Cutaneous Mycobacterium shigaense Infection in Immunocompetent Woman, China

To the Editor: Mycobacterium shigaense is a novel, slow-growing, scotochromogenic mycobacterium (1), initially reported in 2012 as an opportunistic pathogen isolated from skin biopsy samples from a patient with a history of Hodgkin disease and severe cellular immunodeficiency. We describe the identification of this species in a chronic cutaneous infection in an immunocompetent woman.

A 56-year-old woman was admitted to our inpatient department in August 2011 with reddish papules, nodules, plaques, and scars on her face and neck (Figure, panel A) that had developed over >1 year, starting in June 2010. Initially, a few small papules appeared on her face; the primary papules gradually enlarged, spreading to the neck and developing into nodules, plaque, partly purulent lesions, and sometimes fistulas associated with moderate pain and scarring. The patient reported no history of trauma or surgical procedure and could not recall any potential inducement of the lesions or previous receipt of immunosuppressant therapy.

On physical examination, the patient was thin, with normal vital signs. Results of routine laboratory tests were unremarkable. Cephalic and cervical radiographs and computed tomography scans revealed lymphadenectasis of the neck only; a purified protein derivative test showed an erythema ≈8 mm in diameter on the forearm. Results of testing for HIV and human T-lymphotropic virus 1 antibody detection tests were negative. Cell-mediated immunity levels were detected by flow cytometry of peripheral blood cells; cell counts for CD3+, CD4+, CD8+ T-cells, and T-cell receptors and for CD19+ B-cells were within reference ranges.

Skin samples were collected from the face and neck of the patient and subjected to histopathologic examination, smear testing, and culture. Histopathologic examination showed a hyperplastic epidermis and nongranulomatous infiltrates of lymphocytes, histiocytes, and multinucleate giant cells in the dermis (Figure, panel B). Results of Ziehl-Neelsen staining of smears for acid-fast bacilli and periodic acid–Schiff staining for fungi were negative; fungal and other bacterial cultures were sterile. Samples streaked on Löwenstein-Jensen medium at 32°C and 37°C for 4 weeks formed smooth, creamy, yolk yellow colonies (Figure, panel C); however, such colonies did not grow at 25°C and 45°C. Ziehl-Neelsen staining of samples from the colonies revealed acid-fast bacilli (Figure, panel D). Infection with a mycobacterium was suspected on the basis of these results.

The isolated bacilli were subjected to PCR restriction fragment-length polymorphism analysis and sequencing of the mycobacterial hsp65 gene to identify the bacteria and strain (http://app.chuv.ch/prasite/index.html) (2,3). However, a match for the restriction pattern or the sequence of the isolated organism and Mycobacterium spp. was not found. Therefore, complete DNA sequences of the 16S rRNA, hsp65, and rpoB genes and the 16S–23S rRNA ITS region were determined by using primers and PCR protocols described previously (4–7). The sequences of 16S rRNA, rpoB and the 16S–23S rRNA ITS region were identical to those of the nontuberculous mycobacterium species M. shigaense; the hsp65 gene showed 94% similarity (online Technical Appendix, wwwnc.cdc.gov/EID/article/19/5/12-1022-Techapp1.pdf).

Results of biochemical tests of the isolate were positive for Mycobacterium spp. by a 2-week culture on MacConkey agar and a heat catalase test but

Figure. Cutaneous Mycobacterium shigaense infection in a 56-year-old Immunocompetent woman, China. A) Plaques, scars with scabbing, nodules, and concave scars on the face and neck. B) Histopathologic results, showing hyperplastic epidermis and infiltration with lymphocytes, neutrophilic leukocytes, multinucleate giant cells, and epithelioid cells in the dermis. C) Samples streaked on Löwenstein–Jensen medium at 32°C formed smooth, yolk yellow creamy colonies. D) Ziehl–Neelsen staining of bacilli from the colonies that tested positive. E) Visible improvement of lesions after 4 months of treatment.
weakly positive by 3-day arylsulfatase and 2-week catalase testing (8). Results of nitrate reduction, semiquantitative catalase, growth in 5% NaCl medium, urease, and Tween 80 hydrolysis testing were negative. In vitro drug susceptibility was investigated by using the microdilution method, according to Clinical and Laboratory Standards Institute guidelines (9). The isolate was susceptible to moxifloxacin, amikacin, clarithromycin, rifampin, ethambutol, streptomycin, and ofloxacin.

The patient was treated with orally administered rifampin (450 mg 1×/d), moxifloxacin (400 mg 1×/d), and clarithromycin (500 mg 2×/d) for 6 months. The lesions subsided, leaving hyperplastic and atrophic scars (Figure, panel E). New nodules did not recur, and no notable side effects were found.

In general, culture-based identification methods using biochemical tests are slow and inadequate in differentiating species of mycobacteria. Laboratory methods with better performance, such as genetic investigations using nucleic acid amplification and sequencing, are increasingly used for identification. In this case, the complete gene sequences of 16S rRNA, 16S–23S rRNA ITS region, rpoB, and hsp65 of the isolate were used to find the most consistent and highest scoring match across all 4 loci in GenBank (www.ncbi.nlm.nih.gov/genbank).

In conclusion, our results strongly suggest that this chronic cutaneous infection in an immunocompetent patient was caused by M. shigaense. Our observations provide further evidence that this species should be classified as a nontuberculous mycobacterium that can cause disease in immunocompromised and immunocompetent patients. The isolate we identified was classified as clinically pathogenic and have the print issue delivered

Acknowledgments

We thank all the medical workers in ward of our hospital for co-operation and clinical assistance and the colleagues in our mycobacteria laboratory for technical guidance.

This study was supported by grants from the National Natural Science Foundation of China (30972651) and the fund for Key Clinical Program of the Ministry of Health (2010-2012-125).

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DOI: http://dx.doi.org/10.3201/eid1905.121022

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Technical Appendix

DNA sequences for *Mycobacterium shigaense* genes

All sequences are shown in a 5’→3’ orientation.

16S rRNA (*rrs*) (1,346 bp)

CGTGGGTAATCTGACCTGCACTTCGAGATAAGCTGGGTAATATACCGG
ATAGGACCATTTAGCGATGCTTTGTTGTTGGAAGCTTTTGCAGTGTTGGGATGAGCC
CGCGGCTATAGCGTCTTGTTGTGGTGAGGACCGGCTACCAAGGGCGACAGACGGGTAGCGC
CGGCCCTGTGAGAGGGTGCCGGACACTGGGACTGAGATACGGCCCAGACTCCTACCG
GGAGGACGGTGTGGGAATATTGACAATGGGCGACAGCTGGATGCAGCGACGCCG
CGTGCCGAGTGGGACCGCCCTCCGACAGGTAAACCTCTCTACGCAGGGGGAAGGCTG
AGATAGGGTGCGTGCGTTGCACTGGGCTAAAGAGCTCGTAGGTGGTTTGCAGTTGACGG
GTCGCGCTTGTTGCAGAAGTGGAATTACCTGGGCTAAAGAGCTCGTAGGTGGTTTGC
AGACTGAGTACTGCAAGGGAGACTGGAATTACCTGGTGTAGCGGTGGAATGCGCACG
ATATCAGGAGGAACACCGGTGGCAGGGCAGGCTCTTGGGCAGTAACTGACGGTGA
GGAGCAAGGACAGCGTGGGAGGAGAACAGGATTAGATTCCCTCCGGGCGGCTACTACGC
ACGTTGAGTTACTAGGTGTGGGTTTCTCTCTTCCTGGGAATCCGTGCGCGTAGCTAAGC
GATCAGCGGCTCTGCGGGAGTACGGCCGCAAGGCTAAAACTCAAAGGAATTGGGCG
GGGCCGCAACAGCGGCGAGCATGTGGGATTTAATTCCGGTAGCAGTGGCAGAAGCTT
ACCTGGGTTCGACATGCAGGACGCCGCGTGCATGCGTTCCCTTGTGGCCCTGT
GTGCAGGTGGTGCATGGCTGTCGTCAGCTCGTGTCGTGAGATGTTGGGTTAAGTCCC
GCAACGAGCAGCAACCCCTTGGTCTCATGTTGCCAGCAGCTAATGGCGGGGACTCGTGA
GAGACTGCCCGGGGCTCAACTCGGAGGAAGGTTGGGATGAGCTCAAGTCATCAGTCC
CTTATGTCAGGGCTTTCACACATGCTACAATGGGCCGCTACAAGTCGCTGGCAAATCGC
CAAGGTTAAGCGAAATTTTTAAGGCCGCTCTCAAGTTCGAGATCGGGGTCTGCAACTC
GACCCCGTGAAGTGCGGACTGCTAGTAATCGCAGATCGCAACGCTCGGGGTCTGGAATA
CGTTCCCCGGCCTTGTACACACACCGGGGCTCAAGTCATGAAATGCTGGTAACACCCGAA
GCCAGTGCCCTAACCTTTTGGAGGGAGCTGTAAGGTGGGATCGGCGA
GGAGGGCCCTTTCGGGGGTACT

16S–23S rRNA internal transcribed spacer region (197 bp)

TCTCCAATTGGTGTTGGGTGTGAGCCGTGAGGGGTTCTCGTCTGTAGTGAGGACGGG
GGGTCGACCAACAGCAGGGCAATGCCAGACACACTATAAGGGCCCTGAGGAAGGCAACT
CAGTTTCTTCCGAGATGCGTCTCCATCTCATTTGGGTGAGGGGTGGGTGGTGGTGGATAGT
GATAGTGTTGCGAGCATCTAAATGA

rpoB (3,362 bp)

GCACCTAACCGAGTTTTCTTTTGCAAGTCGCTCAGACCGCTTGAGTTCCCGGGGCTT
CTCGATGTGCAGCCGACTCGTTTTGAGTGGCTGATCGGCTCGCCCCCGCTGGCAGG
ACGCAGCGCCACCCGGCGAGTCAGCGCCCGGCTGAGGCGTGGCCCTGAGGAGGTGCTCA
CGAGCTGTCCGGCATTGAGGACTTCTCTCCCCGCTGCTCGCTGCTCTCCGACC
GCCTTTCGACGAGGTAAGCCGGCGGTGCAAGGAGTGCAAGAAGAAGGACATGCGT
ACGAGAGCCCGCTGTACTGCTGATCGCCAGCTGACAGAAGAAGGACATCGT
AAAAGCCAGACGGTTGCTCAGGGGGACCTCTCCCGATGAGTGAGGAGGACAAAGGACAC
CATCATCAACGGGACCGAGGCAGGCTGCTCAGCCAGCTGCTCGGCTGGGCCGCTG
TGTACCTCAGGACTCATGCAAACTGCTGACAGCAGGAGAAGGACATCGT
GTGACCCGAGCGGGGCGCTGCTGAGGCGTGGCTGACAGTGGTGAGGCGTGGGCTGGCGC
CGGCTGCGCATCGACCCGACAGGAGCCGGCCAGCCGGACCGGTCACCGGTCTGCTGCTCAAGGCG
TGGGCTGGGACACGAGAAGTACACCGAAGCGGTTCCGGCTTTCTCCGAGATCATGATCT
GCGCCGAGGAGATCACGCGGGACATCCCGAACGTCTCCGACGAGGTGCTCGCCGAC
CTGGGCGCGGACGCGGCATCGCCTCGCTATCGGTGTCGCCAGTCACGCGGTCTCCGC
GCCGACCGGTGAGTCCCGGCAAGGGGTGACTCGTAGTATCCGGGGTTCTCTCCCGCA
ACGACGAATCTGCCCGGCACAGGCTGCTGCCGACACGTCTCGACATCGTGAAGGT
CCGCACACGGGTCGAGACGCGCCACGCAAAAGGGGAAAGCCCGACCGTACATCGG
ACGATCGCTAACCACCCGGACGGGAGCGTGAAGCTCAGGACGAGAAGATCGGCGCC
GACAGCGTCATGCCCTTGCCGACGGGTAAAGGTGACGTTGGGGCCCAAGGGTCGCA
CGTCCTGGAGAAGAAGTGGGGCGCTCCCACGATCACCAACGATGGTGTGTCCATCG
CCAAGGAGATCGAGCTGGAGGACCCCTACGAGAAGATCGGCGCCGAGCTGGTC
AAGGAAGTCGCCAAGAAGACCGACGACGTTGCCGGTGACGGCACGACGACGGCCACCGT
GCTCGCTCAGGCACTCGTCAAAGAGGGCCTGCGCAACGTGGCGGCCGGCGCCAACC

**hsp65 (1,526 bp)**

GCTCATGCCCCTTGCGCAGCAGGGTAAAAAGGTGACTGTTGGGGCCCAAGGGGGTCGA
CGTCCTGGAGAAGAAGTGGGGCGCTCCCACGATCACCAACGATGGTGTGTCCATCG
CCAAGGAGATCGAGCTGGAGGACCCCTACGAGAAGATCGGCGCCGAGCTGGTC
AAGGAAGTCGCCAAGAAGACCGACGACGTTGCCGGTGACGGCACGACGACGGCCACCGT
GCTCGCTCAGGCACTCGTCAAAGAAGGGGCTGCAGAAGACGCTCGAGTGCAGGTGCTCGAGCGAC

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