Group A Streptococcal Isolates Temporally Associated with Acute Rheumatic Fever in Hawaii: Differences from the Continental United States

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**Background.** The annual incidence of acute rheumatic fever (ARF) in Hawaii has remained several times higher than that in the continental United States, particularly among ethnic Polynesians. The *emm* types of *Streptococcus pyogenes* that are associated with this nonsuppurative complication have, to our knowledge, not been previously reported in Hawaii.

**Methods.** Patients with ARF were identified through an active surveillance system at Kapiolani Medical Center (Honolulu, HI), the only pediatric tertiary care referral hospital in Hawaii. Specimens were obtained by throat culture from patients who met the Jones criteria for ARF at the time of presentation (63 patients), prior to penicillin treatment, and from consenting family contacts (10 individuals). Eight patients and 2 close family contacts with positive throat culture results were identified from February 2000 through December 2005. Group A streptococci isolates were characterized by *emm* sequence typing.

**Results.** Unusual *emm* types were temporally associated with the onset of ARF. *Emm* types 65/69 (from 2 patients), 71, 92, 93, 98, 103, and 122 were isolated from the 8 patients with ARF, and *emm* types 52 and 101 were isolated from the 2 household contacts.

**Conclusions.** So-called rheumatogenic *emm* types and/or serotypes, which were previously associated with ARF in the continental United States, were not found in this study. Instead, *emm* types that are not commonly included among group A streptococci isolates in the continental United States and that are seldom, if ever, temporally associated with ARF were identified. These findings suggest that unusual group A streptococci *emm* types play a significant role in the epidemiology of ARF in Hawaii.

Group A streptococci (GAS) cause a wide spectrum of diseases and have been associated with serious nonsuppurative complications, such as acute rheumatic fever (ARF) [1–5]. ARF and rheumatic heart disease are estimated to affect nearly 20 million people worldwide; they are the leading causes of cardiovascular death during the first 5 decades of life in developing countries [1, 6, 7]. By contrast, the incidence of ARF has decreased dramatically in most developed countries; only a few outbreaks of ARF have occurred during the past 2 decades in certain US cities [8–14]. A major exception has been the state of Hawaii, where the annual incidence of ARF has remained extraordinarily high during the past several decades [15–17].

Certain GAS M types, such as M1, M3, M5, M6, M14, M18, M19, M24, M27, and M29, have been associated previously with ARF, after evaluating the epidemiology of GAS and correlating this with the epidemiology of ARF [1, 5, 10, 11, 18, 19]. Such associations between GAS M/*emm* types and ARF have not been previously studied in Hawaii. Based on our preliminary studies that GAS *emm* types that are...
influenza. They were also compared with the available Centers for Disease Control and Prevention database (including mostly isolates from sterile site cultures), with data from a US streptococcal pharyngitis surveillance study, and also with emm types included in a candidate multivalent GAS vaccine [10, 24–27].

RESULTS

Sixty-three patients with ARF were identified during the study period. The vast majority of patients with ARF (86%) belonged to Pacific Islander ethnicity groups. Of these patients, the majority were Samoans (87%), and a few were Tongans and Micronesians. Ten of the 63 patients with ARF (15.8%) who had culture performed prior to antibiotic treatment had GAS-positive throat culture results, and all of the patients with positive throat culture results were Polynesians. Of the 10 family members who consented to have cultures performed, 2 had positive throat culture results. Eight of the 10 isolates from patients with ARF and 2 isolates from 2 separate patients’ family contacts were available for the current analysis. Two of the ARF-associated isolates could not be retrieved from the clinical microbiology laboratory. None of the ARF-associated isolates had mucoid colony morphology on blood agar plates.

emm Types 65/69 (isolated from 2 patients), 71, 92, 93, 98, 103, and 122 were isolated from the throats of patients with ARF, and emm types 52 and 101 were isolated from the 2 contacts at the time of disease onset [28] (table 1). Two siblings from the same family received a diagnosis of ARF (only 1 of these siblings had a GAS-positive culture result, which was emm 122). None of the emm types that we report here are classically known to be associated with ARF. These emm types were also

Table 1. The distribution of emm types among patients with acute rheumatic fever (ARF) in Hawaii, compared with pharyngitis isolates from Hawaii and sterile site isolates from the continental United States [24, 28].

<table>
<thead>
<tr>
<th>emm types isolated from patients with ARF in Hawaii</th>
<th>No. of pharyngitis isolates from Hawaii</th>
<th>No. of isolates from the continental United States</th>
</tr>
</thead>
<tbody>
<tr>
<td>65/69</td>
<td>12</td>
<td>4</td>
</tr>
<tr>
<td>71</td>
<td>14</td>
<td>1</td>
</tr>
<tr>
<td>92</td>
<td>16</td>
<td>50</td>
</tr>
<tr>
<td>93</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>98</td>
<td>6</td>
<td>1</td>
</tr>
<tr>
<td>103</td>
<td>4</td>
<td>1</td>
</tr>
<tr>
<td>122</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>

NOTE. There were 1258 total pharyngitis isolates from Hawaii and 3424 isolates (from the period 1995–2001) included in the Centers for Disease Control and Prevention surveillance data from the United States.

* Isolated from 2 patients.

MATERIALS AND METHODS

Patients and bacterial isolates. Active prospective surveillance of pediatric patients (age range, 5–16 years) with ARF was performed through notification of pediatric infectious diseases, cardiology, and rheumatology consultants at Kapiolani Medical Center (affiliated with the John A. Burns School of Medicine, University of Hawaii, Honolulu, HI), a 197-bed tertiary-level referral hospital in Honolulu that serves pediatric patients in Hawaii and throughout the Pacific Islands. The diagnosis of ARF was made according to the updated Jones criteria [21]. Essentially, all patients with ARF are seen and/or followed up by pediatric specialists associated with Kapiolani Medical Center. Because patients with ARF are occasionally seen at the Tripler Army Medical Center (Honolulu, HI), specialists at the Tripler Army Medical Center were informed of the active surveillance study to identify any patients with ARF. These measures were taken to maximize ARF surveillance. Inpatient charts and records of outpatients who received a diagnosis of ARF during the study period were identified after appropriate institutional review board approvals. Patient records were retrospectively reviewed to identify any possible missed and unrecorded manifestations at the time of diagnosis.

GAS isolates obtained from patients with ARF prior to penicillin treatment and from consenting family contacts were systematically collected from February 2000 through December 2005. These isolates were characterized as part of an ongoing study of 1660 GAS isolates identified in Hawaii from sterile and nonsterile body sites during the same study period (G.E., unpublished data). Clinical isolates were identified as Streptococcus pyogenes by standard methodology. Morphology of β-hemolytic bacterial colonies was also studied. T agglutination pattern, M serotype, and type-specific serum opacity factor typing were determined for 30 isolates at the World Health Organization Collaborating Center for Reference and Research on Streptococci at the University of Minnesota (Minneapolis) [22, 23]. All isolates were characterized by emm sequencing, as described elsewhere [24]. Emm types were further compared according to available information from the Centers for Disease Control and Prevention surveillance studies [24, 25]. The distribution of emm types classically associated with ARF and emm types identified in patients with ARF in Hawaii were then compared with the sample of 1660 total isolates, which included

1258 pharyngeal GAS isolates. They were also compared with the available Centers for Disease Control and Prevention database (including mostly isolates from sterile site cultures), with data from a US streptococcal pharyngitis surveillance study, and also with emm types included in a candidate multivalent GAS vaccine [10, 24–27].
among the infrequently identified \textit{emm} types in Hawaii (table 2). Admittedly, an isolate identified from the throat of a patient with ARF or from a family member cannot be assumed with certainty to be the exact one that triggered the ARF process, because colonization with another GAS strain is a possibility.

We also evaluated the frequency of classic ARF-associated \textit{emm} types in Hawaii. These comprised only \(~14\)% of the 1660 total GAS isolates and only \(~4\)% of the 1258 pharyngeal isolates in Hawaii (versus \(~18\)% of the 3969 pharyngeal isolates in the continental US surveillance studies) [10]. Only 1 \textit{emm} type (\textit{emm} 92) from the patients with ARF in Hawaii was included in the candidate 26-valent vaccine [26].

**DISCUSSION**

We did not find the previously reported, classic ARF-associated “rheumatogenic” \textit{emm} types and/or serotypes in our study. Instead, we identified \textit{emm} types that are uncommon among the continental US isolates and that are seldom, if ever, reported to be temporally associated with ARF. These results suggest that uncommon GAS \textit{emm} types may have a significant role in the epidemiology of ARF in Hawaii. It might be argued that factors other than the specific GAS \textit{emm} types observed are also important in the increased rates of ARF among our population.

Environmental conditions, host susceptibility, and other biologic characteristics of GAS are known to relate to high rates of ARF [1, 5, 10, 11]. Among these, the degree of crowding and overall income affecting access to health care in affected population groups could be considered in Hawaii. However, no obvious difference was found between the annual incomes of families with a patient with ARF and an ethnicity-based control group in Hawaii [29]. Moreover, pediatricians and other health care professionals practicing in Hawaii do not see a significant difference in access to health care in the affected population and age groups (R. Rudoy and M. Melish, personal communications). Another environmental factor, climate, may be important. Pharyngitis symptoms appear to be less common than skin infections in Hawaii, where high ARF incidence rates and the subtropical climate are similar to those in certain regions in Australia and New Zealand (R. Rudoy and M. Melish, personal communications) [9, 28, 30–32]. As in Samoan persons, certain Polynesian populations, such as the Maori in New Zealand, have been shown to have higher rates of ARF [9, 30]. The unusually high number of Polynesian patients affected by ARF obviously would warrant further research for specific host factors.

In addition to environmental and host factors, changes in the biologic characteristics of GAS have been associated with the fluctuations and changes in the rates of ARF [1, 10, 11]. Infection with GAS as the inciting event in the development of ARF is well established. Some of the proposed characteristics of the so-called rheumatogenic strains of GAS include the following: affinity for the pharynx, mucoid colony morphology, belonging to a relatively small number of specific M types, failure to produce serum opacity factor, possessing Class I M protein, and sharing epitopes with heart tissue [2, 10, 33]. Encapsulation and other possible virulence factors were not features of this study, but it is worth observing that none of the 8 temporally ARF-associated isolates showed mucoid colony formation on blood agar plates, and 2 isolates belonged to serum opacity factor–positive \textit{emm} types (\textit{emm} 92 and 103) [23].

The M proteins of GAS are not only important virulence factors and provide the basis for M and \textit{emm} typing schemes, but they are also widely studied in association with rheumatogenicity [2, 34–39]. On the basis of carefully performed studies from decades earlier, pharyngitis- and pyoderma-related M types and ARF-associated M types have been identified in patients living in temperate climates [1, 40, 41]. In 1 recent surveillance study, the reduction in the incidence of ARF in the continental United States was found to parallel reductions in the prevalence of almost all putative rheumatogenic types of GAS over the past 4 decades [10, 11]. Similar to these results, putative rheumatogenic types were uncommon in Hawaii. However, the association of the decrease of ARF in the continental United States with a decrease in classic “rheumatogenic” types was not true in Hawaii, where a high rate of ARF persists in the absence of these \textit{emm} types. More importantly, the ARF-associated GAS isolates belonged to \textit{emm} types that were prevalent neither in the continental United States, nor in Hawaii. This may be explained partially by the different climates in Hawaii and the continental United States.

In tropical areas and in developing countries, where ARF incidence is high and where impetigo is more common than

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**Table 2.** The distribution of “rheumatogenic” \textit{emm} types in the continental United States and Hawaii [24, 28].

<table>
<thead>
<tr>
<th>“Rheumatogenic” types</th>
<th>No. (%) of isolates from Hawaii</th>
<th>No. (%) of isolates from the continental United States</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>197 (11.9)</td>
<td>685 (20)</td>
</tr>
<tr>
<td>3</td>
<td>16 (1.0)</td>
<td>316 (9.2)</td>
</tr>
<tr>
<td>5</td>
<td>2 (&lt;1)</td>
<td>58 (1.7)</td>
</tr>
<tr>
<td>6</td>
<td>12 (&lt;1)</td>
<td>83 (2.4)</td>
</tr>
<tr>
<td>14</td>
<td>1 (&lt;1)</td>
<td>1</td>
</tr>
<tr>
<td>18</td>
<td>3 (&lt;1)</td>
<td>50 (1.5)</td>
</tr>
<tr>
<td>19</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>24</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>27</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>29</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

**NOTE.** There were 1660 total pharyngitis isolates from Hawaii and 3424 sterile site isolates (from the period 1995–2001) included in the Centers for Disease Control and Prevention surveillance data from the United States.
pharyngitis, very few comprehensive studies have been conducted and the M/emm types associated with disease are not well documented [9, 11, 32, 42]. Higher rates of impetigo and lower GAS pharyngeal carriage rates, with endemically higher rates of ARF, suggest that the dynamics of GAS acquisition may be different in some tropical climates, and this may also be true in Hawaii [9]. Almost all GAS isolates from patients with ARF in this study belonged to less prevalent emm types in the United States, as well as in the convenience sample from Hawaii. These types have not been previously reported to be associated with ARF, and none of the emm types isolated from our patients with ARF belonged to known “rheumatogenic” GAS emm types.

Studies from regions with a high incidence of ARF, such as Australia and New Zealand, have similarly shown that previously identified “rheumatogenic” serotypes were not commonly encountered among patients with streptococcal disease in these locales [9, 30]. The higher number of emm types identified in Hawaii, with an overall higher number of unusual and infrequent M/emm types among GAS isolates, was also similar. In populations with a wider range of circulating GAS M/emm types, such as in Hawaii, the association between the disease and the specific, so-called rheumatogenic M/emm types may be less clear. Moreover, the assumed link between the disease and the disease-causing emm types is only temporal and may itself not be sufficient to build a hypothesis for a possible association of initial infection site and ARF. Nevertheless, the identification of M/emm types that were previously unrecognized as being “rheumatogenic” is significant.

The prevalence of GAS emm types circulating in a community with high rates of ARF and other streptococcal infections needs to be considered when designing and customizing candidate vaccines to induce type-specific immunity. Our results from Hawaii are particularly important during a time when type-specific GAS vaccine trials are underway in the continental United States and in other regions of North America [11, 26]. Based on our sample, such a candidate vaccine would only provide coverage for 1 emm type isolated from patients with ARF in Hawaii. These findings suggest that GAS strains associated with ARF may be quite different in these areas with high rates of ARF, complicating future vaccine strategies.

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Potential conflicts of interest. All authors: no conflicts.

References

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