clinicians should test for these infectious agents to suggest correspondingly suitable treatment regimens.

Declaration of generative AI and AI-assisted technologies in the writing process

None.

CRediT authorship contribution statement

Nancy E Aguilar Gómez: Writing – review & editing, Methodology, Investigation. **Uriel Pérez Blanco:** Writing – review & editing, Writing – original draft. **Patricia Saltigeral Simental:** Methodology, Investigation. **Sara Espinosa Padilla:** Writing – review & editing, Supervision. **Jacinta Bustamante:** Writing – review & editing, Supervision. **Lizbeth Blancas Galicia:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Conceptualization.

Declaration of competing interest

The authors declare no competing interests (Nancy E. Aguilar Gómez, Uriel Pérez Blanco, Patricia Saltigeral Simental, Sara Espinosa Padilla, Jacinta Bustamante, Lizbeth Blancas Galicia) The authors declare no competing interests.

Data availability

Data will be made available on request.

Funding

The writing of this report was supported by the FUMENI A.C.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.clicom.2024.06.001.

References

[1] L. Blancas-Galicia, E. Santos-Chavez, C. Deswarte, Q. Mignac, I. Medina-Vera, X. Leon-Lara, et al., J. Clin. Immunol. 40 (3) (2020) 475–493.
[2] C. Deffert, M.G. Schappi, J.C. Pache, J. Cachat, D. Vesin, R. Bisig, et al., PLoS. Pathog. 10 (9) (2014) e1004325.
[3] R. Labrosse, J. Abou-Diab, A. Blincoe, G. Cros, T.M. Luu, C. Deslandres, et al., Front. Immunol. 8 (2017) 1167.
[4] R.S. Weening, M. De Boer, T.W. Kuijpers, V.M. Neefjes, W.W. Hack, D. Roos, Clin. Exp. Immunol. 122 (3) (2000) 410–417.
[5] S. Ohga, K. Ikeuchi, R. Kadoya, K. Okada, C. Miyazaki, S. Suita, et al., J. Infect. 34 (2) (1997) 147–150.
[6] D.M. Allen, H.H. Chng, J. Infect. 26 (1) (1993) 83–86.
[7] M.J. Chusid, J.E. Parrillo, A.S. Fauci, JAMA 233 (12) (1975) 1295–1296.
[8] T. Li, X. Zhou, Y. Ling, N. Jiang, J. Ai, J. Wu, et al., Front. Immunol. 10 (2019) 73.
[9] H. Salvator, N. Mahlaoui, E. Catherinot, E. Rivaud, B. Pilmis, R. Borie, et al., Eur. Respir. J. 45 (6) (2015) 1613–1623.
[10] D. Abd Elaziz, R. El Hawary, S. Meshaal, R. Alkady, S. Lotfy, A. Eldash, et al., J. Clin. Immunol. (2023).
[11] S.F. Bode, S. Ammann, W. Al-Herz, M. Bataneant, C.C. Dvorak, S. Gehring, et al., Haematologica 100 (7) (2015) 978–988.