The Changing Epidemiology of *Streptococcus pneumoniae* Serotype 19A Clonal Complexes

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(See the article by Beall et al, on pages 1360–8.)

*Streptococcus pneumoniae* is a leading cause of pneumonia throughout the world and a major cause of serious invasive disease, especially in the very young and the elderly. The major virulence factor of *S. pneumoniae* is its polysaccharide capsule, which can be expressed as 91 different capsular serotypes [1, 2]. Seven serotypes (those of high prevalence in children prior to 2000; serotype [sero]4, sero6B, sero9V, sero14, sero18C, sero19F and sero23F) constitute a very effective protein conjugate vaccine, PCV7, which has been in use throughout the world, including the United States, since 2000. While the vaccine serotypes contained in PCV7 have declined in prevalence over the last decade, other serotypes, most notably sero19A, have become more prevalent throughout North America and Europe [3–8]. To add cause for concern, many of the sero19A strains circulating are also resistant to multiple antimicrobial agents, making them difficult to treat. Six years after the introduction of PCV7, sero19A is the most common invasive pneumococcal serotype in the United States, with a substantial proportion of its strains being antimicrobial resistant [9].

Although the serotype designation is important for understanding pneumococcal epidemiology, to gain a deeper perspective of the pneumococci circulating globally, a technique termed multilocus sequence typing (MLST) is used. MLST is a nucleotide sequence–based approach to characterizing bacterial strains [10]. It involves the DNA sequencing of internal fragments of 7 housekeeping genes from the pneumococcal genome. The sequences are compared with previously identified sequences (alleles) and assigned an allele number for each of the 7 loci. The 7 number combinations denote an allelic profile of the strain that is then assigned a sequence type (ST) designation. Sequence types are grouped into “clonal complexes.” To be part of clonal complex, members must have typically 5 or 6 loci that match. Interestingly and importantly, isolates with different serotypes can be members of the same clonal complex. Detailed information regarding *S. pneumoniae* MLST analysis is provided on the pneumococcal MLST website, hosted by Imperial College, London (http://spneumoniae.mlst.net/). The information provided by MLST is global in scope but more in-depth than the serotype designation.

In this issue of the *Journal*, Beall and colleagues describe the recent epidemiology of *S. pneumoniae* sero19A in the United States from isolates collected through the Centers for Disease Control and Prevention (CDC) Active Bacterial Core (ABC) surveillance program. Their detailed MLST analysis of sero19A isolates has shown that while the prevalence of sero19A strains seems to have stabilized since 2005, the numbers of isolates within specific sero19A clonal complexes have fluctuated significantly. These changes have resulted in declines in clonal complex prevalence, increases in clonal complex prevalence, or appearance of new sero19A clonal complexes. Currently, 3 main clonal complexes dominate the invasive pneumococcal disease sero19A landscape in the United States. These have been designated clonal complex (CC)19919A, CC320/27119A, and CC69519A. These 3 clonal complexes collectively comprise approximately 87% of the sero19A isolates collected from 2005–2007 [11]. Brief descriptions of each of these clonal complexes with respect to prevalence over recent years is given below.

**CC19919A**

Prior to 1999 and the introduction of PCV7, CC19919A strains were the most...
prevalent sero19A strains detected in the United States, accounting for 77% of the sero19A collected in the ABC’s surveillance program [12–14]. This pattern continued during the first few years of PCV7 use. CC199\textsuperscript{19A} was the predominant sero19A in children aged \textless 5 years in specimens collected in 2003–2004 by the ABC’s group, accounting for 72% of the sero19A isolates collected in this survey. Based on pre- and post-PCV7 CC199 numbers, it would have been realistic to predict that CC199\textsuperscript{19A} would continue to be the predominate sero19A clonal complex in circulation. The surveillance data presented by Beall et al, however, show this not to be the case. They show that cases of invasive pneumococcal disease caused by CC199\textsuperscript{19A} appear to be gradually declining. In 2005, CC199\textsuperscript{19A} accounted for 59% of the sero19A isolates detected dropping slightly to 57% in 2006 and declining further to 40% in 2007 [11]. A decline from a high of 77% prior to 1999 to a low of 40% in 2007 indicates changes are occurring with respect to sero19A epidemiology. Interestingly, most CC199\textsuperscript{19A} strains collected in the United States are intermediately resistant to penicillin, with minimum inhibitory concentrations (MICs) of .12–1 \(\mu\)g/mL [9, 13]. Even with this phenotype, CC199\textsuperscript{19A} strains are in decline. 

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\textbf{CC320/271\textsuperscript{19A}}

In contrast to CC199\textsuperscript{19A}, strains of CC320/271\textsuperscript{19A} were not detected by the ABC’s surveillance program in 1999. It was not until a follow-up ABC survey in 2003–2004 that investigators noted CC320/271\textsuperscript{19A} accounted for 13.4% of the sero19A isolates collected from cases of invasive pneumococcal disease in the United States. As noted previously, different serotypes can occur within the same clonal complex. CC320/271 isolates within the pneumococcal MLST database from the pre-PCV7 era (prior to 2000) circulated as sero19F rather than sero19A. MLST analysis suggests that a capsule-switching event may have occurred, involving ST236\textsuperscript{19F} (a serotype in PCV7) and a sero19A strain, which gave rise to CC320/271\textsuperscript{19A} subsequent to PCV7 introduction [9, 15]. The work by Beall and colleagues found CC320/271\textsuperscript{19A} continued to climb in prevalence in the United States, attaining a 21% detection rate in 2005, 23% in 2006, and 33% in 2007 [11]. With respect to antimicrobial resistance, unlike most CC199\textsuperscript{19A} isolates, 98% of CC320/271\textsuperscript{19A} isolates recovered in the 2005–2007 survey period were fully penicillin resistant (MIC \(\geq 2 \mu\)g/mL), as well as resistant to erythromycin and clindamycin, suggesting the possibility that high-level antimicrobial resistance may be playing a role in driving this increase.

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\textbf{CC695\textsuperscript{19A}}

The pneumococcal strains from CC695\textsuperscript{19A} are considered moderately antimicrobial-resistant vaccine-escape strains. Prior to 2003–2004, CC695\textsuperscript{19A} isolates were unknown. The first 5 CC695\textsuperscript{19A} isolates were identified in 2003–2004, 3 years after the introduction of PCV7 [16], and have continued to increase in numbers. This clonal complex comprised 7.5% of the sero19A strains in 2005, 9.0% in 2006, and 13.9% in 2007 in the United States [11]. Previous evidence indicated that CC695\textsuperscript{19A} arose through a serotype switch event involving a ST695\textsuperscript{4} and a ST199\textsuperscript{19A}. A ST199\textsuperscript{19A} donated the \textit{pbp2x-cps} locus-\textit{pbp1a} to a ST695\textsuperscript{4} recipient, giving rise to a moderately antibiotic resistant sero19A strain in a sero4 background (penicillin MIC of .06–.2 \(\mu\)g/mL).

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\textbf{OTHER SERO19A CLONAL COMPLEXES}

Whereas CC199, CC320/271, and CC695 comprise most sero19A strains circulating in the United States, other clonal complex variants are present, and of these, many are first documented in the article by Beall et al [11]. What is perhaps most interesting about these new sero19A sequence types is that MLST showed many of them were documented in serotypes other than sero19A. As Beall and coauthors suggest, these data point to a number of serotype switching events having occurred within the last few years. Considering the plasticity of the pneumococcal genome, this change may be not that unusual, and we should expect to find more of these variants in subsequent surveys.

As mentioned, the proportion of invasive pneumococcal disease caused by sero19A strains seems to have stabilized in the population surveyed by the ABC program. However, the proportions of the different clonal complexes that comprise the circulating sero19A in the United States have not remained stable.

The reasons for the changes in the epidemiology of the CC199\textsuperscript{19A}, CC320/271\textsuperscript{19A}, and CC695\textsuperscript{19A} strains over the last decade are probably multifactorial. The implementation of the 7 valent conjugate vaccine program in 2000 has no doubt put immunological pressure on the 7 serotypes contained in this vaccine, leading to their rapid decline. Removal of these vaccine serotypes has possibly provided niches for other serotypes to occupy, such as those that make up the sero19A clonal complexes. Antimicrobial nonsusceptibility also likely plays a role, as antimicrobial-resistant clonal complexes are increasing in prevalence in each year surveyed. Whether these factors are the only drivers for clonal complex sero19A fluctuations is not clear now.

The surveillance program of the ABC group in the United States, coupled with the detailed MLST laboratory analysis of pneumococcal isolates, allows us to identify and track pneumococcal strains that have increased in prevalence following PCV7 introduction. Although sero19A strains now cause a substantial amount of invasive pneumococcal disease and are becoming increasingly resistant to antimicrobials, making these isolates more difficult to treat, a new tool has recently become available to prevent
these events from occurring. A protein-conjugate vaccine targeting 13 pneumococcal serotypes (PCV13; PCV7 serotypes plus 1, 3, 5, 6A, 7F, and 19A) will replace PCV7 and should prove as effective as the current conjugate vaccine. With the introduction of the PCV13 vaccine, invasive pneumococcal disease and the antibiotic resistance associated with the sero19A should decline to levels currently seen with the PCV7 serotypes, especially in children for whom the PCV13 vaccine will be targeted. However, as evidenced by the work described by Beall et al and by others, there is a possibility that S. pneumoniae will simply switch its serotype to escape the effects of the PCV13 vaccine. Should this switch occur (and based on evidence in the past decade, it most likely will), we will need to focus efforts on targets other than the capsule as potential pneumococcal vaccine candidates.

References