OPEN

Abnormal T Cells Function Associated With Intraspinal Cold Abscess Caused by Macrolide-resistant *Mycoplasma pneumoniae* in a Patient With X-linked Agammaglobulinemia

Ying-Ying Jin, MD, PhD,† Jing Wu, PhD,†‡ Fei Ding, MD,* Hua Huang, MD,* Xue-Mei Xu, MD,* Qi-Min Chen, MD,§ Min-Zhi Yin, MD,¶ Yu-Min Zhong, MD, PhD,‖ and Yan-Liang Jin, MD, PhD**

Abstract: Intraspinal cold abscesses caused by macrolide-resistant *Mycoplasma pneumoniae* in patients with X-linked agammaglobulinemia have not yet been described to our knowledge. Here we describe a patient with X-linked agammaglobulinemia who developed an intraspinal cold abscess caused by macrolide-resistant *M. pneumoniae*. Genetic analysis revealed a hemizygous c.1566 + 1G > C (IVS15 + 1G > C) mutation in *BTK* gene. The patient showed relatively naive T cells and a significant proliferative defect.

Key Words: T cells, intraspinal cold abscess, macrolide-resistant mycoplasma, X-linked agammaglobulinemia

(*Pediatr Infect Dis J* 2025;44:e45–e48)

X-linked agammaglobulinemia (XLA) is characterized by severe or recurrent infections, panhypogammaglobulinemia, lack of recall humoral response to antigens and markedly reduced numbers of B cells (less than 2%) with an X-recessive pattern of inheritance that affects males.[1] The disease is caused by mutation of the *Brutons tyrosine kinase* (*BTK*) gene, which affects the differentiation-transition blockage of B cell progenitors to mature B lymphocytes.[2] Patients with XLA usually present with recurrent bacterial infections, mainly in the respiratory and gastrointestinal tracts. The most common organism identified was *Haemophilus infuenzae*, followed by *Staphylococcus aureus* and *Streptococcus pneumonia*.[3] Mycoplasma infection has been reported in patients with XLA, especially *Ureaplasma*, mainly presenting with pneumonia or arthritis.[4,5] Here, we describe a XLA patient with an intraspinal cold abscess caused by macrolide-resistant *Mycoplasma pneumoniae* when administered immunoglobulin replacement therapy, which has not yet been described to our knowledge, and his T-cell function was further analyzed. This case may cast new light on the spectrum of infections in patients with XLA and may help us pay more attention to the roles of T cells in XLA.

CASE

A 3-year-old male child, being the third offspring of unrelated parents without a familial history of immunodeficiency (as detailed in Figure, Supplemental Digital Content 1, http://links.lww.com/INF/F751), initially experienced an infection at the age of 1 year. This infection manifested as pneumonia, subsequently complicated by otitis media, plantar blister suppuration and the development of a subcutaneous abscess on the extensive region of the right thigh.

At the age of two and a half years, the individual experienced a recurrence of pneumonia along with the development of arthritis in the left wrist joint and right sternoclavicular region accompanied by elevated white blood cell, C-reactive protein and erythrocyte sedimentation rate. On examination, the RNA test for *M. pneumoniae* in the respiratory tract[6] yielded a negative result. Laboratory findings revealed a significantly low level of IgG at 0.07 g/L, which falls below the normal range of 4.24–10.51 g/L, IgM at 0.20 g/L, which is also below the normal range of 0.48–1.68 g/L, and IgA at 0.26 g/L, which remains within the normal range of 0.14–1.23 g/L. Further investigation showed an abnormal percentage and absolute number of $CD19^+$ B cells, with the percentage being 0.20% (normal range: 13.23%–26.39%) and the absolute number being 13.41 cells/μL (normal range: 461–1456 cells/μL). Given these findings, a suspicion of XLA was raised, and DNA analysis was subsequently performed. The patient was administered anti-infection therapy with cefuroxime, followed by linezolid, and received intravenous immunoglobulin (IVIG, 500 mg/kg) replacement therapy. On discharge, the arthritis had resolved. Subsequently, the patient received IVIG every 4 weeks, with trough IgG levels varying from 7.12 to 8.31 g/L.

Three months later, he was readmitted because of pain in the right lower limb with myasthenia. A mass of approximately 7 × 10 cm was noted on the left back with a slightly hard texture and no local increase in skin temperature. Enhanced magnetic resonance imaging revealed abnormal signals in the left erector spinalis muscle, psoas major muscle (about the level of T12–L3) and spinal canal (about the level of T12 lower edge to L3 upper edge) indicating abscess formation. Irregular hypoechoic areas with cord-like separation could be seen in the subcutaneous soft tissue muscle layer of the main complaint mass on the back, with a size range of approximately 32 × 25 × 47 mm. The cystic mass in the muscular layer of the back, with destruction of the surrounding pyramidal bone cortex, was an abscess, considering the possibility of infectious lesions (Fig. 1A). The laboratory data showed normal level of

Accepted for publication September 9, 2024

From the Departments of *Rheumatology/Immunology, †Allergy/Immunology Innovation Team, ‡Institute of Pediatric Translational Medicine, §Pediatric Surgery, ¶Pathology, and ‖Radiology, Shanghai Children's Medical Center, Shanghai Jiao Tong University School of Medicine, Shanghai, China.

The authors have no funding or conflicts of interest to disclose.

All relevant data are within the paper and its supporting information files. Y.-Y.J. and J.W. are the co-first authors. The authors have contributed equally to this work.

This study was conducted in accordance with the recommendations of the Shanghai Children's Medical Center, Shanghai Jiao Tong University School of Medicine, with written informed consent from all subjects. All participants provided written informed consent in accordance with the Declaration of Helsinki. The study protocol was approved by the Shanghai Children's Medical Center, Shanghai Jiao Tong University School of Medicine.

Supplemental digital content is available for this article. Direct URL citations appear in the printed text and are provided in the HTML and PDF versions of this article on the journal's website (www.pidj.com).

Address for correspondence: Yan-Liang Jin, MD, PhD, Departments of Rheumatology/Immunology, Shanghai Children's Medical Center, Shanghai Jiao Tong University School of Medicine, 1678 Dongfang Road, Pudong New District, 200127, Shanghai, China. E-mail: jinyanliang@scmc.com.cn.

ISSN: 0891-3668/25/442-e45e48
DOI: 10.1097/INF.0000000000004569

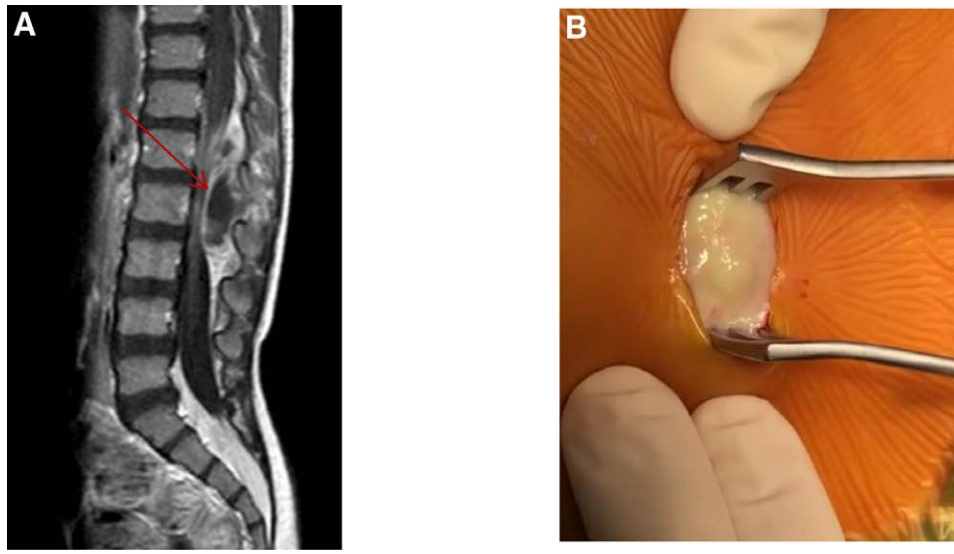

FIGURE 1. The MRI and intraoperative photo of abscess of the patient.A: Sagittal T1-W MRI demonstrating abscess formation with destruction of the surrounding pyramidal bone cortex (arrow). B: Intraoperative photo showed a large amount of bean curd residue like thin pus gushed out from the mass. MRI indicates magnetic resonance imaging.

IgG (7.86 g/L), low level of IgM (<0.18 g/L), IgA (<0.07 g/L) and IgE (<4.3 IU/mL) (data were obtained 3 weeks after the last IVIG infusion). The lymphocyte subsets are reported as follows: $CD3^+$ comprises 85.77%, which is above the normal range of 53.88%–72.87%; $CD3^+CD4^+$ comprises 41.89%, which falls within the normal range of 24.08%–42.52%; $CD3^+CD8^+$ comprises 37.86%, which is above the normal range of 19.00%–32.51%; $CD19^+$ comprises 0.04%, which is below the normal range of 13.23%–26.39%; and natural killer cells comprise 13.05%, which falls within the normal range of 7.21%–20.90%.

A surgical laminectomy and mass drainage procedure were conducted. During the surgical intervention, a significant quantity of pus-like material, akin to bean curd residue, was extracted from the mass (Fig. 1B). Cultures taken from the paravertebral abscesses, which contained purulent material, did not yield any growth of Mycobacterium tuberculosis, bacteria or fungi. Using next-generation sequencing technology,[7] the pathogen present in the pus was identified as *M. pneumoniae*. Following anti-infective therapy with azithromycin, analysis of the drug-resistant gene[8] revealed the presence of a macrolide-resistant *M. pneumoniae* strain harboring an A2063G mutation. Subsequently, the patient's clinical condition improved after substituting azithromycin with minocycline. The boy's muscular strength gradually regained its normal function. Administration of minocycline via the oral route was continued for a period of 3 months, with the dosage gradually tapered off until cessation. Both ultrasonography and enhanced magnetic resonance imaging confirmed the complete resolution of the mass.

Genetic analysis revealed a hemizygous c.1566+1G > C (IVS15+1G > C) mutation in *BTK* gene of the patient. His mother and elderly sister were carriers, whereas the other sister did not carry this mutation.

Heparinized or ethylene diamine tetraacetie acid-treated peripheral blood was obtained by venipuncture from the patient and healthy controls. Informed consent was obtained before analysis. The study protocol was approved by the Ethics Committee of Shanghai Children's Medical Center, Shanghai Jiao Tong University School of Medicine (SCMCIRB-W2022004). We analyzed the effector memory status of $CD4^+$ and $CD8^+$ T cells, including naive (TN, $CCR7^+CD45RA^+$), central memory (TCM, $CCR7^+CD45RA^-$), effector memory (TEM, $CCR7^-CD45RA^-$) and terminally differentiated effector memory $CD45RA^+$ (TEMRA, $CCR7^-CD45RA^+$) cells in the patient and 6 age-matched healthy controls (data were shown as mean of the 6 samples), as shown in Figure 2. The percentage of naive T cells was significantly higher in the XLA patients than in controls both in $CD4^+$ (67.00% vs. 43.6%) and $CD8^+$ T cells (47.40% vs. 34.13%). The percentages of central memory and effector memory T cells were lower in the XLA patients than in controls, both in $CD4^+$ (25.00% vs. 34.28%, 7.62% vs. 19.42%) and $CD8^+$ T cells (7.87% vs. 12.85%, 14.60% vs. 36.51%). The percentage of TEMRA cells was higher in the XLA patients than controls in $CD8^+$ T cells (30.10% vs. 16.51%). These results indicate that both $CD4^+$ and $CD8^+$ T cells in patients with XLA were relatively naive.

We assessed the proliferative capacities of $CD4^+$ and $CD8^+$ T cells. On day 3, proliferative capacity was determined by the decrease in the fluorescence of carboxyfluorescein diacetate succinimidyl ester (CFSE)-labeled $CD4^+$ T cells and $CD8^+$ T cells. As is shown in Figure, Supplemental Digital Content 2, http://links.lww.com/INF/F752, the percentage of $CFSE^-$ proliferating $CD4^+$ T cells was lower than that in healthy controls (data were shown as mean of the 6 samples), both with phytohemagglutinin (PHA) (61.90% vs. 87.50%) and CD3 mAb (36.60% vs. 60.47%) stimulation. While for $CD8^+$T cells, the percentage of $CFSE^-$ proliferating cells was also lower than that in healthy controls after PHA (79.00% vs. 90.33%) and CD3 mAb (59.30% vs. 78.08%) stimulation. These data demonstrated a significant proliferative defect in both $CD4^+$ and $CD8^+$ T cells in patients with XLA.

We further assessed the cell cycle of the $CD4^+$ and $CD8^+$ T cells. On day 3, the cell cycle in both XLA patients and healthy controls was determined by 5-ethynyl-2'-deoxyuridine and propidium iodide staining, both with PHA and CD3 mAb simulation. As shown in Figure, Supplemental Digital Content 3, http://links.lww.com/INF/F753, in $CD4^+$ T cells, the DNA content in G1 phase was significantly higher (PHA: 72.70% vs. 65.52%; CD3 mAb: 86.8% vs. 66.28%) and that in S phase was lower (CD3 mAb: 10.70% vs. 29.07%). While for $CD8^+$T cells, the DNA content of G1 phase was slightly higher (CD3 mAb: 69.40% vs. 56.52%) and that of S phase was lower (CD3 mAb: 25% vs. 35.45%) also. These data

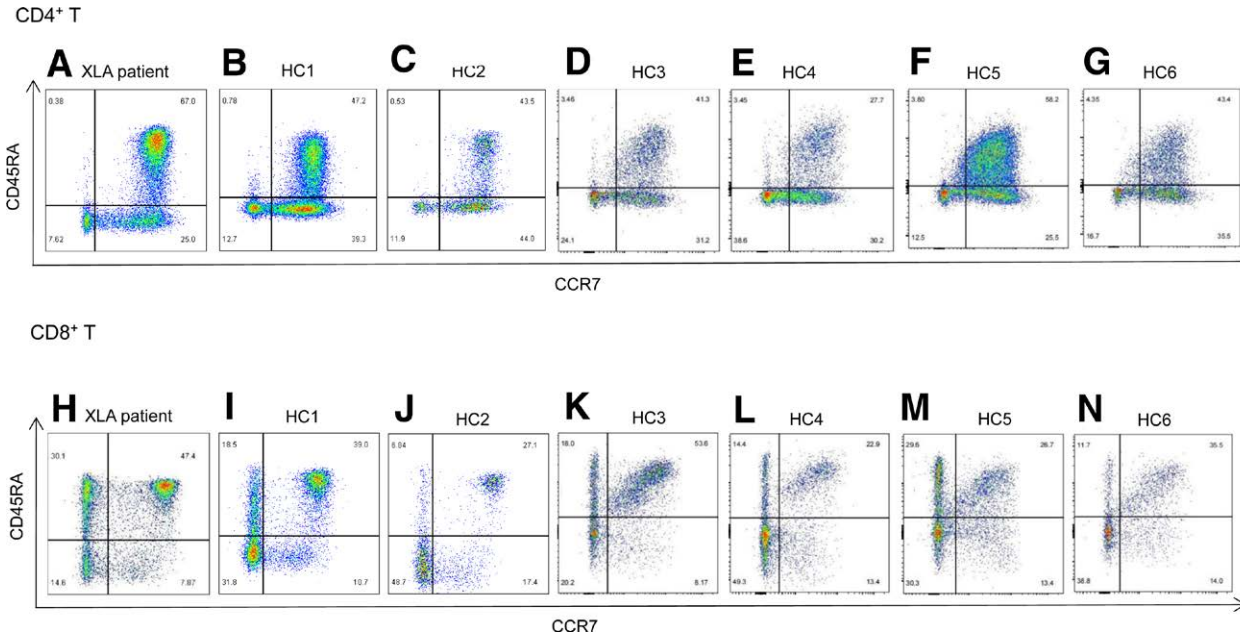

FIGURE 2. The effector memory status of peripheral blood T lymphocytes of the patient. Plots are naive (TN, CCR7$^+$CD45RA$^+$), central memory (TCM, CCR7$^+$CD45RA$^-$), effector memory (TEM, CCR7$^-$CD45RA$^-$) and terminally differentiated effector memory CD45RA$^+$ (TEMRA, CCR7$^-$CD45RA$^+$) cells of the patient (A/H), and 6 age-matched healthy controls (B/C/D/E/F/G/I/J/K/L/M/N). The cells were gated on CD4$^+$ (B/C/D/E/F/G) and CD8$^+$ populations (I/J/K/L/M/N). Numbers in the plots indicate the percentage of cells in each quadrant. HC indicates healthy control.

demonstrate that the T cell cycle of patients with XLA had a G1-S block.

DISCUSSION

In this article, we describe a patient with XLA with a *BTK* gene mutation (IVS15 + 1G > C) who developed an intraspinal cold abscess caused by macrolide-resistant *M. pneumoniae* even when administered an appropriate immunoglobulin replacement therapy and showed abnormal T cell function with naive effector memory status and proliferative capacity defect.

Since the expression of BTK protein in T lymphocytes is very low, the number of peripheral T cells and T cell subsets seemed to be unchanged in patients with XLA; therefore, studies showing T lymphocyte counts and function in these patients are rare. Previous studies mainly focused on the antibody defect caused by the B cell defect, but several clinical problems cannot be explained only by B cell defects, such as why patients with XLA are susceptible to enterovirus, why XLA patients will suffer from poliomyelitis or have prolonged and chronic poliovirus excretion following vaccination with live poliovirus vaccine and why hematopoietic stem cell transplantation can cure intestinal failure caused by enterovirus infection in XLA.[9] These clinical phenomena suggest that although the number of T cells is likely to be normal in children with XLA, its function may be defective.

Patients with XLA usually present with recurrent bacterial infections, mainly in the respiratory and gastrointestinal tracts. Intraspinal cold abscess is extremely rare, especially that caused by macrolide-resistant *M. pneumoniae*, which was very interesting in our case. Cold abscesses are usually induced by *Mycobacterium tuberculous* infection.[10] Nontuberculosis cold abscesses are very rare and has only been reported sporadically. Hyper IgE syndrome is prone to cold abscess caused by *S. aureus* infection because of its immune function defect. Another type of bacterial cold abscess is melioidosis, an infection caused by *Burkholderia pseudomallei*. An impaired neutrophil function in diabetics and chronic alcoholics appears to be the mechanism underlying the pathogenesis of cold abscess in patients with melioidosis. Cold abscess caused by *M. pneumoniae* has not yet been described to our knowledge and the mechanism needs to be further clarified.

However, why did our patient develop macrolide-resistant *M. pneumoniae* infection? The mechanisms underlying macrolide-resistant *M. pneumoniae* infections in patients with XLA are not well understood. Specific antibodies and T cells may play a major role in the elimination of *M. pneumoniae*. Immunoglobulin replacement therapy reduces the frequency and severity of infections and can lower morbidity and mortality rates. The patient in our study still developed intraspinal cold abscess caused by macrolide-resistant *M. pneumoniae* even when administered an appropriate immunoglobulin replacement therapy and the trough IgG level was normal.

We evaluated the effector memory status, proliferation function and cell cycle of peripheral blood T lymphocytes and found that both CD4$^+$ and CD8$^+$ T cells in patients with XLA were mainly naive, with a low percentage of TCM and TEM cells. The data also showed that the proliferative capacity was defective with a G1-S block of the cell cycle in patients with XLA. There are contrasting data on T-lymphocyte function in patients with XLA. On one hand, Plebani et al[11] showed normal T cell proliferation in vitro in response to either mitogens or tetanus toxoid in XLA patients up to 6 months after a tetanus toxoid booster immunization. Moreover, a good in vitro T-cell response to hepatitis B virus was demonstrated in 9 XLA patients up to 24 months after vaccination. On the other hand, an impaired maintenance of T cell memory to Neisseria meningitidis was shown in patients with XLA. Furthermore, Martini et al[12] found that the CD4$^+$ T cell memory compartment was reduced in patients with XLA which is consistent with our findings. It is

also reported that $CD4^+$TEM cells form the major population of the pathogen-specific interferon-γ response in children with mycoplasma pneumoniae infection.[13] This may be one of the causes of *M. pneumoniae* infection in our patient but needs to be further verified by large sample data.

Why is T-cell function abnormal in XLA? It has been suggested that B lymphocytes are not only involved in humoral immune response but also play a critical role in influencing T-cell immunity, so that the lack of B lymphocytes affects some subset of T lymphocytes. However, the exact mechanism of T cell defects in XLA still needs to be further clarified in future studies.

CONCLUSION

Here, we describe a patient with XLA with a *BTK* gene mutation who developed an intraspinal cold abscess caused by macrolide-resistant *M. pneumoniae* even when administered an appropriate immunoglobulin replacement therapy. The patient showed abnormal T cell function with naive effector memory status and proliferative defects.

ACKNOWLEDGMENTS

We are grateful to the patient and his family member for their cooperation.

REFERENCES

1. Cardenas-Morales M, Hernandez-Trujillo VP. Agammaglobulinemia: from X-linked to autosomal forms of disease. *Clin Rev Allergy Immunol*. 2022;63:22–35.
2. Corneth OBJ, Klein Wolterink RGJ, Hendriks RW. BTK signaling in B cell differentiation and autoimmunity. *Curr Top Microbiol Immunol*. 2016;393:67–105.
3. O'Toole D, Groth D, Wright H, et al. X-linked agammaglobulinemia: infection frequency and infection-related mortality in the USIDNET registry. *J Clin Immunol*. 2022;42:827–836.
4. Conley ME, Rohrer J, Minegishi Y. X-linked agammaglobulinemia. *Clin Rev Allergy Immunol*. 2000;19:183–204.
5. Paccoud O, Mahlaoui N, Moshous D, et al; CEREDIH network. Current spectrum of infections in patients with X-linked agammaglobulinemia. *J Clin Immunol*. 2021;41:1266–1271.
6. Kakuya F, Kinebuchi T, Fujiyasu H, et al. Genetic point-of-care diagnosis of mycoplasma pneumoniae infection using LAMP assay. *Pediatr Int*. 2014;56:547–552.
7. Zhao N, Cao J, Xu J, et al. Targeting RNA with next- and third-generation sequencing improves pathogen identification in clinical samples. *Adv Sci (Weinh)*. 2021;8:e2102593.
8. Guo D, Hu W, Xu B, et al. Allele-specific real-time PCR testing for minor macrolide-resistant mycoplasma pneumoniae. *BMC Infect Dis*. 2019;19:616.
9. Shillitoe BMJ, Ponsford M, Slatter MA, et al. Haematopoietic stem cell transplant for norovirus-induced intestinal failure in X-linked agammaglobulinemia. *J Clin Immunol*. 2021;41:1574–1581.
10. Ben Saad S, Kallel N, Gharsalli H, et al. Cold abscess in the immunocompetent subject. *Tunis Med*. 2018;96:302–306.
11. Plebani A, Fischer MB, Meini A, et al. T cell activity and cytokine production in X-linked agammaglobulinemia: implications for vaccination strategies. *Int Arch Allergy Immunol*. 1997;114:90–93.
12. Martini H, Enright V, Perro M, et al. Importance of B cell co-stimulation in CD4(+) T cell differentiation: X-linked agammaglobulinaemia, a human model. *Clin Exp Immunol*. 2011;164:381–387.
13. Pánisová E, Unger WWJ, Berger C, et al. Mycoplasma pneumoniae-specific IFN-gamma-producing CD4(+) effector-memory T cells correlate with pulmonary disease. *Am J Respir Cell Mol Biol*. 2021;64:143–146.