Acellular Pertussis Vaccines and Pertussis Resurgence: Revise or Replace?

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ABSTRACT The resurgence of pertussis (whooping cough) in countries with high vaccination coverage is alarming and invites reconsideration of the use of current acellular pertussis (aP) vaccines, which have largely replaced the old, reactogenic, whole-cell pertussis (wP) vaccine. Some drawbacks of these vaccines in terms of limited antigenic composition and early waning of antibody levels could be anticipated by the results of in-trial or postlicensure human investigations of B- and T-cell responses in aP versus wP vaccine recipients or unvaccinated, infected children. Recent data in experimental models, including primates, suggest that generation of vaccines capable of a potent, though regulated, stimulation of innate immunity driving effective, persistent adaptive immune responses against *Bordetella pertussis* infection should be privileged. Adjuvants that skew Th1/Th17 responses or new wP (detoxified or attenuated) vaccines should be explored. Nonetheless, the high merits of the current aP vaccines in persuading people to resume vaccination against pertussis should not be forgotten.

PERTUSSIS RESURGENCE AND FAILURE OF ACELLULAR PERTUSSIS VACCINES

Pertussis (whooping cough) continues to be a relevant public health problem. Of particular concern, the incidence of pertussis has seen a brisk acceleration in recent years, with disease outbreaks in countries with high vaccination coverage. In Australia, the number of reported new cases has increased more than 100 times from 1991 to 2011 (1). In the Californian epidemic of 2010, over 9,000 cases were recorded, the highest number in 60 years (2), and in the 2012 epidemic in Washington, the rate of disease exceeded that in the California epidemic (3). In the United Kingdom, there were 10 deaths in infants under 12 months old in 2012, the highest rate of mortality from pertussis since 1982 (4). No doubt, there is a resurgence of pertussis despite vaccination.

Since the early 1990s, acellular pertussis (aP) vaccines have largely replaced the old inactivated whole-cell pertussis (wP) vaccine, which was efficacious but so reactogenic as to bring about a rather dramatic drop of the vaccination coverage in most industrialized countries (3, 5).

Replacing the wP vaccine with the aP vaccines has resulted in an increase in pertussis vaccination coverage in newborns and infants, although some gaps of coverage may remain. Overall, the resurgence of pertussis in the countries mentioned above means some failure of aP vaccines to protect against the disease.

It is not easy to explain the reason for this failure. In a recent expert working group meeting on pertussis (6), several potential factors, possibly cooperatively acting, were identified. These factors include genetic changes in circulating *Bordetella pertussis* (1, 7), as well as increased recognition and reporting of pertussis by the application of new, more sensitive, laboratory diagnostic tests (8, 9). Nonetheless, age-related waning of protective immunity conferred by the aP vaccines has emerged as a major contributing factor (5, 10–12).

In this editorial, we will focus on composition of, and immune responses to, aP and wP vaccines, since a better knowledge of these factors may provide some clues about both aP vaccine failure and potential remedies, e.g., whether we need to revise composition and usage of the current vaccines or simply replace them with new ones.

Immunity persistence in pertussis vaccine recipients. In dealing with immunity persistence in aP vaccine recipients, it would be wise to acknowledge that immunity waning at a certain distance from vaccination is a "normal" phenomenon that is seen with all vaccines, hence the need of boosters. This seems to be of particular relevance for pertussis vaccines, since pertussis, at variance with some other bacterial and viral diseases, does not leave a persistent protective immunity (3). The problem here is that a "long" persistence of protective immunity, and the consequent delay in the time of immunity waning in children immunized with aP vaccines, was likely overestimated from the data of aP vaccine trials and follow-up studies (5, 13). A few observations regarding both aP vaccine composition and immune responses in aP vaccine recipients, compared to those of wP recipients, suggest that some early waning of protective immunity in the former might have been anticipated (14).

The composition issue. The varied antigenic formulation of aP vaccines approved for human use is a distinctive and unusual feature with respect to all other bacterial vaccines for human use. In fact, aP vaccines formulated with two (pertussis toxin [PT] and filamentous hemagglutinin [FHA]) or five (PT, FHA, pertactin, and fimbriae 1 and 2) immunizing antigens have been licensed for human use (5, 13). In principle, vaccines with such different antigenic formulations should not be expected to confer equal levels of efficacy and long-term protection.

Pathogenesis of *Bordetella pertussis* is due to the expression of multiple virulence traits that are poorly recognized by the immune system during natural infection, hence conferring to the bacterium a high degree of immune escape (15, 16). Unless unknown compensatory mechanisms among the various antigens are admitted, it is expected that the aP vaccines with three or more

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antigens are not only more protective than those with fewer components for a short period postvaccination but are also less sensitive to a rapid decay of the protective immunity. This difference among the aP vaccines would hardly be seen during time- and population-limited efficacy trials (5).

There are very few investigations on possible differences in time or degree of waning of protection from pertussis in children who receive different aP vaccines. More precisely, is the immunity similarly waning in recipients of 5- and 2-component aP vaccines? If the FHA component of aP vaccines does not significantly contribute to vaccine efficacy (13, 17), then some children have been immunized with one protective antigen and other children have been immunized with four protective antigens. Is this divergence really irrelevant to long-term aP vaccine efficacy? Evidence suggests that it makes a difference. In fact, 5 years after primary vaccination with 2- or 3-component aP vaccines, the children immunized with the 3-component vaccine showed a trend to a longer persistence of anti-PT IgG after the preschool boost compared with the recipients of the 2-component vaccine (18). In this line, approval by the regulatory authorities of vaccines with such a different antigenic composition for preventing the same disease remains somewhat surprising.

Immunological issues. That protection from pertussis could not last long while immunity mediated by *B. pertussis* virulence-neutralizing antibodies was rapidly waning could be inferred from studies by ourselves and others, some of which were performed during clinical trials of aP vaccines or soon after their approval (5, 19). These studies suggested that equally high, long-term protective immunity could not follow from vaccines such as the wP and aP vaccines that had manifestly quite different ways of inducing T-cell memory responses and the consequent help for antibody production (20–22). In addition, protection conferred by a wP vaccine was much closer to that which follows from natural infection than the protection conferred by aP vaccines. Immune responses in aP vaccine recipients could change from a mixed Th2/Th1 profile to a robust Th1 profile following a natural booster (12, 14, 23–25). Curiously enough, if long-term protection by aP vaccines depends on natural boosters, those aP vaccines which are more effective in retarding the circulation of *B. pertussis* could be less efficacious in the long run.

However, the problem of aP vaccines is not limited to only waning immunity, which could be at least mitigated by vaccine boosters (26). In a recent study with a baboon model, the authors demonstrated that current aP vaccines fail to prevent *B. pertussis* colonization and transmission (27). Baboons vaccinated with aP vaccine were protected from severe, pertussis-associated symptoms, but not from colonization, and were unable to arrest the transmission of *B. pertussis* to unvaccinated contacts. Vaccination with wP vaccine induced a more rapid *B. pertussis* clearance compared with naive and aP-vaccinated animals. Although all vaccinated and infected animals had robust serum antibody responses, the key difference was in T-cell immunity. Infected and wP-vaccinated animals showed strong *B. pertussis*-specific Th17 and Th1 memory responses, whereas aP vaccination induced a Th1/Th2 response. The importance of Th17 in the immune protection in pertussis was also demonstrated in mouse and *ex vivo* human models (28, 29).

The observation that aP vaccines induce an immune response that fails to prevent colonization and transmission adds to the waning of antipertussis protective immunity in providing a plausible explanation for the resurgence of pertussis. Thus, optimal control of this disease may require the development of better vaccines. This could be done by either modifying the composition of the existing subunit vaccines or by generating new wP vaccines, inactivated or attenuated.

Revise the existing subunit vaccines. As suggested by several researchers (11, 30), it would be possible to improve aP vaccines by replacing the alum with other adjuvants, particularly those enabling potent Th1 and Th17 responses (11). Possible candidates are Toll-like receptor agonists. This vaccine formulation would induce a persistent immunological memory and hence be more effective than current aP vaccines. New subunit vaccines could be generated by adding new putative protective antigens to the existing antigen formulations. Possible candidates include the adenylate cyclase toxin, the autotransporter BrkA, and an antigen induced by iron starvation, named IRP1-3 (30).

A new vaccine made up of a conjugate of the core oligosaccharide of *Bordetella bronchiseptica* with bovine serum albumin (BSA) and shown to raise bactericidal antibodies against *B. pertussis* has been proposed by Robbins and collaborators (31). The carrier protein in the final vaccine formulation is planned to be genetically altered PT (31).

New whole-cell vaccines. For the multiple virulence traits possessed by *B. pertussis* and the ascertained differences in the types of immune responses between aP and wP vaccines (as discussed above), the generation of new wP vaccines appears to be a promising alternative for the replacement of current aP vaccines. New, improved wP vaccines are expected to stimulate a high level of innate immunity that is instrumental in driving the generation of persistently protective Th1/Th17-skewed adaptive immunity. The rich cytokine milieu generated by a strong and regulated activation of antigen-presenting cells and other immune effectors such as monocytes and natural killer cells may be the critical factor for a sustained expansion of antigen-specific T and B memory cells. Of interest in this context, the recent report by Wu et al. (32) that DTwP (diphtheria, tetanus, and whole pertussis) vaccines are better than DTaP vaccines in maintaining high anti-DT antibody titers. Subunit vaccines, whatsoever their complexity, may be less suitable to activate high levels of innate immunity than whole-cell vaccines with their rich repertoire of immunomodulatory constituents.

A return to the use of the old wP vaccine after making it less reactogenic has been suggested (30). However, it is not known how to make that vaccine less reactogenic without losing some efficacy. Particularly extensive investigations have been made with a recently developed live attenuated nasal *B. pertussis* vaccine, BPZE1. Similar to natural infection, nasally applied BPZE1 induces both mucosal and systemic immune responses, which result in faster and broader immunity, compared to parenteral vaccine administration. In preclinical mouse studies, BPZE1 induced long-lasting protection, significantly higher than aP vaccine (33), and BPZE1 vaccination induced rapid protection compared to aP vaccines (30). In a human preclinical model, BPZE1 promotes human dendritic cell CCL21-induced migration and, in agreement with the data in baboons, it drives a Th1/Th17 response (29). The vaccine was tested in a placebo-controlled, double-blind, dose-escalating safety trial that ended in 2010 (registered at Clinicaltrials.gov under registration no. NCT01188512) that indicated that the BPZE1 vaccine is safe in healthy adults and able to transiently colonize the nasopharynx. It induces immune re-

sponses in all colonized individuals (34). Such a vaccine can be an excellent carrier for antigenic determinants of other infectious pathogens that affect early childhood, such as respiratory syncytial virus (30). Concern for BPZE1 vaccine administration in newborns should not preclude further investigations and its use as a booster dose in children and adults. Overall, wP and aP vaccine combination schedules could be usefully exploited.

CONCLUSION: FAILURES VERSUS MERITS

It is clear that a revision of current aP vaccine usage, improvements in aP vaccines (such as adjuvants and antigenic formulation), reintroduction of a less reactogenic wP vaccine, or the replacement of current pertussis vaccines with totally new ones are all possible options to enhance our capacity to fight pertussis. This should not lead us to forget the high merits of the current aP vaccines. While they have seemingly not met the initial expectations of long-term protective immunity, nonetheless, their high efficacy against severe pertussis in infants and their recognized low reactogenicity levels have reconciled people with vaccination against pertussis in countries where this vaccination had been largely abandoned because of the adverse events of wP vaccines.

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