Bacterial Otitis Media, the Chinchilla Middle Ear, and Biofilms

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(See the article by Reid et al., on pages 786–94.)

Otitis media in children has a significant impact on health care and health-care economics [1]. It is the number one reason children are seen in physicians’ offices and the most common reason children are given antibiotics [2]. The economic impact was estimated in 1998 to be between $3 billion and $5 billion per year in the United States. This estimate does not take into consideration the indirect costs associated with lost time at work for the parents of children with chronic and recurrent middle ear infections. In addition, the widespread use of antibiotics for children treated for otitis media is believed to play an important role in the marked increase in the extent of antibiotic resistance as well as the increasing number of resistant strains isolated worldwide. The importance of understanding the pathogenesis of otitis media, the host adaptive and innate immune responses, and the mechanisms for vetting reasonable vaccine candidates is great. Central to these goals is an animal model that mimics human disease.

The chinchilla inner and middle ear are uniquely suited to serve as a relevant model for human otological diseases. The inner ear has been used by audiologists and otolaryngologists since 1970 for studies on hearing loss [3]. Giebink et al. demonstrated over 30 years ago that the chinchilla middle ear could be used as a model for pneumococcal otitis media [4]. Since that time, Haemophilus influenzae and Moraxella catarrhalis have been shown to be capable of inducing bacterial otitis media in the chinchilla [5, 6]. The model bears many similarities to human disease and can be induced by intranasal instillation of bacteria after inoculation with a respiratory virus such as influenza or adenovirus. In 2001, Post showed that both chinchillas and children with otitis media had evidence of biofilm formation on middle ear epithelial surfaces, in their middle ear secretions, and on tympanostomy tubes [7].

In the article by Reid et al. in this issue of the Journal [8], the formation of biofilms by pneumococci in a chinchilla middle ear model is clearly demonstrated. The authors propose that the existence of the biofilms leads to bacterial persistence. Using live-dead staining, the authors have shown that viable bacteria are present in the entire biofilm and have confirmed pneumococcal identity by staining with labeled antibodies for 4 different well-defined pneumococcal proteins. There was a considerable amount of neutrophil infiltration in this biofilm. It appears that neutrophil extracellular traps (NET) are formed in the biofilm in otitis media. These NET are made by activated neutrophils and have been characterized as bacterial traps [9]. They are formed by the release of granule proteins and chymotrypsin to form extracellular fibers 15–17 nm in thickness alternating with globular domains ~25 nm in size. Immunochromical analysis has shown that these fibers contain proteins from the azurophilic granules such as neutrophil elastase, cathepsin G, and myeloperoxidase [9]. This observation in the chinchilla is important in relation to the innate immune mechanisms that operate to clear this infection. Much additional work is necessary to understand the requirements for pneumococcal biofilm formation, including the structural features and composition of the biofilm and the role of quorum sensing in biofilm evolution.

It is clear to all who have worked with the chinchilla model of otitis media that it replicates almost all aspects of human disease. To exploit this model fully, a number of important resources are needed. Most importantly, the genome of this animal should be sequenced. Currently, studies into the chinchilla innate (or acquired) immune response to otitis media rely on seeking proteins homologous to human or murine receptors or effectors.
so as to use commercially available reagents.

The implications of biofilm formation by organisms that colonize and infect humans have undergone considerable discussion in the scientific community. The studies of Costerton et al. in the 1980s led the way to the realization that bacterial biofilms could form on the inert surfaces of indwelling urinary and vascular catheters [10, 11]. This view was at the time very controversial, but it is taken for granted today. The studies of Pearson et al. demonstrated that quorum sensing was involved in biofilm formation [12, 13] by the human opportunistic pathogen *Pseudomonas aeruginosa*. Since that time, the importance of biofilm formation as a component of human disease and colonization has evolved to include an increasing list of microbes and now includes the concept that biofilms can form on biological surfaces, such as the airway of patients with cystic fibrosis or the middle ear of a young child. The concept that organisms in biofilms are significantly more resistant to antimicrobial therapy than the same organisms under standard culture conditions has implications for the management of infections, emerging antimicrobial resistance, and the role of biofilm formation in disease outcome and recurrence. The chinchilla model of otitis can serve as an excellent tool to study the contribution of biofilms to the evolution of antimicrobial resistance in pneumococcal and nontypeable *H. influenzae* infections.

Many factors influence the frequency and severity of otitis media in children including genetics, age, anatomy, and environment. The goal of limiting or eliminating this childhood infection by vaccination or enhancement of innate immune responses should be a priority for future biomedical research funding.

References