Diagnosis of Hyper IgM syndrome in a Previously Healthy Adolescent Boy Presented with Cutaneous and Cerebral Cryptococcosis

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Abstract: X-linked hyper IgM (X-HIGM) syndrome is a combined immunodeficiency disease caused by mutations in the *CD40LG* gene, leading to a defect in immunoglobulin (Ig) class switching recombination and effector T-cell responses. X-HIGM patients usually present in early life with pyogenic bacterial and opportunistic infections. Herein, we report a previously healthy 13-year-old Thai boy who first presented with cutaneous and meningoencephalitis cryptococcosis. Whole-exome sequencing revealed that he was hemizygous for a missense c.514T>C (p.Tyr172His) in *CD40LG*, confirming a diagnosis of X-HIGM. This report demonstrates that X-HIGM could have an age of onset in teens and systemic cryptococcosis could be its presenting symptoms.

Key Words: X-linked hyper IgM syndrome, cryptococcosis, late-onset primary immunodeficiency, *CD40LG*

(*Pediatr Infect Dis J* 2020;XX:00–00)

X-linked hyper IgM syndrome (X-HIGM, MIM 300386) is an immunodeficiency disorder caused by genetic mutations in the *CD40LG* gene (Cluster of differentiation 40 Ligands gene). The encoded protein "CD40Ligand" (CD40L; CD154) was expressed on activated CD4+ T-cells and binds its receptor CD40 on B cells. The mutation on this gene leads to a defect in immunoglobulin (Ig) class switching recombination resulted in a marked elevation of serum IgM with low or absent IgG, IgA and IgE.[1] CD40L also binds CD40 on macrophages and dendritic cells, leading to the priming of T cells and then resulting in an effective T-cell response.[2] Consequently, the mutations in CD40LG can result in a combined immunodeficiency clinical phenotype. They usually first present in their early childhood with recurrent sinopulmonary tract infections and gastrointestinal manifestations including chronic diarrhea, hepatosplenomegaly, liver diseases and sclerosing cholangitis. They are susceptible to various microbes such as bacteria, fungi and opportunistic organisms including *Pneumocystis jirovecii* and *Cryptosporidium* spp.[2]

Here, we report an unusual case of X-HIGM who manifested with the adolescent onset of disseminated cryptococcosis.

CASE REPORT

A 13-year-old Thai boy presented with progressive jaundice for 1 month and an alteration of consciousness for 1 week. He lived with his parents in a houseboat in Chao Phraya River at Phra Nakhon Si Ayutthaya province, Thailand. He was previously healthy and had never been hospitalized from any illnesses. His immunization was up to date with no history of complications from vaccines. He was born at term to nonconsanguineous Thai parents (Fig. 1A). There was no history of immunodeficiency or infantile death in his family members. He had normal growth parameters with weight and height at 75–95 percentile. Physical examination showed stupor but responded to pain, icteric sclerae, hepatosplenomegaly, molluscum contangiosum-liked lesions on his face, trunk and back (Fig. 1B) with no focal neurologic signs. Investigation revealed eosinophilia 15,100/μL (normal range < 450/μL), high white blood cell count (WBC) 24,890/μL, with normal hematocrit and platelet count. The liver function test showed transaminases with cholestasis jaundice: aspartate aminotransferase 54 U/L (normal range: 15–40), alanine aminotransferase 61 U/L (normal range: 10–55), alkaline phosphatase 1362 U/L (normal range: 100–390), albumin 27 g/L (normal range: 36–52), globulin 24 g/L (normal range: 20–35), total bilirubin 7 mg/dL (normal range: <1.5) and direct bilirubin 6 mg/dL (normal range: <0.2). Cerebrospinal fluid (CSF) examination showed WBC 5900/μL (normal range: 0–7). The cryptococcal antigen was detected in his CSF and the blood with the titer of over 1:1024. *Cryptococcus neoforman* was detected in his blood culture within 5 days.

Histopathology of a skin biopsy showed histiocyte infiltration throughout the whole dermal layer, and many small and large extracellular yeast cells scattering in the infiltrate transepidermal elimination of the inflammatory cells (Fig. 1C). Histopathology of a liver biopsy showed cryptococcosis with granulomatous inflammation and cholestasis (Fig. 1D). Serum anti-HIV was negative. The immunologic evaluation showed a normal number of peripheral blood lymphocyte phenotypes for age with very low IgG and IgA but high IgM (Table 1 and Figure, Supplemental Digital Content 1, http://links.lww.com/INF/E162). Serum IgG to Tetanus and Rubella was negative. The assessment for T-cell function by mitogen stimulation lymphoproliferation showed the normal result. His immunologic abnormalities were compatible with hyper IgM syndrome.

The diagnosis of X-HIGM was confirmed by whole-exome sequencing which found a hemizygous missense c.514T>C variant (GRCh37/hg19 coordinate chrX:135741302 T/C, p.Tyr172His) in the *CD40LG* gene. The p.Tyr172His is located in the TNF homology domain (including amino acids 123–261) as reported by Karpusas et al.[4] The variant was identified in his mother and his older

Accepted for publication September 22, 2020.
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This study was supported by Health System Research Institute and Thailand Research Fund (DPG618000).
The authors have no funding or conflicts of interest to disclose.
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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)
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ISSN: 0891-3668/20/XXXX-0000
DOI: 10.1097/INF.0000000000002945

TABLE 1. Immunological Profiles of the Patient

Lymphocyte Subset	% (Normal Range*)	Cells/μL (Normal Range*)
$CD3^+$T cell	62.3 (53–46)	2473 (430–1500)
$CD3^+CD4^+$T cells	26.78 (31–52)	1052 (530–1300)
$CD3^+CD8^+$ T cell	24.12 (18–35)	948 (330–920)
$CD3^+CD4^+CD45RA^+$	25.16 (33–66)	622 (230–770)
$CD3^+CD8^+CD45RA^+$	16.97 (69–97)	420 (380–1100)
$CD19^+CD20^+$	0.67 (6–23)	2 (110–570)
Immunoglobulin Level	mg/dL (Normal range*)	
IgG	81.5 (698–1194)	
IgA	8.1 (22–274)	
IgM	590 (59–99)	

*Normal range value was obtained from Stiehm's Immune deficiencies textbook.[3]

sister but absent in his father (Fig. 1A). The mutation has previously been reported in Bajorath et al[5] and in the ClinVar database (accession ID is VCV000855047.1), but it is absent in gnomAD and our in-house Thai Exome (N = 2163) databases. In addition, it is predicted to be pathogenic by 10 prediction programs (https://varsome.com/variant/hg19/chrX-135741302-T-C) including BayesDel_addAF, DANN, DEOGEN2, FATHMM-MKL, M-CAP, MVP, MutationAssessor, MutationTaster, REVEL and SIFT.

He received Amphotericin B combined with fluconazole for cryptococcal infection for 6 weeks, and then was switched to oral fluconazole for 8 weeks. His peripheral blood eosinophil count and liver function test turned to be within the normal range after treatment. After the consolidation phase, he received fluconazole for fungal prophylaxis. Intravenous immunoglobulin 500 mg/kg/dose was given to him every 3 weeks with a daily sulfamethoxazole-trimethoprim. Since then, he had no infection requiring hospitalization. Allogenic hematopoietic stem cell transplantation (HSCT), the only curative treatment for X-HIGM, was discussed with parents. However, neither matched sibling nor unrelated donors were available. Subsequently, the child underwent a haploidentical HSCT with his mother as the donor at the age of 14 years and 6 months. After the HSCT, he had complications from graft-versus-host diseases stage IV including chronic diarrhea, upper gastrointestinal bleeding, acute kidney injury, bronchiolitis obliterans, air leak syndrome and infectious associated hemophagocytosis syndrome, and passed away at the age of 15 years.

DISCUSSION

Patients with X-linked hyper IgM (X-HIGM) frequently present with recurrent sinopulmonary tract infections or chronic diarrhea leading to the investigation for the diagnosis before four years of age.[6] Apart from the pyogenic bacterial infection, more than half of patients with X-HIGM presented with opportunistic infection from *P. jirovecii*.[6] Cryptococcocosis is not a common opportunistic infection in patients with X-HIGM. In a case series of infections in 79 X-HIGM patients, only 1 patient had cryptococcosis.[6] Up to now, only 11 patients with X-HIGM were reported to have cryptococcosis as their opportunistic infection. Six patients had a cryptococcal infection in the central nervous system, 5 patients had lymphonodular cryptococcosis, 3 patients had cutaneous cryptococcosis and 2 had evidence of cryptococcal infection in the liver.[7–16]

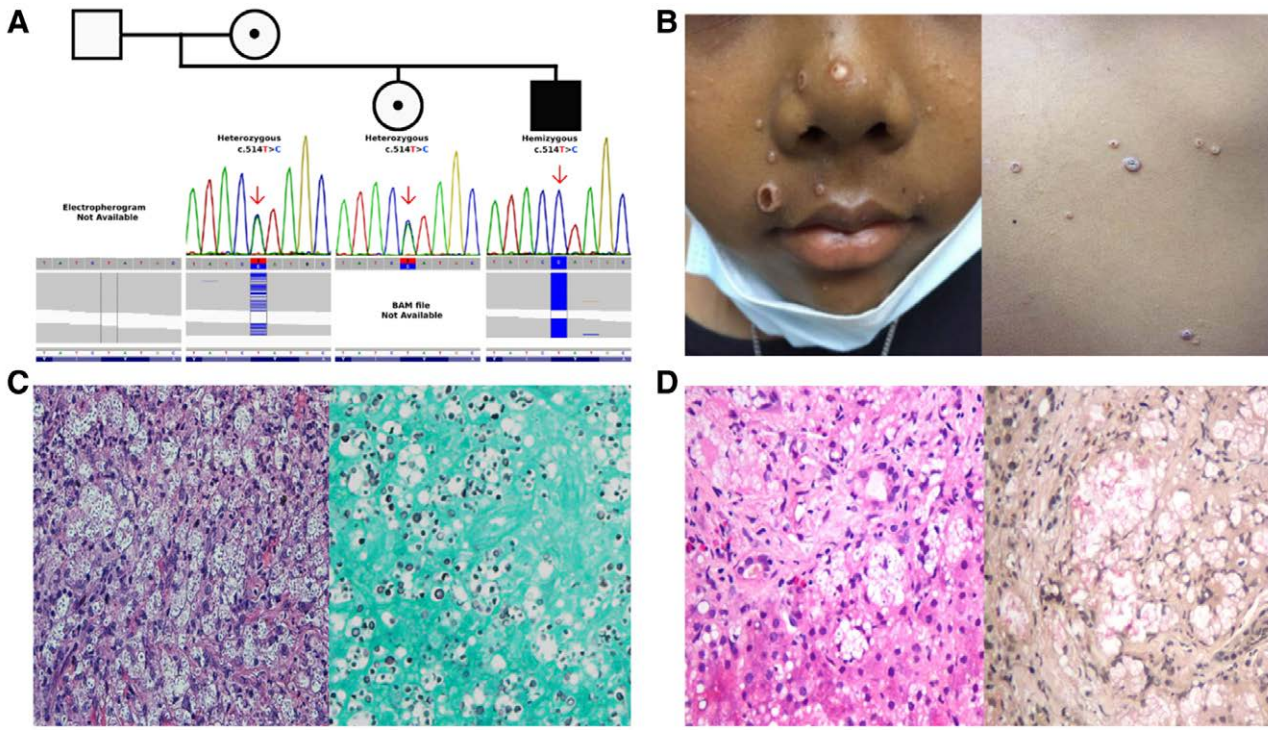

FIGURE 1. Clinical and pathologic features of the CD40L- deficient patient. A: Pedigree (upper panel), Sanger sequencing validation of the identified c.514T>C (p.Y172H) variant in the CD40LG gene (middle panel), and the BAM files showing read depths of exome sequencing of the family members. B: Cutaneous lesions on the face and trunk of the patient. C: Skin pathology with the hematoxylin-eosin stain on the left panel shows abundant encapsulated yeasts which are characteristic features of *Cryptococcus* spp. and the Gomori methenamine silver stain on the right panel confirming the presence of large yeast cells within the infiltration. D: Liver pathology with the hematoxylin–eosin stain on the left panel and the Mucicarmine stain on the right panel showing large yeast cells within the infiltration.

Eight patients experienced cryptococcosis even receiving IVIG replacement therapy and daily sulfamethoxazole-trimethoprim.[7–13] From this observation, the standard treatment with immunoglobulin replacement therapy may not prevent cryptococcal infection in X-HIGM. Time to experiencing cryptococcosis X-HIGM ranged from 2 to 26 years after the diagnosis.[7–13] Only 3 patients initially presented with cryptococcosis at the age of 3 years leading to the diagnosis of X-HIGM.[14–16] Notably, our X-HIGM patient had been healthy up to the age of 13 years old when he developed an unusual invasive fungal infection, disseminated cryptococcosis at the skin, central nervous system, liver and blood.

X-HIGM is the disease caused by the mutations in the CD40 Ligand gene (*CD40LG*) resulting CD40-ligand (CD40L, CD154) deficiency on activated T cells. CD40L binds its receptor CD40 on B cells leading to immunoglobulin isotype switching. CD40L also binds to CD40 on macrophages for their activation.[17] In addition, peripheral blood neutrophils from patients with X-HIGM demonstrated defective respiratory burst and microbicidal activity.[18] These defects on macrophage activation and functional defects of peripheral neutrophils would increase the risk of cryptococcal infections.[17]

Our patient was hemizygous for the c.514T>C (p.Tyr172His) of the *CD40LG* gene. This mutation was previously identified in one patient without demographic data, clinical manifestations, and natural history(unpublished data in Graf et al, 1993).[5] It is located in the extracellular domain of the protein, as seen in the majority of mutations in *CD40LG*.[5] There are some correlations between genotypes and phenotypes in X-HIGM. Large deletions are associated with severe clinical manifestations while missense mutations may have less severity and delayed onset of the disease.[19–21] However, patients with identical mutations may have various presentations and different severity.[22]

Treatment of X-HIGM consists of immunoglobulin replacement therapy and prevention of Pneumocystis infections with trimethoprim-sulfamethoxazole. With conventional treatment, X-HIGM patients may have fatality from viral encephalitis and hepatic malignancy. HSCT is the curative treatment and the best survival outcome is associated with early age at transplant and the absence of liver disease.[2] However, patients undergoing HSCT have a chance to develop graft-versus-host diseases, especially in haploidentical HSCT. Nevertheless, the prognosis of patients with X-HIGM is not good, with only 20% of reported survival cases reached the age of 20 years.[6] The unfavorable result of HSCT in our patients may partly explain from the later age at transplant. As a result, performing HSCT in X-HIGM with advanced age may need to weigh between the risk for complications from HSCT and the chance for developing fatal encephalitis or hepatic malignancy if only conventional treatment is given.

In conclusion, we report a case of X-HIGM in a previously healthy 13-year-old boy first presented with disseminated cryptococcosis. Our case would emphasize the awareness of the late-onset primary immune deficiency in previously healthy adolescents.

ACKNOWLEDGMENTS

to the authors thank Asst. Prof. Thipwimol Timaroon for the result of the Sanger sequencing.

REFERENCES

1. Qamar N, Fuleihan RL. The hyper IgM syndromes. *Clin Rev Allergy Immunol*. 2014;46:120–130.
2. de la Morena MT. Clinical phenotypes of hyper-IgM syndromes. *J Allergy Clin Immunol Pract*. 2016;4:1023–1036.
3. Jyonouchi S. Appendix. In: Sullivan KE, Stiehm ER, eds. *Stiehm's Immune Deficiencies*. 1st ed. London: Academic Press; 2014:1111–1113.
4. Karpusas M, Hsu YM, Wang JH, et al. 2 A crystal structure of an extracellular fragment of human CD40 ligand. *Structure*. 1995;3:1426.
5. Bajorath J, Seyama K, Nonoyama S, et al. Classification of mutations in the human CD40 ligand, gp39, that are associated with X-linked hyper IgM syndrome. *Protein Sci*. 1996;5:531–534.
6. Winkelstein JA, Marino MC, Ochs H, et al. The X-linked hyper-IgM syndrome: clinical and immunologic features of 79 patients. *Medicine (Baltimore)*. 2003;82:373–384.
7. Jo EK, Kim HS, Lee MY, et al. X-linked hyper-IgM syndrome associated with *Cryptosporidium parvum* and *Cryptococcus neoformans* infections: the first case with molecular diagnosis in Korea. *J Korean Med Sci*. 2002;17:116–120.
8. Malheiro L, Lazzara D, Xerinda S, et al. Cryptococcal meningoencephalitis in a patient with hyper immunoglobulin M (IgM) syndrome: a case report. *BMC Res Notes*. 2014;7:566.
9. de Górgolas M, Erice A, Gil A, et al. Cryptococcal meningitis in a patient with X-linked hyper-IgM1 syndrome. *Scand J Infect Dis*. 2005;37:526–528.
10. Lee MY, Chung JH, Shin JH, et al. Lymphonodular cryptococcosis diagnosed by fine needle aspiration cytology in hyper-IgM syndrome. A case report. *Acta Cytol*. 2001;45:241–244.
11. Tabone MD, Leverger G, Landman J, et al. Disseminated lymphonodular cryptococcosis in a child with X-linked hyper-IgM immunodeficiency. *Pediatr Infect Dis J*. 1994;13:77–79.
12. Pacharn P, Phongsamart W, Boonyawat B, et al. Disseminated cryptococcosis in two boys with novel mutation of CD40 ligand-associated X-linked hyper-IgM syndrome. *Asian Pac J Allergy Immunol*. 2018. doi: 10.12932/AP-140218-0265. [Online ahead of print]
13. Acker KP, Fetch A, Schnell SA, et al. Scalp lesions in a pediatric patient with hyper IgM Syndrome: clinical and histologic mimicry of *Cryptococcus neoformans* infection. *J Pediatr*. 2018;192:256–258.
14. Kyong CU, Virella G, Fudenberg HH, et al. X-linked immunodeficiency with increased IgM: clinical, ethnic, and immunologic heterogeneity. *Pediatr Res*. 1978;12:1024–1026.
15. Iseki M, Anzo M, Yamashita N, et al. Hyper-IgM immunodeficiency with disseminated cryptococcosis. *Acta Paediatr*. 1994;83:780–782.
16. Mohanty SK, Thakral D, Gupta D, et al. Diminished CD40L expression on T-cells in a case of disseminated cryptococcosis. *Indian J Pathol Microbiol*. 2018;61:137–140.
17. Pietrella D, Lupo P, Perito S, et al. Disruption of CD40/CD40L interaction influences the course of *Cryptococcus neoformans* infection. *FEMS Immunol Med Microbiol*. 2004;40:63–70.
18. Cabral-Marques O, França TT, Al-Sbiei A, et al. CD40 ligand deficiency causes functional defects of peripheral neutrophils that are improved by exogenous IFN-γ. *J Allergy Clin Immunol*. 2018;142:1571–1588.e9.
19. López-Granados E, Cambronero R, Ferreira A, et al. Three novel mutations reflect the variety of defects causing phenotypically diverse X-linked hyper-IgM syndrome. *Clin Exp Immunol*. 2003;133:123–131.
20. Seyama K, Nonoyama S, Gangsaas I, et al. Mutations of the CD40 ligand gene and its effect on CD40 ligand expression in patients with X-linked hyper IgM syndrome. *Blood*. 1998;92:2421–2434.
21. Tsai HY, Yu HH, Chien YH, et al. X-linked hyper-IgM syndrome with CD40LG mutation: two case reports and literature review in Taiwanese patients. *J Microbiol Immunol Infect*. 2015;48:113–118.
22. Alt FW. *Hyper-IgM Syndrome*. 1st ed. Cambridge, MA: Academic Press; 2004.