

***Neisseria meningitidis* Serogroup W135 Isolates Associated with the ET-37 Complex**

To the Editor: As of April 20, 2000, >300 laboratory-confirmed cases of meningococcal disease caused by *N. meningitidis* serogroup W135 (NMW135) have been reported in Saudi Arabia and nine other countries among Hajj pilgrims or their close contacts (1). This is the first reported multinational outbreak of W135 meningococcal disease. NMW135 accounts for <5% of meningococcal disease in the United States and worldwide. The main cause of meningococcal disease outbreaks, especially in the African "meningitis belt," which extends from Ethiopia in the east to Senegal in the west, has long been *N. meningitidis* serogroup A. An epidemic, caused by a particular clonal group of serogroup A, identified by multilocus enzyme electrophoresis (MEE) as subgroup III-1, occurred in Nepal (1983 and 1984), Pakistan, and India (1985) (2) and may have caused earlier epidemics in China. In 1987, an outbreak of group A meningococcal disease caused by the same clonal group occurred in association with the annual Moslem pilgrimage to Mecca (Hajj) (3). At that time, Saudi Arabia implemented vaccination requirements for all entering pilgrims. The vaccine formulations vary; bivalent A/C vaccine or quadrivalent A/C/Y/W135 vaccine (as licensed in the United States) is used.

We present molecular characterization results of NMW135 isolates from four U.S. patients with meningococcal disease. These cases were epidemiologically linked to this year's Hajj in Saudi Arabia, which concluded on March 17, 2000 (4). We examine the origin and potential relatedness of the NMW135 isolates to some of the major *N. meningitidis* virulent clones. Two patients (both vaccinated with quadrivalent A/C/Y/W135 vaccine) were returning pilgrims; one was a close household contact of another pilgrim, the other had other possible contacts with the U.S. pilgrims. Three isolates were identified as NMW135 at the New York City Health Department and the fourth at the California Department of Health. Serogroup identification was confirmed at the Centers for Disease Control and Prevention (CDC) by standard microbiologic methods (5).

MEE has long been the standard method for molecular subtyping of *N. meningitidis* and has allowed identification of several major epidemic-prone clones. When the MEE system established by Caugant et al. (6) and that used at CDC were recently compared, they demonstrated excellent correlation segregating individual electrophoretic types (ETs) into similar clusters. A virulent clone, designated ET-37 complex, which contains >50 different ETs, is mainly composed of *N. meningitidis* serogroup C. In the United States, strains of ET-24 (within the ET-37 complex) are the main cause of meningococcal disease outbreaks and among the most frequent causes of sporadic meningococcal disease (7). The four Hajj-associated NMW135 isolates were of ET-927, which is located in a small cluster more closely related to the ET-37 complex (at a genetic distance <0.20) than to any other NMW135 isolate or any other major virulent clonal group among >2,000 *N. meningitidis* isolates in our collection (8). This cluster contains two other NMW135 isolates: one from Indonesia from a pilgrim returning from the 1996 Hajj and another from Canada (1997) for which additional epidemiologic information is not available.

Serotyping and serosubtyping showed that all four W135 isolates were 2a:P1.5,2, most frequently seen in *N. meningitidis* serogroup B and C isolates (9). Sequencing of the variable regions (VR) of the *porA* gene showed that these four isolates had VR1 and VR2 sequences identical to those of the prototype P1.5,2 strain. Strains of the same serogroup/serosubtype have been already identified in France and the Netherlands isolated from four patients with Hajj 2000 association (1), and earlier in Gambia (1990-1995) and Mali (1995) (10). A single NMW135 isolate from Gambia and one from Mali are listed in the multilocus sequence typing (MLST) database (Oxford database) as being of sequence type 11, typically seen in isolates of the ET-37 complex (11). MLST provides results comparable to those of MEE for classification and taxonomic purposes. We are evaluating the usefulness of MLST for outbreak studies. The DNA sequences of the 16S rRNA gene of the four Hajj-associated NMW135 isolates were identical to each other and to those of the California (1995) outbreak-related serogroup C ET-24 strains and an Ohio (1997)

sporadic serogroup C ET-24 strain. The 16S rRNA gene sequences of 66 *N. meningitidis* isolates representing serogroups A, B, C, W135, Z, and Y were diverse, with nine different sequences among the NMW135 isolates. Finally, all four recent NMW135 isolates had identical *NheI* pulsed-field gel electrophoresis (PFGE) patterns distinct from patterns seen in other NMW135 isolates. All these molecular markers were clearly unique in NMW135 isolates previously identified in the United States or isolated at the same time as the Hajj-associated isolates but with no epidemiologic link to the current outbreak. These unique markers allowed easy differentiation of the imported, Hajj-associated isolates from other sporadic NMW135 isolates circulating in the United States.

It has been shown previously that NMW135 strains can exist in widely divergent clonal groups. Our data suggest that strains like those associated with this year's Hajj have been in circulation in human populations for at least several years in different parts of the world. Given that the Hajj is a large, yearly event, high-level exposure of pilgrims to respiratory secretions and subsequent spread of infection to many countries by returning pilgrims may turn W135 meningococcal disease into a global health threat. Continued surveillance, as well as increased awareness of meningococcal disease caused by *N. meningitidis* of this serogroup by physicians and the public, is needed. Efforts to measure the efficacy of the quadrivalent meningococcal vaccine for prevention of W135 meningococcal disease should be considered. To get a better global understanding of W135 meningococcal disease, we are conducting a large multicenter study on molecular characterization of >50 Hajj-associated NMW135 isolates from Saudi Arabia, France, Singapore, and Finland, and 50 other W135 isolates from throughout the world.

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Gnathostomosis in Fish from Tres Palos Lagoon, Guerrero, Mexico

To the Editor: Since the first two cases of human gnathostomosis in Mexico were reported in 1970 (1), >1,500 cases have been reported in Nayarit, Sinaloa, Oaxaca, Guerrero, Veracruz, and Tamaulipas states (2). In Acapulco, Guerrero, 98 cases have been described; the intermediate or definitive hosts in this region are unknown (3,4).