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Molecular Epidemiology of an Outbreak of Meningococcal Disease in a University Community

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Over a 2-month period, five cases of serogroup C meningococcal disease occurred in Iowa City, Iowa. Two patients were unacquainted university students who had independently visited another university with endemic meningococcal disease. Isolates from these patients had DNA fingerprints identical to those of the isolates responsible for infections on the other campus. Three cases for which the patients' isolates had a different DNA fingerprint were linked to visiting a local tavern. To disrupt the outbreak, the University of Iowa offered free meningococcal vaccine to all students. This report demonstrates that outbreaks of meningococcal disease may be due to more than one circulating strain and illustrates the utility of pulsed-field gel electrophoresis in defining the molecular epidemiology of meningococcal infections.

Although the incidence of meningococcal disease in the United States has declined in recent years (7), the number of outbreaks of serogroup C meningococcal disease has increased dramatically. In the 30-month period from 1991 through mid-1993, 10 outbreaks were reported (6), 4 of which occurred on college campuses (3, 5, 6, 14). This contrasts strikingly with the 11 outbreaks reported for the entire decade 1981 through 1990 (6). We describe an outbreak of serogroup C meningococcal disease occurring in 1992 in Iowa City, Iowa, a university community located in a semirural county (population, 96,000). The mean countywide incidence rate of meningococcal disease for 1987 to 1991 was 0.6 case per 100,000 persons per year. For 1992 the incidence was 5.2/100,000 ($P < 0.0001$).

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On 23 October 1992, a 20-year-old male University of Iowa student (patient 1) presented with meningococcal meningitis and bacteremia. One week previously, he had attended a football game and social activities at the University of Illinois, Urbana-Champaign. The Illinois campus had experienced an endemic problem with meningococcal disease, with nine cases (three deaths) occurring between February 1991 and April 1992 (5). In February 1992 meningococcal vaccine was administered to approximately 16,000 students at the Illinois campus (6).

On 3 November, an 18-year-old female University of Iowa student (patient 2) presented with meningococcemia and right-shoulder septic arthritis. Although she had attended the same football game as patient 1, they were not acquainted and had no known contact or common social activities in Iowa City or Urbana-Champaign. Antimicrobial susceptibility patterns from the first two cases were nearly identical: notably, both isolates were susceptible to rifampin and resistant to trimethoprim-sulfamethoxazole, as shown by the E test (AB Biodisk, Solna, Sweden). Minor differences in MICs were noted for erythromycin, penicillin, and vancomycin.

Patient 3, a 22-year-old male bartender who was not a student, presented on 19 November with meningococcemia and polyarticular septic arthritis. An epidemiologic link with the first two cases could not be established. Moreover, the antibiogram of this patient's isolate was different from those for the first two patients; patient 3's isolate was resistant to rifampin and susceptible to trimethoprim-sulfamethoxazole. Ten days later, a 21-year-old male college student (patient 4) presented with meningococcal meningitis. His isolate's antibiogram was similar to that of the isolate from patient 3 (rifampin resistant, trimethoprim-sulfamethoxazole susceptible). Of note, patient 4 was a frequent patron of the tavern where patient 3 worked as a bartender, although they were not personally acquainted. On 15 December 1992, patient 5, a 21-year-old woman who was not a college student, presented with meningococcemia. She had visited the tavern associated with patients 3 and 4 approximately 1 week before her illness developed. The antibiogram of her isolate matched those of patients 3 and 4. Table 1 summarizes the epidemiological data for all five patients.

Molecular typing was performed for isolates associated with the Iowa outbreak, the previous outbreak at Urbana-Champaign, and cases in other areas of Iowa and Illinois which appeared not to be epidemiologically linked to the outbreaks on either campus. Genomic DNA was prepared in agarose plugs and digested with the restriction enzyme *Sfi*I. DNA digests were separated by pulsed-field gel electrophoresis with a CHEF-DR II electrophoresis cell (Bio-Rad, Melville, N.Y.). Gels were stained with ethidium bromide to yield DNA fingerprints (2, 10) and were photographed. Two distinct patterns were found (Fig. 1): the patterns for isolates from patients 1 and 2 and some previously obtained Illinois isolates (fingerprint A) were readily distinguished from the patterns for isolates from patients 3, 4, and 5 (fingerprint B).

After the third case was reported (no deaths occurred at Iowa), the University of Iowa launched a mass, free vaccination program for its students. Over a 5-day period, approximately 18,000 (67%) of the university's 27,000 students received the tetravalent meningococcal vaccine. In the ensuing

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TABLE 1. Summary of cases in the meningococcal disease outbreak

Patient no.	Age (yr)/ gender[a]	Occupation	Potential site of exposure	Date of presentation (mo/day/yr)	Clinical presentation	Site(s) yielding positive culture(s)[b]	DNA fingerprint pattern
1	20/M	University of Iowa student	Football game in Illinois[c]	10/23/92	Headache, fever, rash	Blood, CSF	A
2	18/F	University of Iowa student	Football game in Illinois[c]	11/03/92	Fever, rash, shoulder arthritis	Blood, right shoulder	A
3	22/M	Bartender at bar A	Bar A	11/10/92	Fever, rash, polyarticular arthritis	Blood, pharynx	B
4	21/M	University of Iowa student	Bar A	11/29/92	Fever, headache, meningismus	CSF	B
5	21/F	Unemployed	Bar A	12/15/92	Fever, rash, headache, meningismus	Blood	B

[a] M, male; F, female.
[b] CSF, cerebrospinal fluid. With patient 3, gram-negative diplococci were seen in aspirates from both wrists, both ankles, and the left knee, but cultures were sterile.
[c] 17 October 1992.

year, no cases of group C meningococcal disease were reported for University of Iowa students. One case of meningococcemia due to serogroup B (a serogroup not included in the vaccine) occurred in a student in April 1993.

In this outbreak of five cases of serogroup C meningococcal disease, the epidemiology and antimicrobial susceptibility profiles suggested the existence of two subclusters. Genomic analysis confirmed that two different strains were responsible and allowed the organisms' sources to be traced. For the first two cases, the exact site of transmission is undetermined; however, the infections at Iowa and Illinois appeared to be linked to attendance at a football game between the two schools and/or to related social activities.

The remaining three cases were all linked epidemiologically to a local Iowa tavern and included a bartender. Interestingly, a case-control study at Urbana-Champaign showed that students with meningococcal disease were significantly more likely to have patronized campus bars (5). Moreover, group C meningococcal carriage rates were 0.2% for students without exposure to bars, 0.5% for patrons of any bar, 4% for patrons of one particular bar, and 14% for employees of any bar (5). A

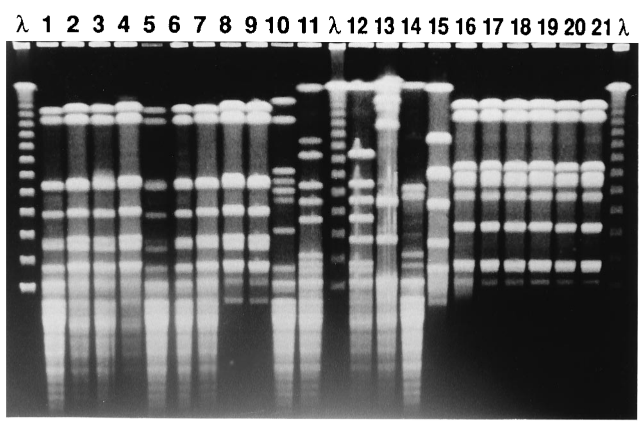

FIG. 1. DNA fingerprints of serogroup C meningococcal isolates. Isolates were collected (February 1991 to December 1992) from patients with invasive meningococcal disease and were prepared with restriction enzyme *Sfi*I. Electrophoretic parameters: initial pulse, 5 s; final pulse, 60 s; pulse angle, 120°; voltage, 6 V/cm; time, 23 h; temperature, 13°C. Lanes 8 and 9, cases 1 and 2, respectively; lanes 16 to 18, cases 3 to 5, respectively; lanes 2 to 5 and 11, Urbana-Champaign isolates; lanes 1, 6, 7, 10, and 12 to 14, other Illinois (non-Urbana-Champaign) isolates; lanes 15 and 19 to 21, isolates from western Iowa (significant geographic distance from Iowa City); lanes λ, lambda phage DNA ladders (interval = 48.5 kbp).

previous study in England found that persons who had meningococcal disease were more likely to have regularly visited pubs than controls (12) and suggested that this may be related to cigarette smoke exposure. Several case-control studies have evaluated active and passive smoking as risk factors for meningococcal carriage or disease. Three of these studies found active smoking to be a risk factor for carriage (1, 11, 13). Passive smoking was demonstrated to be a risk factor for meningococcal carriage in two studies (11, 13) and to be a risk factor for invasive disease in three studies (4, 9, 12).

At the time of the Iowa City outbreak there were no available recommendations on when to initiate a mass vaccination to interrupt an outbreak. The decision to vaccinate the student body was based on the evidence that a meningococcal strain, associated with endemic infection and deaths at another campus, was introduced into the University of Iowa community and appeared to be spreading. Subsequently, the Canadian Laboratory Centre for Disease Control issued guidelines on the control of meningococcal disease, although specific recommendations for college settings were not given (8). Currently, incoming students at the University of Iowa are offered but not required to receive meningococcal vaccine. On the basis of the accelerated activity of meningococcal disease on college campuses and the specter of life-threatening illness, a cost-benefit analysis of mandatory vaccination at the time of matriculation is warranted.

Although multilocus enzyme electrophoresis has been widely used to study the epidemiology of meningococci, pulsed-field gel electrophoresis has the advantage of being able to identify subclones of the same electrophoretic type (10). In this outbreak we demonstrated the utility of this technique in separating an outbreak into two subclusters caused by different organisms. Moreover, it allowed us to epidemiologically link meningococcal disease on our campus to that involving another university in an adjacent state.

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