

BRIEF COMMUNICATION

A report on the first outbreak of a single clone group A *Streptococcus* (emm-type 89) tonsillopharyngitis in China



Ying-Mei Liu^{a,c}, Jian-Zhong Zhao^{b,c}, Bin-Bin Li^a,
Jun-Yong Yang^b, Xiao-Gen Dong^b, Jian-Jun Zhang^{b,**},
Bin Cao^{a,*}

^a Department of Infectious Diseases and Clinical Microbiology, Beijing Chao-Yang Hospital, Capital Medical University, Beijing, People's Republic of China

^b Center for Disease Control and Prevention, Fengtai, Beijing, People's Republic of China

Received 30 March 2013; received in revised form 15 July 2013; accepted 8 August 2013
Available online 8 October 2013

KEYWORDS

emm-type 89;
Film crew;
Group A
Streptococcus;
Outbreak;
Tonsillopharyngitis

In 2012, 28 out of 140 staff working for a film crew in a Beijing movie and television base experienced a sudden onset of fever, sore throat, and/or tiredness, headache within the 24 hour period of July 26–27. All of the patients visited the hospital and were diagnosed as having tonsillopharyngitis. On July 28, 2012, a team of Centers for Disease Control and Prevention staff arrived and initiated an outbreak investigation. Pharyngeal swabs were obtained from patients for microbiologic analysis. All isolates of the outbreak were analyzed for toxin-genes and drug-resistance genes by polymerase chain reaction, and were performed for the *emm* typing and pulsed-field gel electrophoresis typing. On July 30, 2012, Group A *Streptococcus* was isolated from eight of the 16 throat swab specimens obtained on site. These isolates were found to have the same genotype *emm* 89. This is the first report to identify Group A *Streptococcus emm* type-89 as a cause of tonsillopharyngitis in Beijing, China.

Copyright © 2013, Taiwan Society of Microbiology. Published by Elsevier Taiwan LLC. All rights reserved.

* Corresponding author. Department of Infectious Diseases and Clinical Microbiology, Beijing Chao-Yang Hospital, Capital Medical University, Number 8 Gongren Tiyuchang South Road, Chaoyang District, Beijing 100020, People's Republic of China.

** Corresponding author. Center for Disease Control and Prevention, Number 3 Xi An Road, Fengtai District, Beijing 100071, People's Republic of China.

E-mail addresses: ft8302@yahoo.com.cn (J.-J. Zhang), caobin1999@gmail.com (B. Cao).

^c These authors contributed equally to this work.

Introduction

Group A streptococcal (GAS) pharyngitis is a common disease of epidemic nature, and responsible for 5–15% of cases of pharyngitis in adults and 20–30% of cases in children. GAS strains are categorized by a variation in the nucleotide sequence of the gene (*emm*) that encodes the M protein. Currently, more than 200 distinct *emm* types of GAS have been recognized. In distinct geographic areas, the predominant *emm* types often vary in frequency from year to year for reasons that are not fully understood. The *emm* type-89 is highly prevalent in some parts of the world.¹ Regional outbreaks of *emm* 89 strains have been documented previously, including a clonal epidemic that occurred in northern Italy,² and a food-borne outbreak of streptococcal pharyngotonsillitis in Denmark.³ The prevalence of the *emm* 89 in China is currently unknown; this strain has not been isolated from GAS carriers or detected from patients with GAS infections. In this study, we report an outbreak of tonsillopharyngitis caused by GAS (*emm*-type 89) among staff of a film crew in Beijing, China.

Methods

The outbreak

On July 8, 2012, a film crew that included 140 staff started shooting a movie in Beijing Fengtai Movie and Television Base of People's Liberation Army (PLA), China. On July 26, 2012, 10 staff presented with fever, sore throat, and/or tiredness, headache. These patients all visited the local hospital in Beijing with the same complaints and were treated symptomatically for a presumed upper respiratory tract infection. By the next day, there were 18 cases with same symptom; the presumptive diagnosis was changed to bacterial tonsillopharyngitis. The treatment was initiated by the hospital's physicians and was not uniform. On July 28, 2012, the Center for Disease Control and Prevention (CDC) of Beijing was notified of unusually widespread tonsillopharyngitis among staff serving in the film crew. A team of CDC staff arrived on the day of notification and initiated an outbreak investigation. Pharyngeal swabs were obtained from 16 (57.1%) of 28 patients and were sent to the laboratory of CDC for microbiologic analysis. Drinking water samples were collected from the rooms where the staff stayed in and were also sent for microbiologic analysis on the same day.

Infection-control strategies implemented

Environmental surfaces were rigorously cleaned every day and kept fresh air in the bedrooms. All GAS-infected patients were placed in single rooms in order to reduce contact with each other.

Identification and antimicrobial susceptibility testing of strains

Pharyngeal swabs were identified using conventional microbiologic methods. Minimal inhibitory concentrations

for isolated GAS were determined by broth microdilution methods, which were performed and interpreted according to the guidelines established by the Clinical and Laboratory Standards Institute.

Assignment of *emm* type

For *emm* typing, lysate preparation, polymerase chain reaction amplification and sequencing were performed according to the protocol described by the CDC. High quality sequences were trimmed and compared with reference sequences in the CDC *emm* database (<http://www.cdc.gov/ncidod/biotech/strep/strepblast.htm>).

Pulsed-field gel electrophoresis

Pulsed-field gel electrophoresis (PFGE) was conducted as previously described with minor modifications.⁴ Whole-cell genomic DNA was digested with the restriction enzyme *Apal*. PFGE patterns were interpreted using the criteria proposed by Tenover et al.⁵

Detection of virulence and resistance genes

All isolates of the outbreak were analyzed for amplification of virulence and drug-resistance genes as described previously.^{6,7}

Results

Epidemiological findings

All of the crew members were staying in two courtyards with several standard bedrooms (2 staff shared a room) with good hygienic conditions but not a good airflow. Take-away food was ordered for the staff from a local restaurant every day. Between the evening of July 26, 2012 and the evening of July 27, 2012, a total of 28 staff experienced a sudden onset of fever, sore throat, and/or tiredness, and headache. Physical examination revealed tonsil enlargement (Table 1). The overall attack rate was 20% (28/140). Full recovery occurred within 3 days.

Results of microbiological testing

On July 30, 2012, GAS was isolated from eight of the 16 throat swabs. We found resistance to erythromycin, azithromycin, clindamycin, and tetracycline among all GAS isolates. All 8 strains tested were still highly susceptible to penicillin, levofloxacin, vancomycin, linezolid, ceftriaxone, and meropenem. These isolates were all found to have the same genotype (*emm* 89), the same PFGE type, the same resistance-gene profiles, and the same toxin-gene profiles (Fig. 1).

Discussion

We present an epidemiologic analysis of an outbreak of streptococcal tonsillopharyngitis in a film crew in China. The causing organism was a single clone GAS (*emm*-type

Table 1 Epidemiological, clinical, and laboratory characteristics of 28 patients in this outbreak

| Case | Onset of fever (2012) | T (°C) | Cough | Snivel | Sore throat | Headache | Tiredness | Tonsil enlargement | WBC/mm ³ | CRP (mg/L) |
|------|-----------------------|--------|-------|--------|-------------|----------|-----------|--------------------|---------------------|------------|
| 1 | Evening July 26 | 39.1 | — | — | + | + | + | + | 19.3 | 10 |
| 2 | Morning July 27 | 38.3 | — | — | + | + | — | + | 9.7 | 8 |
| 3 | Evening July 26 | 38.0 | — | — | + | + | + | + | 12.5 | 48 |
| 4 | Morning July 27 | 38.4 | — | — | + | — | — | + | 10.6 | 24 |
| 5 | Morning July 27 | 39.4 | — | — | + | — | + | — | 17.1 | 11 |
| 6 | Morning July 27 | 38.3 | — | — | + | — | + | + | 14.6 | 26 |
| 7 | Evening July 26 | 37.0 | — | — | + | — | — | — | 6.7 | 1 |
| 8 | Morning July 27 | 39.6 | — | — | + | — | — | + | 15.2 | 86 |
| 9 | Morning July 27 | 38.4 | — | — | + | + | — | + | 8.3 | 11 |
| 10 | Evening July 26 | 38.4 | — | — | + | — | — | — | — | — |
| 11 | Evening July 26 | 38.9 | — | — | + | — | — | — | 13.2 | 1 |
| 12 | Morning July 27 | 37.8 | — | — | + | — | — | + | 12.1 | 6 |
| 13 | Evening July 26 | 40.0 | — | — | + | + | + | + | 14.7 | 88 |
| 14 | Morning July 27 | 38.2 | — | — | + | — | — | + | 11.6 | 19 |
| 15 | Morning July 27 | 38.2 | — | — | + | — | — | + | 11.9 | 17 |
| 16 | Evening July 26 | 38.3 | — | — | + | + | + | + | 9.6 | 26 |
| 17 | Evening July 26 | 38.0 | — | — | + | + | + | + | 10.8 | 40 |
| 18 | Evening July 26 | 39.4 | — | — | + | + | + | + | 16.5 | 11 |
| 19 | Evening July 26 | 38.7 | — | — | + | — | + | + | 13.7 | 52 |
| 20 | Morning July 27 | 38.3 | — | — | + | — | — | + | 13.0 | 15 |
| 21 | Morning July 27 | 38.2 | — | — | + | — | — | + | 12.9 | 28 |
| 22 | Morning July 27 | 39.4 | — | — | + | — | — | + | 12.2 | 93 |
| 23 | Morning July 27 | 38.5 | — | — | + | — | — | + | 11.6 | 1 |
| 24 | Morning July 27 | 37.5 | — | — | + | — | — | + | 11.5 | 17 |
| 25 | Evening July 27 | 37.9 | — | — | + | — | — | + | 15.8 | 96 |
| 26 | Morning July 27 | 37.8 | — | — | + | + | — | + | 13.9 | 83 |
| 27 | Morning July 27 | 36.7 | — | — | + | — | — | — | 5.8 | 1 |
| 28 | Morning July 27 | 38.0 | — | — | + | — | — | + | 16.6 | 133 |

CRP = C-reactive protein; WBC = white blood cell count.

89). This type is highly prevalent in some parts of the world.¹ However, *emm*-type 89 was absent from a sample collected in India⁸ and in a survey during 1997–2008 in Taiwan,⁹ illustrating that the predominant *emm* types vary with geographical location. In mainland China, the most prevalent *emm* genotype identified from clinical specimen in recent years were *emm* 1.0, *emm* 12.0, *emm* 4.0, and *emm* 22.0. Because research on GAS is mainly focused on children in China, to date the information on adults is scarce. The prevalence of the *emm* 89 in China is currently unknown; this strain was isolated from neither GAS carriers nor patients with GAS infections. Our current study is the first report of *emm* 89 in mainland China.

In a previous study, the rate of erythromycin-resistance isolates was high in China (97.6%), and the rate of resistance to tetracycline was also high in China (94%). The high rates of resistance in mainland China may be correlated with the overuse of these antibiotics. Consistent with previous studies,^{10,11} all the isolates in the current study exhibited resistance to erythromycin, azithromycin, clindamycin, and tetracycline. Penicillin is the treatment of choice in GAS infections. However, among patients with β -lactam allergy, macrolides, and lincosamides are first-line alternative therapy. Fortunately, the GAS strains in this outbreak were still highly susceptible to penicillin and quinolone. Quinolone may represent an alternative for

treating infections with macrolid-resistance GAS strain in patients allergic to β -lactams.

In the current study, airborne transmission was the initial suspicion. However, further epidemiological investigations showed that individuals who stayed in the same rooms with index cases did not develop the same illness. In addition, all of the patients presented similar symptoms within 24 hours and no further new cases were identified after this period. The sudden onset of the outbreak was a characteristic of food-borne streptococcal pharyngitis.¹² Most food-borne GAS outbreaks were reported from high-income countries such as Australia, Denmark, and the USA.^{1,3,12} Contamination of food with streptococci could present with unusual outbreaks that may be difficult to recognize in the early stages. In fact, most food-borne GAS outbreaks are ignored by healthy personnel who are not aware of this kind of transmission or they may detect it but do not report it. Thus, the current outbreak is a reminder of a possible route of streptococcal dissemination, and suggests that food-borne GAS outbreaks are probably under-recognized.

In the current study, the proportion of GAS-positive cultures was relatively low (8 out of 16), probably due to the fact that half of the cases had already taken antibiotics before the samples were taken. Moreover, our results suggest that this outbreak in Beijing could be food-borne.

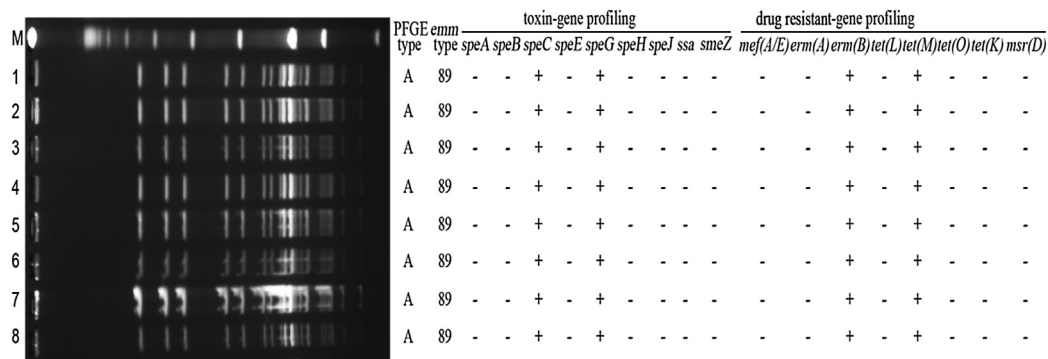


Figure 1. Pulsed-field gel electrophoresis (PFGE) patterns of *Apal*-restricted chromosomal DNA, *emm* typing, drug resistance-gene profile and toxin-gene profile of Group A *Streptococcus emm*-type 89 strains isolated during the outbreak.

However, we were unable to obtain samples of food consumed by the crew members; therefore, it is difficult to draw a firm conclusion on the transmission route.

Conflicts of interest

The authors declare that there are no conflicts of interest.

Acknowledgments

The authors thank Shoushan Qu, Fang Li, Shanshan Wang, Chunxia Yang, Chunlei Wang, Jiuxin QU, Zhengjia Liu, and Peng Wang for their assistance. This work was financially supported by National Natural Science Foundation of China (81070005/H0104) and Program for New Century Excellent Talents in University (NCET-10-0006) for Dr Bin Cao.

References

- Levy M, Johnson CG, Kraa E. Tonsillopharyngitis caused by foodborne group A *Streptococcus*: a prison-based outbreak. *Clin Infect Dis* 2003;**36**:175–82.
- Creti R, Imperi M, Baldassarri L, Pataracchia M, Recchia S, Alfarone G, et al. *emm* types, virulence factors, and antibiotic resistance of invasive *Streptococcus pyogenes* isolates from Italy: what has changed in 11 years? *J Clin Microbiol* 2007;**45**: 2249–56.
- Falkenhorst G, Bagdonaite J, Lisby M, Madsen SB, Lambertsen L, Olsen KE, et al. Outbreak of group A streptococcal throat infection: don't forget to ask about food. *Epidemiol Infect* 2008;**136**:1165–71.
- Chiou CS, Liao TL, Wang TH, Chang HL, Liao JC, Li CC. Epidemiology and molecular characterization of *Streptococcus pyogenes* recovered from scarlet fever patients in central Taiwan from 1996 to 1999. *J Clin Microbiol* 2004;**42**:3998–4006.
- Tenover FC, Arbeit RD, Goering RV, Mickelsen PA, Murray BE, Persing DH, et al. Interpreting chromosomal DNA restriction patterns produced by pulsed-field gel electrophoresis: criteria for bacterial strain typing. *J Clin Microbiol* 1995;**33**: 2233–9.
- Schmitz FJ, Beyer A, Charpentier E, Normark BH, Schade M, Fluit AC, et al. Toxin-gene profile heterogeneity among endemic invasive European Group A streptococcal isolates. *J Infect Dis* 2003;**188**:1578–86.
- Rubio-López V, Valdezate S, Alvarez D, Villalón P, Medina MJ, Salcedo C, et al. Molecular epidemiology, antimicrobial susceptibilities and resistance mechanisms of *Streptococcus pyogenes* isolates resistant to erythromycin and tetracycline in Spain (1994–2006). *BMC Microbiol* 2012;**21**(12):215.
- Sagar V, Kumar R, Ganguly NK, Chakraborti A. Comparative analysis of *emm* type pattern of Group A *Streptococcus* throat and skin isolates from India and their association with closely related SIC, a streptococcal virulence factor. *BMC Microbiol* 2008;**8**:150.
- Chiang-Ni C, Wu AB, Liu CC, Chen KT, Lin YS, Chuang WJ, et al. Emergence of uncommon *emm* types of *Streptococcus pyogenes* among adult patients in southern Taiwan. *J Microbiol Immunol Infect* 2011;**44**:424–9.
- Liang Y, Liu X, Chang H, Ji L, Huang G, Fu Z, et al. Epidemiological and molecular characteristics of clinical isolates of *Streptococcus pyogenes* collected between 2005 and 2008 from Chinese children. *J Med Microbiol* 2012;**61**:975–83.
- Liu X, Shen X, Chang H, Huang G, Fu Z, Zheng Y, et al. High macrolide resistance in *Streptococcus pyogenes* strains isolated from children with pharyngitis in China. *Pediatr Pulmonol* 2009;**44**:436–41.
- Hill HR, Zimmerman RA, Reid GVK, Wilson E, Kilton RM. Foodborne epidemic of streptococcal pharyngitis at the United States Air Force Academy. *N Engl J Med* 1969;**280**:917–21.