Published in final edited form as:

Pediatr Infect Dis J. 2009 January ; 28(1): 13–16. doi:10.1097/INF.0b013e31818128ce.

Streptococcal emm Types in Hawaii: A region with high incidence of acute rheumatic fever

Guliz Erdem, MD^{*},¹, Carla Mizumoto, BSci¹, David Esaki, BA¹, Lucienne Abe, PhD², Karen M. Yamaga, PhD², Venu Reddy, MD¹, and Paul V. Effler, MD⁴

¹ Department of Pediatrics, John A. Burns School of Medicine, University of Hawaii at Manoa

² Department of Tropical Medicine Medical Microbiology and Pharmacology, John A. Burns School of Medicine, University of Hawaii

³ Clinical Microbiology Laboratories, Saint Francis Medical Center

⁴ Hawaii State Department of Health, Honolulu

Abstract

Background—The clinical epidemiology of group A streptococcal (GAS) infections in Hawaii appears different from that in the continental U.S. with frequent skin infections and endemically high rates of acute rheumatic fever (ARF).

Methods—GAS emm types in Hawaii were determined to identify any possible association between the emm types and specific clinical manifestations. A convenience sample of 1,482 Hawaii GAS isolates collected between February 2000 and December 2005 was used. All isolates were characterized by emm sequence typing. The distribution of emm types in Hawaii was compared with the published continental U.S. data for pharyngeal and invasive GAS strains, the CDC database from similar time periods as well as with emm types present in a candidate GAS vaccine.

Results—Ninety three distinct emm types were recognized among the 1,482 GAS isolates. The most frequently identified emm types were emm 12,1,28,4,22,7781,58,65/69,49,74,85,92,75,101>2. Of this study sample, 27 of the 50 invasive GAS isolates belonged to uncommon continental U.S. emm types (54 % in Hawaii cultures vs. 10 % reported from the continental U.S.). Of the 1179 pharyngeal isolates 509 belonged uncommon continental U.S. emm types (43 % in Hawaii cultures vs. 27 % reported from the continental U.S.).

Conclusion—The prevalent *emm* types in Hawaii differ from those in the continental U.S. These unusual *emm* types might limit the effectiveness of any proposed multivalent type specific GAS vaccine in Hawaii.

Keywords

Streptococcus pyogenes; *emm* sequencing; epidemiology; acute rheumatic fever; streptococcal vaccine

Corresponding author: Guliz Erdem, M.D. Associate Professor Department of Pediatrics John A. Burns School of Medicine University of Hawaii at Manoa 651 Ilalo Street, BSB 325D Honolulu, Hawaii 96813 Telephone: 808-692-1666 Fax: 808-692-1984 guliz@hawaii.edu.

Potential conflicts of interest. All authors: no conflicts.

Introduction

Group A streptococci (GAS) are ubiquitous Gram-positive bacteria responsible for a wide array of human illnesses ranging from mild to moderate infections, such as impetigo and tonsillopharyngitis, to more severe diseases, including streptococcal necrotizing fasciitis and streptococcal toxic shock syndrome (1). The clinical epidemiology of GAS infections in Hawaii appears to differ from the continental U.S. in that the non-suppurative sequelae such as acute rheumatic fever (ARF) and nephritis are common (2-6). Unlike the continental U.S. and other developed countries, the annual incidence rate of ARF in Hawaii has remained very high (2,7).

GAS strains are now commonly classified on the basis of sequence variation in the emm gene, which encodes the M protein (8-12). Despite the high number of distinct emm types (more than 125), only five of these are responsible for approximately half of the invasive GAS disease in the continental U.S. (12-15). Furthermore, 23 distinct emm types accounted for approximately 90% of all emm typed sterile body site cultures during continental U.S. surveillance studies in 2003 and early 2004 (15).

Previously, we reported that uncommon emm types were observed among patients with invasive and noninvasive GAS infections in Hawaii when compared with the published studies and available CDC data from the mostly sterile site continental U.S. GAS isolates (3,12,14,16,17). These earlier studies were done in limited number of isolates (< 150 isolates) collected during and following two community-associated necrotizing fasciitis clusters (3,4). In the current report, we determined the GAS emm types in a larger sample of isolates collected over a period of six years and examined the association between the specific/unique GAS types and clinical manifestations in Hawaii.

Materials and Methods

Characterization of isolates and typing methods

A total of 1,482 GAS isolates, collected from patients with mostly noninvasive GAS infections in the state of Hawaii during February 2000 through December 2005 were studied. The majority of GAS isolates were obtained from one of two clinical microbiology laboratories serving most of the civilian hospitals and outpatient clinics throughout the state of Hawaii. This clinical microbiology laboratory receives isolates not only from the island of Oahu but from all of the Hawaiian Islands and serves to major civilian hospitals and outpatient clinics throughout the islands. Approximately 5,000 throat swabs are received annually. The study isolates were identified year around by the clinical laboratories staff and approximately ten isolates were then collected bi-weekly by the study personnel. Isolates from patients with invasive GAS disease were from normally sterile sites (blood, cerebrospinal fluid, synovial fluid, sterile aspirates of deep tissue obtained during surgery).

Clinical isolates were identified as *Streptococcus pyogenes* by standard methodology. All isolates were characterized by emm sequencing as previously described (12). Emm types were further compared to the published continental U.S. data for pharyngeal and invasive GAS strains, and to available information from the CDC surveillance studies of the sterile site cultures (14,17,18). Certain emm types (emm types 1, 12, 28, 89, 6, 49, 44, 75, 77, 4, 22, 11, 2, 5, 92, 18, 27, 73, 82, 58, 114, 94) were observed over 1% of the isolates collected at CDC during 2003 and 2004 studies. These types were also among the commonly identified emm types during the pharyngitis surveillance studies and GAS isolates identified from sterile sites from an earlier time period (14,17,19). The emm types that were not listed were either occasionally identified during these studies or never identified in the continental U.S. during or before surveillance studies or, if identified previously, not for several

decades. In relation to these emm types that are not identified during the surveillance studies, the CDC web site listed the body site and geographic region those strains were recovered and provided comprehensive and publicly accessible information even if these isolates were not part of the surveillance studies and were identified from non-sterile body sites.

Emm types included in a candidate 26-valent streptococcal vaccine used in early phase I and phase II studies were compared with the emm types identified among 1,482 Hawaii isolates (20).

Results

Characterization of isolates and emm/M protein typing

Of the 1,482 GAS isolates which were typed by emm sequencing, 1,179 (80 %) isolates were from throat, 238 (16 %) were from skin cultures; 50 (3 %) were from sterile sites; and 15 (1%) other were from sites including sputum, tracheal aspirate and vagina. A total of 93 distinct emm types were found. Twenty (22%) of these emm types belonged to the types that were also observed over 1% of the continental U.S. (14,17,18). Eighty six distinct emm types were identified among throat cultures, 61 emm types were identified among skin cultures and 24 emm types were identified among sterile site cultures. In aggregation, emm types 1, 2, 4, 12, 22, 28, 49, 58, 65/69, 74, 77, 81, 85, 92 and 101 were the most prevalent types in Hawaii and each of these emm types was identified in >1.5 % of the 1,482 GAS isolates.

Twenty-seven of the 50 (54%) Hawaii sterile site cultures belonged to the emm types that were not commonly identified in the continental U.S. during surveillance studies. By contrast, in continental U.S., only 9.5% of the reported isolates belonged to these unusual emm types (14). Similar to the sterile site cultures, the majority of the skin isolates belonged to the uncommon continental U.S. emm types (128 of the 238 isolates, 54%) (12,21). Most commonly, sterile site cultures belonged to emm types 92 (7 isolates), 49 (6 isolates) and 1 (5 isolates). Emm types 85 (18 isolates), 87 (11 isolates), 74 (10 isolates), 1 (10 isolates), and emm type 81 (9 isolates) were frequently identified among skin and soft tissue infections.

The majority of the Hawaiian throat isolates belonged to the emm types frequently observed among continental U.S. pharyngitis patients [47% of the 1,179 throat isolates] (Table 1) (17,19). Still a significant proportion of the pharyngeal isolates were found to represent infrequent emm types such as emm 71, 74, 78, 81, 85, 95, 109 and st2147. The pharyngeal isolates provided a better measure for possible fluctuations of emm types during the study period. Although, an increase in the number of pharyngeal isolates (83 isolates) belonging to emm type 12 was observed in 2004, there was no other major difference in the percent distribution of the isolates in any study year.

One hundred and fifteen (8% of 1,428 isolates) isolates from Hawaii represented 21 different emm types that were either never reported from the continental U.S. during or before surveillance studies, or if identified, not for several decades. These emm types were emm 67 (3 isolates), 74 (44 isolates), 90 (2 isolates), 93, 97 (2 isolates), 106 (3 isolates), 109 (15 isolates), 113, 121 (4 isolates), 122, st106M, st11014 (7 isolates), st1389, st2147 (14 isolates), stiL62, st7700, std633, stil103 (2 isolates), stknb4, stns1033 (6 isolates), stns554 and stns90 (3 isolates).

To assess the significance of this unusual distribution of emm types, data from Hawaii and continental U.S. isolates were compared to emm types included in a candidate 26-valent streptococcal vaccine (Figure 1) (12,20). Overall, 50 % of the Hawaiian non-sterile site

isolates and 40% of the Hawaii sterile-site isolates were included among the emm types in this candidate vaccine (20). In contrast, this particular candidate vaccine would theoretically provide 77.3% coverage of the invasive isolates reported by CDC and 78.2% of the invasive isolates identified in year 2000 and early 2001 (14). Certain emm types included in the candidate vaccine such as emm 13, 19, 24, 29, 43 were never identified among Hawaiian GAS isolates. Other emm types such as emm 14 (1 isolate), 59 (1 isolate), 76 (1 isolate), 5 (2 isolates), 18 (3 isolates) were rarely identified during the years studied.

Discussion

Epidemiologic studies in U.S. suggest that a significant percentage of the serious suppurative and non-suppurative sequelae of GAS infections are associated with a relatively small number of specific emm/M protein types (1,14,17,19,21-24). Certain GAS emm/M protein types (e.g., M1, M3, M5, M6, M14, M18, M19, M24, M27, M29) are considered to be “rheumatogenic” and certain serotypes have been associated with invasive disease (1,14,17,19,21,22). In contrast, few M/emm types (e.g., M4 and M12) are more frequently associated with uncomplicated pharyngitis (17,19). In geographic areas like Hawaii, where skin infections are commonly observed, very few comprehensive studies have been conducted and the M/emm types associated with each type of infection have not been well documented (23,25,26).

Hawaii offers a unique setting for studying GAS epidemiology in an industrialized country because of its isolated geography and high GAS infection and post-infectious complication rates such as ARF and acute glomerulonephritis (2,6,7). As in reports from other developed countries, emm/M types like emm 1, 12, and 28 were frequently present in Hawaii isolates throughout the study period (14,17-19,21,27-29). Emm type 12 appeared to be the most frequent type. However, the overall frequency of these types, especially emm type 1, was much lower in our Hawaiian sample than they were in continental U.S. studies and these types were mostly associated with non-invasive infections (14,17). Some other emm/M types such as types 3 and 18; that have been associated with ARF and other serious diseases in the continental U.S. were not common among this study sample from Hawaii.

Although our study sample was small and was from a convenience sample rather than a population based prospective surveillance, GAS isolates from patients with invasive infections, such as bacteremia, meningitis, osteomyelitis, arthritis, necrotizing fasciitis and puerperal sepsis, appeared to be associated with emm types not commonly observed in the continental U.S. This finding in invasive isolates parallels the overall study findings in that emm types frequently shared with the continental U.S. (such as emm/M 1, 12 and 28) comprise less than 40% of all Hawaii isolates.

Since the study was largely limited to molecular typing of GAS isolates, clinical information was not systematically collected for isolates belonging to uncomplicated pharyngeal or skin infections. Nevertheless, the large number of isolates studied from patients with different clinical presentations over several years supports that the differences in emm types we observed in the state of Hawaii, compared to those reported from the continental U.S. during a similar time period, are real.

The prevalence of disease-causing GAS emm types circulating in a community with high rates of ARF and other streptococcal infections needs to be carefully considered in designing candidate vaccines made to utilize protection by type-specific immunity. Our results from the state of Hawaii are particularly important during a time when type specific GAS vaccine trials are underway in U.S. and North America (30-32). The M/emm types found in the 26-valent GAS vaccine were chosen on the basis of the most prevalent types associated with

significant invasive disease and uncomplicated pharyngitis in the continental U.S. (15,19,20). Unfortunately, based on this sample of sterile and non-sterile site isolates, such a vaccine may not provide adequate type-specific coverage for our population. This is concerning because Hawaii has consistently had the highest endemic rates of ARF in the entire U.S. (2,5).

In summary, this study reveals in a large GAS convenience sample that the distribution of unusual *emm*/M types in Hawaii appears different from the continental U.S. Several *emm* types infrequently identified in the continental U.S. studies accounted for a significant proportion of the isolates associated with severe GAS disease as well as uncomplicated infections in Hawaii. Comprehensive prospective epidemiological studies will be required when considering the make up of any GAS vaccine based on the type specific immunity.

Acknowledgments

This research was made possible by grant P20RR018727 from the National Center for Research Resources, National Institutes of Health. Preliminary findings were presented at the XVIth Lancefield International Symposium on Streptococci and Streptococcal Diseases in Cairns, Australia and at the annual meeting of the Infectious Diseases Society of America, San Francisco, CA, in October 2005.

References

1. Bisno A. Group A streptococcal infections and acute rheumatic fever. *NEJM* 1991;325:783–793. [PubMed: 1870652]
2. Chun LT, Reddy DV, Yim GK, Yamamoto LG. Acute rheumatic fever in Hawaii: 1966 to 1988. *Hawaii Med J* 1992;51:206–211. [PubMed: 1399542]
3. Erdem G, Ford J, Kanenaka RY, Abe L, Yamaga K, Effler P. Molecular epidemiologic comparison of two group A streptococcal necrotizing fasciitis community clusters in Hawaii. *Clin Infect Dis* 2005;40:1851–1854. [PubMed: 15909276]
4. Erdem G, Abe L, Kanenaka RY, et al. Characterization of a community cluster of group a streptococcal invasive disease in Maui, Hawaii. *Ped Infect Dis J* 2004;23:677–679.
5. Erdem G, Reddy DV, Yamaga K, et al. Group A streptococcal isolates temporally associated with acute rheumatic fever in Hawaii: differences from the continental United States. *Clin Infect Dis* 2007;45:e20–e24. [PubMed: 17599299]
6. Yim H. Rheumatic fever in Hawaii. *Am J Dis Child* 1962;103:715–717. [PubMed: 14009109]
7. Kurahara D, Tokuda A, Grandinetti A, et al. Ethnic differences in risk for pediatric rheumatic illness in a culturally diverse population. *J Rheumatol* 2002;29:379–383. [PubMed: 11838859]
8. Beall, B.; Facklam, R.; Hoenes, T.; Schwartz, B. Application of *emm* gene sequencing and T antigen serology for typing group A streptococcal systemic isolates. In: Hraud, T.; Bouvet, A.; Leclercq, R.; de Montclos, H.; Sicard, M., editors. *Streptococci and the Host*. Plenum Press; New York: 1997. p. 21-27.
9. Fischetti VA. Streptococcal M protein: molecular design and biological behavior. *Clin Microbiol Rev* 1989;2:285–314. [PubMed: 2670192]
10. Johnson, DR.; Kaplan, EL.; Sramek, J. *Laboratory Diagnosis of Group A Streptococcal Infections*. World Health Organization; Geneva: 1996.
11. Johnson DR, Kaplan EL, Vangheem A, Facklam RR, Beall B. Characterization of group A streptococci (*Streptococcus pyogenes*): correlation of M protein and *emm*-gene type with T-protein agglutination pattern and serum opacity factor. *J Med Microbiol* 2006;55:157–164. [PubMed: 16434707]
12. Centers for Disease Control and Prevention (CDC) website
<http://www.cdc.gov/ncidod/biotech/strep/strepindex.htm>. Accessed November 26, 2007
13. Hoe NP, Fullerton KE, Liu M, et al. Molecular genetic analysis of 675 group A streptococcus isolates collected in a carrier study at Lackland Air Force Base, San Antonio, Texas. *J Infect Dis* 2003;188:818–827. [PubMed: 12964112]

14. Li Z, Sakota V, Jackson D, Franklin AR, Beall B. Array of M protein gene subtypes in 1064 recent invasive group A streptococcus isolates recovered from the active bacterial core surveillance. *J Infect Dis* 2003;188:1587–1592. [PubMed: 14624386]
15. O'Brien KL, Levine OS, Schwartz B. The changing epidemiology of group A streptococcus infections. *Semin Ped Infect Dis* 1997;8:10–16.
16. Erdem G, Ford J, Abe L, Yamaga K, Johnson D, Kaplan EL. Erythromycin resistant group A streptococcal isolates from Oahu, Hawaii. *J Clin Microbiol* 2005;43:2497–2499. [PubMed: 15872294]
17. Shulman S, Stollerman G, Beall B, Dale JB, Tanz RR. Temporal changes in streptococcal M protein types and the near-disappearance of acute rheumatic fever in the United States. *Clin Infect Dis* 2006;42:441–447. [PubMed: 16421785]
18. Centers for Disease Control and Prevention (CDC) website
http://www.cdc.gov/ncidod/biotech/files/emmttype_freq2003-04.ppt. Accessed November 26, 2007
19. Shulman ST, Tanz RR, Kabat W, et al. Group A streptococcal pharyngitis serotype surveillance in North America, 2000–2002. *Clin Infect Dis* 2004;39:325–332. [PubMed: 15306998]
20. Hu MC, Walls MA, Stroop S, Reddish MA, Beall B, Dale JB. Immunogenicity of a 26-valent group A streptococcal vaccine. *Infect Immun* 2002;70:2171–77. [PubMed: 11895984]
21. Johnson DR, Stevens DL, Kaplan EL. Epidemiologic analysis of group A streptococcal serotypes associated with severe systemic infections, rheumatic fever, or uncomplicated pharyngitis. *J Infect Dis* 1992;166:374–382. [PubMed: 1634809]
22. Anthony BF, Kaplan EL, Wannamaker LW, Chapman SS. The dynamics of the streptococcal infections in a defined population of children: serotypes associated with skin and respiratory infections. *Am J Epidemiol* 1976;104:652–666. [PubMed: 793381]
23. Bessen DE, Carapetis JR, Beall B, et al. Contrasting molecular epidemiology of group A streptococci causing tropical and nontropical infections of the skin and throat. *J Infect Dis* 2000;182:1109–1116. [PubMed: 10979907]
24. Efstratiou A. Group A streptococci in the 1990s. *J Antimicrob Chemother* 2000;45(Suppl.):3–12. [PubMed: 10759357]
25. Carapetis JR, McDonald M, Wilson NJ. Acute rheumatic fever. *Lancet* 2005;366:155–168. [PubMed: 16005340]
26. Tewodros W, Muhe L, Daniel E, Schalen C, Kronwall G. A one year study of streptococcal infections and their complications among Ethiopian children. *Epidemiol Infect* 1992;109:211–225. [PubMed: 1397112]
27. Beall B, Facklam R, Hoenes T, Schwartz B. Survey of emm gene sequences and T-antigen types from systemic *Streptococcus pyogenes* infection isolates collected in San Francisco, California; Atlanta, Georgia; and Connecticut in 1994 and 1995. *J Clin Microbiol* 1997;35:1231–1235. [PubMed: 9114412]
28. Davies HD, McGeer A, Schwartz B, et al. Invasive group A streptococcal infections in Ontario, Canada. *NEJM* 1996;335:547–554. [PubMed: 8684408]
29. Centers for Disease Control and Prevention (CDC) website
<http://www.cdc.gov/ncidod/dbmd/abcs/survreports.htm>. Accessed November 26, 2007
30. Kotloff KL, Corretti M, Palmer. Safety and immunogenicity of a recombinant multivalent group A streptococcal vaccine in healthy adults: phase 1 trial. *JAMA* 2004;292:709–715. [PubMed: 15304468]
31. Lee GM, Wessels MR. Changing epidemiology of acute rheumatic fever in the United States. *Clin Infect Dis* 2006;42:448–450. [PubMed: 16421786]
32. McNeil SA, Halperin SA, Langley JM, et al. Safety and immunogenicity of 26-valent group A streptococcus vaccine in healthy adult volunteers. *Clin Infect Dis* 2005;41:1114–1122. [PubMed: 16163629]

Figure 1. Comparison of the frequencies of M/emm types included in a multivalent GAS vaccine (20). The candidate vaccine will theoretically provide 77.3% coverage of 3,424 isolates in CDC database (12). This vaccine would cover 50% of all the isolates and 40% of the sterile site isolates in this study.

Table 1

Frequency of emm types identified in GAS isolates from throat cultures in the continental U.S and their frequency among pharyngeal GAS isolates in Hawaii. Percentages used in the table since the U.S. data were given in percentages (17).

<i>Emm</i> type	Hawaii, (1,179 isolates)	United States, 2000-2004 (3,969 isolates)
1	11.4	17.4
2	1.4	7.3
3	0.8	9.3
5	0.2	1.7
6	0.8	6.7
4	5.1	8.3
12	13.7	17.4
22	3.7	1.7
28	9.8	10.9
Others	53.1	19.4