MENINGOCOCCAL DISEASE IN AUSTRALIA

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Abstract

Although the overall incidence if invasive meningococcal disease in Australia is not high by international standards, *Neisseria meningitidis* is a major cause of bacterial meningitis and septicaemia in a number of patient subgroups. Clusters of cases are not infrequently recorded. This situation presents significant challenges to public health responses in both the immediate and longer term. Resolution of these problems would be advanced by more accurate and complete data on disease patterns, invasive meningococcal subtypes and antibiotic susceptibility patterns. The prospect of more efficacious conjugate vaccines gives further impetus to the need for improvement of these data. ***Comm Dis Intell* 1996;20:368-371.**

Introduction

Neisseria meningitidis is a strictly human, but highly versatile pathogen responsible for invasive disease throughout the world. Meningococcal infection is manifest most frequently as a meningitis or as an often fulminant septicaemia. It occurs in epidemics, smaller outbreaks and sporadically without apparent linkage to other cases. Rapid and accurate characterisation of isolates is an essential part of the preventative public health response to a possible outbreak.

This review will summarise features of N. meningitidis which allow differentiation of strains, what is known about the epidemiology of meningococcal disease in Australia, antimicrobial susceptibility patterns and their influence on treatment recommendations, outcome of infection, and potential roles of new vaccines in Australia.

Epidemiology of meningococcal disease in Australia

Incidence of disease

Data on many aspects of meningococcal disease in Australia are incomplete. The National Notifiable Diseases Surveillance System provides information on sex, age, onset date and place of occurrence of cases, but provides no data on strains causing disease, clinical presentation and outcome. Meningococcal disease was not a separate notifiable disease in some States until the late 1980s. It is an infection with cyclical peaks of incidence. Notification of 'meningitis' showed a peak of 33.1 cases per 100,000 in 1942 (2,371 cases); this was followed by a slow decline in incidence until another peak in the early 1970s. By the late 1970s notifications were categorised nationally as meningococcal disease[1]. There was then another decline to less then 0.5 cases per 100,000 until 1987, when an increase was again seen. The increase has been sustained with annual rates of 1.6 to 2.2 per 100,000 1991-94[1,2,3]. The highest attack rates are seen in children less than five years of age, particularly in Aboriginal communities in central and northern Australia. In Aboriginal children less than five years of age in the Northern Territory, Western Australia and New South Wales, the incidence since 1991 is estimated to be 66 per 100,000[4]. The incidence decreases with age, but in Australia, as in many other developed countries, there is a second smaller peak of incidence in the 15-20 years age group. The peak incidence for meningococcal disease is during winter, July to September.

Knowledge of strains causing disease in Australia

The National Neisseria Network (NNN), a laboratory-based meningococcal surveillance system commenced providing standardised national information on serogroups, antibiotic susceptibility patterns, patterns of disease and mortality rates in 1994[5]. National data on serotypes and subtypes were provided from 1995. Otherwise, data on strains and patterns of disease in Australia have been sporadic and regional. Large-scale outbreaks of meningococcal disease have been confined to Aboriginal communities in central and northern Australia.

In 1971 to 1973, there was an epidemic in central Australia affecting predominantly Aboriginal communities with an annual attack rate of 321 per 100,000[6]. Organisms which were serogrouped from this outbreak were serogroup A. In 1987 to 1991, another epidemic, predominantly serogroup A, recurred in central Australia[7]. Both these outbreaks had characteristics of epidemics of serogroup A seen regularly in Africa and other developing countries. Apart from these outbreaks, serogroup A organisms are rarely isolated[5,8,9]. Serogroup B organisms have been the main cause of sporadic disease in Australia, as in other developed countries[5,8,9]. From 1987 to the early 1990s, there were reports from different areas of Australia of an increase in the frequency of serogroup C isolations.

Meningococcal strain differentiation

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Strain differentiation is undertaken to determine isolate relatedness and identify possible outbreaks, to map the spread of virulent strains, and to assist in determining vaccine efficacy.

Serogroup, serotype, subtype

The most universal methods of strain differentiation involve the sequential serological procedures of serogrouping, serotyping and subtyping. Organisms isolated from systemic sites have polysaccharide capsules which allow differentiation into 13 distinct serogroups. Serogroups A, B, and C account for 90% of invasive infections. There are five different structural classes of outer membrane proteins (OMPs) located in the cell wall of the organism. *Neisseria meningitidis* is a transformable organism capable of frequent recombination events causing alterations in antigens exposed on its surface. Variations in these OMPs can be used to further differentiate organisms within serogroups. Meningococci of different serogroups can be divided into 20 serotypes with six common ones, using monoclonal antibodies directed against class 2/3 OMPs. Similarly, variations in class 1 OMPs form the basis of a subtyping system involving 13 common subtypes[10].

Most laboratories perform serogrouping of invasive meningococcal isolates. This basic information will provide some valuable epidemiological information. The epidemic forms of meningococcal disease affecting many thousands of people in Africa and other developing countries are almost always serogroup A. In developed countries where endemic and hyperendemic disease is more usual, there are geographical and temporal variations in the occurrence of serogroup B and C strains.

Monoclonal antibodies for serosubtyping were developed over ten years ago, but only in the last three to four years they have been widely available in a standardised form. However, a significant proportion of strains (about 40%) will be nontypable by one or other method. This is partly related to the titre of monoclonal antibody attainable, but more importantly to the inability of monoclonal antibodies to recognise many variants of OMPs caused by recombination events, or to react differently with strains with only very minor genetic differences.

Multilocus enzyme electrophoresis (MLEE)

This is a phenotypic method which identifies 14 to 20 intracellular enzymes by electrophoretic separation and action on a range of substrates[11]. Because these enzymes are less susceptible to genetic variation than cell surface proteins, their electrophoretic mobilities can be used to group organisms into clones or clusters called electrophoretic complexes. All isolates can be assigned an electrophoretic type (ET) and grouped into genetically related ET complexes. The method has been used in many countries, allowing study of the global spread of some clones and changing incidence of particular ETs within countries. Unfortunately, there is very little information about ETs of isolates in Australia.

DNA typing techniques

A range of DNA typing techniques has been applied to meningococci; the most widely used is pulsed field gel electrophoresis (PFGE)[12]. PFGE, which examines the whole chromosome, has mainly been used to examine whether epidemiologically related strains collected during an outbreak are genetically related or are the same strain. It complements the epidemiological data. Interpretation is subjective, and small genetic variations which occur frequently in meningococci which are closely related may further make interpretation difficult. Recent recommendations for criteria for PFGE typing, recognising that evidence for clonality is relative rather than absolute, will assist with these problems[13]. Other methods which examine total chromosomal DNA include restriction fragment length polymorphisms (RFLP) with cloned meningococcal DNA as a probe or with ribosomal RNA as a probe (ribotyping)[14,15].

There are also DNA typing techniques applied to meningococci which examine variation in single genes or small numbers of genes. Polymerase chain reaction (PCR) amplification of the *por* A gene encoding the class 1 OMP with RFLP of PCR products and random amplified polymorphic DNA PCR are two such techniques[16,17].

DNA typing techniques have the advantage over serological techniques in that all strains are typable. In general, DNA typing correlates with results obtained by MLEE and will also discriminate amongst organisms in specific ET complexes. Reports on the use of these techniques have mainly been retrospective analyses of collections of organisms: a number of techniques has reliably identified epidemiologically related strains[18].

Choice of strain differentiation techniques

The choice of a typing technique will depend in part on practical considerations such as cost, technical complexity, and time to obtain results, but will also be influenced by the reason strain differentiation is being undertaken.

Outbreak investigations

If rapid identification of epidemiologiccally related cases is required so that a public health response can be mounted to control a possible outbreak, a DNA typing technique is ideal. Serogrouping and serosubtyping will be helpful, but only if strains are typable. MLEE, although typing all strains, is too slow and complex for outbreak identification. *Por* A PCR amplification with RFLP of products is one of the most rapid DNA typing techniques. Random amplification polymorphic DNA (RAPD) PCR, another rapid typing technique, has lacked reproducibility in our hands. PFGE or RFLP with meningococcal DNA or rRNA probes take several days to perform.

Studying the epidemiology of meningococcal disease over time

Serotyping and MLEE are standardised strain differentiation techniques used in many countries over a number of years. Valuable information can be obtained about the spread of strains in different geographic areas over time and comparisons made with other countries by characterisation of all meningococcal isolates using these two methods.

There has been little work using DNA typing techniques to study prospectively meningococci causing sporadic disease. Ongoing characterisation of serogroup B meningococci in New Zealand has indicated that the marked increase in disease incidence since 1991 coincided with an increase in frequency of one particular genotype as defined by RFLP, this type being uncommon before 1991[19]. DNA typing techniques examining total chromosomal DNA may be more applicable to epidemiological investigations of spread of sporadic strains than techniques examining variation in smaller numbers of genes. Combinations of techniques may be required to give optimal information.

In addition, there were reports of clustering of cases of serogroup C disease, both in urban and rural settings[20,21,22,23]. At that time there was increased serogroup C activity in a number of countries throughout the world. However, data from the NNN in 1994-95 indicate that serogroup B organisms again predominate and have not been associated with clusters or outbreaks of disease in Australia[5].

Data are limited about serotypes and subtypes of meningococci causing disease in Australia, and there has been considerable geographical variation in the prevalence of particular serotypes and subtypes. In South Australia and the Northern Territory in 1971 to 1989, the predominant serotypes were 4, 2a, 15, and 14, with subtypes P1.2, P1.1, and P1.10 in South Australia. In the Northern Territory all serogroup A isolates were 4:P1.10. Serogroup B and C strains showed considerable heterogeneity[24]. In south-western Sydney in 1990-94[25], serogroup C accounted for 60.8% (31 / 51) of invasive meningococcal isolates. Eighty per cent of these serogroup C isolates were serotype 2b and 70% subtype P1.2. Thus during this five year period the phenotype C:2b:P1.2 was the commonest sporadic isolate and was also associated with the cluster of cases described in 1991[20]. The phenotype C:2b:P1.2 has been involved in all the serogroup C outbreaks mentioned above (J. Jelfs, personal communication). Outbreaks many thousands of kilometres apart occurred in 1990-1991. Sporadic cases of the C:2b:P1.2 phenotype continued to occur in New South Wales over subsequent years, but have become less common, along with an overall decrease in serogroup C strains in New South Wales in 1994-95[26]. Molecular analysis of outbreak and sporadic strains of this phenotype is currently being undertaken to investigate whether one or more clones are involved.

Antibiotic susceptibility patterns

Many countries have noted the appearance of meningococcal isolates with altered penicillin susceptibilities in recent years. Laboratories of the National Neisseria Network have determined the antibiotic susceptibility of more than 450 invasive isolates, using standardised agar dilution techniques since 1994. Although 72.5% of isolates showed a decreased susceptibility to penicillin (minimal inhibitory concentration (MIC) $\leq$0.06mg / l), the data indicate that penicillin-based treatment regimes remain suitable for use in Australia. A study in Victoria of invasive strains isolated over six years from 1988 found little or no increase in resistance[27]. No beta lactamase-producing isolates have been detected in Australia.

Outcome of meningococcal infection

Factors influencing the outcome of meningococcal infection include the clinical syndrome on presentation, age, timing of commencement of antibiotic treatment and strain causing infection. In the study from south-western Sydney where clinical and laboratory criteria were used to classify patients as having predominantly meningitis (20.7%), meningitis / septicaemia (53.4%), or septicaemia (22.4%), the mortality rate in the meningitis group was 0, in the meningitis / septicaemia group it was 6.5% and in the septicaemia group it was 30.8%[25]. An attempt to prevent rapid bacterial multiplication is the rationale for the early use of parenteral penicillin to improve outcome[28]. This recommendation remains controversial, because there is a subset of patients with meningococcal disease who present with shock and rapidly developing multi-organ failure in whom there is a high mortality rate despite early appropriate antibiotic therapy. However, early recognition and treatment of meningococcal meningitis remains an important goal.

It is not common for Australian general practitioners to use parenteral antibiotics before transferring the patient to hospital. A delay in diagnosis and administration of appropriate intravenous therapy of more than two hours occurred in 36.2% of patients in the south-western Sydney study[25]. By comparison, there was a median delay to treatment in an Auckland, New Zealand, survey of 80 minutes[19]. There is room for improvement in the time taken to diagnose meningococcal infection and institute appropriate intravenous treatment. It is particularly important that intravenous antibiotics are not delayed while investigations such as lumbar puncture or CT scan are performed. The prior use of antibiotics does decrease the number of positive cerebrospinal fluid (CSF) cultures; however, there is very often other laboratory evidence of meningococcal disease[25]. Polymerase chain reaction (PCR) has also been used in blood and CSF to make a non-culture diagnosis of meningococcal disease[29].

Meningococcal vaccines

Meningococcal vaccines currently available in Australia are polysaccharide vaccines directed against serogroup A, C, W_{135} and Y strains. Polysaccharide vaccines are poorly immunogenic and unable to generate immunological memory. Thus response rates to the current serogroup vaccine are poor in young children, antibody titres are of short duration, and there is failure to respond to subsequent vaccination. Polysaccharides can be changed to T cell-dependent antigens by structural modification of the polysaccharide sialic acid polymer subunit so that it can be conjugated to a protein carrier such as tetanus toxoid, CRM (a nontoxigenic mutant diphtheria toxin), or a meningococcal OMP. Immunogenicity trials of these conjugate vaccines in infants and toddlers have shown high levels of bactericidal antibodies with good immunological memory. Similar modifications of the polysaccharide sialic acid polymer subunit of serogroup B organisms coupled with a protein carrier have produced a vaccine giving good levels of bactericidal antibodies and immunological memory[30].

The use of capsular polysaccharide vaccines is preferred because of the lack of strain variability in polymer subunits and the production of protective bactericidal antibodies post-vaccination. Another approach to vaccination, especially against serogroup B organisms whose capsular polysaccharide is poorly immunogenic, is the use of outer membrane protein antigen vaccines. Vaccine antigens may be prepared from outer membrane vesicles and combined with meningococcal capsular polysaccharide for improved solubility. Alternatively multivalent

vaccines against a number of class 1 OMPs can be prepared by insertion of OMP genes into meningococcal strains and subsequent purification of outer membrane vesicles. The limitation of these vaccines against group B meningococci is that the protection induced is serotype and subtype specific. Thus vaccines appropriate for one country or particular geographical area may not be suitable in another area.

The polysaccharide serogroup A, C, W_{135} and Y vaccines formerly have been used successfully to control large outbreaks of serogroup A disease in Australia and for control of some of the smaller outbreaks and clusters of serogroup C disease[7,20,21,23]. It is doubtful whether there would ever be a case for universal vaccination against meningococcal disease in Australia with the newer, more effective vaccines. The overall incidence of meningococcal disease of less than 2 per 100,000 places Australia in the category of a low-incidence country. However, attack rates in Aboriginal communities in central and northern Australia are much higher, approaching the incidence of *Haemophilus influenzae* in Aboriginal children less than five years of age before mass vaccination programs began. In this group, universal vaccination with new meningococcal vaccines effective against serogroups A, B, C, W_{135} and Y would be very worthwhile.

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