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Polysaccharide-Based Conjugate Vaccines for Enteric Bacterial Infections: Typhoid Fever, Nontyphoidal Salmonellosis, and *Escherichia coli* O157:H7

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INTRODUCTION

Bacterial surface polysaccharides, either as capsules or as O-specific polysaccharides (O-SPs) expressed by gram-negative organisms, are common virulence factors and essential protective antigens. It is hypothesized that one of the ways that the licensed purified polysaccharide vaccines (e.g., pneumococcal and *Salmonella typhi* Vi) and conjugate vaccines [e.g., *Haemophilus influenzae* type b (Hib), pneumococcal, and meningococcal] function is by eliciting a critical level of serum IgG that ''exudes'' onto the mucosal sites and lyses pathogens upon contact (1). Polysaccharide vaccines are safe and efficacious, and can be administered in multivalent form without interference. For example, the pneumococcal polysaccharide vaccine contains 23 types and meningococcal vaccine contains 4 (2,3). Also proven in field trials, conjugate vaccine-induced anti-polysaccharide antibodies inhibit colonization by the target bacteria, thereby resulting in ''herd'' immunity (4).

Purified polysaccharide vaccines are T-independent antigens that do not induce booster response on reinjection, the duration of immunity is short (3–5 years), and they are notoriously poor immunogens for young children. Hib conjugate vaccines showed that the immunogenicity of polysaccharide antigens can be significantly improved by linking the capsular polysaccharide to a carrier protein (5,6). This same principle was applied successfully in the development and licensure of pneumococcal and meningococcal conjugate vaccines (7–9). This approach has also been successfully applied to antigens from enteric pathogens such as Vi polysaccharide, the capsular polysaccharide of *S. typhi*; a Vi conjugate vaccine was highly immunogenic and protective in young children in an area of high typhoid endemicity (10).

For gram-negative pathogens that lack a capsule, the O-SP of the outer membrane serves as a virulence and protective antigen. Experimental and clinical data show that immunity in humans following disease is specific to O-SP of *Shigella*, *Escherichia coli*, and *Vibrio cholerae* (11,12). For nontyphoidal *Salmonella*, mice can be protected from *Salmonella typhimurium* infection by passive immunization with a monoclonal IgG directed against O-SP (13). Unlike capsular polysaccharides, the O-SPs are normally poorly immunogenic, probably because of their small size; O-SPs need to be conjugated with carrier proteins to enhance their immunogenicity. Our laboratory has applied this technique to several enteric pathogens including nontyphoidal *Salmonella*, *Shigella*, *V. cholerae*, and *E. coli* O157. In this chapter, *S. typhi*, nontyphoidal *Salmonella*, and *E. coli* O157 vaccine developments will be reviewed.

TYPHOID FEVER

Typhoid fever, which is transmitted to susceptible hosts by food and water contaminated with *S. typhi*, remains a major health problem in developing countries. Since the 1990s, the emergence of antibiotic resistant strains has made the treatment of typhoid fever more difficult (14). Asymptomatic carriers constitute the reservoir of infection (15). With no near-term solution to providing clean water and environment to all populations in developing countries, vaccination is a cost-effective way to control and eliminate this disease (16–18).

Typhoid fever in young children is often unrecognized because of atypical clinical symptoms and difficulties in the volume of blood that can be drawn for culture. Recently, using active surveillance methods a high incidence of typhoid fever was shown in young children in densely populated urban slums in South Asia (19). In some surveys in developing countries, children two to four years old had the highest attack rate in both community- and hospital-based surveys (18,19). Currently, there is no licensed typhoid vaccine suitable for this age group.

There are three licensed typhoid vaccines, each having limitations. The killed whole-cell parenteral vaccine provides ~65% protection but strong adverse reactions limit its usefulness. In the late 1980s and early 1990s, two new and safer vaccines were licensed: the live, attenuated oral vaccine Ty21a and the parenteral administered Vi polysaccharide vaccine. There are major differences between the two vaccines. Vi has consistently provided ~65% to 70% efficacy in field trials in highly endemic areas, with protection lasting at least three years (20–22). Ty21a confers a similar level of protection for up to seven years (23–25). Vi immunization consists of a single injection, whereas Ty21a requires three doses. Vi polysaccharide is poorly immunogenic in infants and data for protection do not exist for Ty21a for children less than three years (26). The immunogenicity of Vi vaccine has been substantially improved by conjugation with carrier proteins (10).