

## Adaptation of *Bordetella pertussis* to Vaccination: A Cause for Its Reemergence?

Frits R. Mooi, Inge H. M. van Loo, and Audrey J. King

National Institute for Public Health and the Environment, Bilthoven, the Netherlands

In the Netherlands, as in many other western countries, pertussis vaccines have been used extensively for more than 40 years. Therefore, it is conceivable that vaccine-induced immunity has affected the evolution of *Bordetella pertussis*. Consistent with this notion, pertussis has reemerged in the Netherlands, despite high vaccination coverage. Further, a notable change in the population structure of *B. pertussis* was observed in the Netherlands subsequent to the introduction of vaccination in the 1950s. Finally, we observed antigenic divergence between clinical isolates and vaccine strains, in particular with respect to the surface-associated proteins pertactin and pertussis toxin. Adaptation may have allowed *B. pertussis* to remain endemic despite widespread vaccination and may have contributed to the reemergence of pertussis in the Netherlands.

*Bordetella pertussis* and *Bordetella parapertussis* are the etiologic agents of whooping cough or pertussis, a respiratory disease that is most severe in infants and young children. Compared to *B. pertussis*, *B. parapertussis* is isolated less frequently from pertussis patients (1% to 5% of pertussis patients in The Netherlands) and generally causes less severe symptoms. Pertussis is highly contagious, and, in the prevaccination era, nearly every child contracted this disease. The clinical course of pertussis is characterized by paroxysms, or bursts, of numerous, rapid coughs followed by a long inspiratory effort, which may be accompanied by a characteristic high-pitched whoop (hence the designation whooping cough). During such an attack, patients may turn blue due to lack of oxygen. In serious cases, this oxygen deprivation may lead to brain damage. The most common complication of pertussis, however, is secondary pneumonia. Young infants are at highest risk for pertussis-associated complications. More than 50% of infants less than 6 months old who contract pertussis require hospitalization. Treatment of pertussis is primarily supportive, and adequate control of the disease depends on effective immunization. Before vaccination was introduced in the 1950s, pertussis was a major cause of infant death throughout the world (1). Widespread vaccination of young children has been successful in controlling the disease (1).

### Vaccination

The high rate of illness and death caused by pertussis stimulated the early development of vaccines composed of whole, killed bacteria. These whole-cell vaccines were introduced in many countries in the 1950s and 1960s and have been highly successful in reducing the incidence of pertussis (1). The desire to avoid the side effects of whole-cell vaccines has stimulated the development of less reactogenic, acellular, vaccines composed of purified *B. pertussis* proteins (2). Acellular vaccines are replacing whole-cell vaccines in many countries.

Address for correspondence: Frits R. Mooi, Research Laboratory for Infectious Diseases (LIO), National Institute of Public Health and the Environment, P.O. Box 1, 3720 BA, Bilthoven, The Netherlands; fax: 31-30-274.4449; e-mail: fr.mooi@rivm.nl

Despite vaccination, pertussis is an endemic disease. Various sero-epidemiologic studies have shown that the frequency of infection may be as high as 1% to 4% (de Melker and Schellekens, pers. comm.) (3). Further up to 30% of people with a persistent cough were found to have been infected with *B. pertussis* (4). It is possible that vaccination initially reduced the circulation of *B. pertussis* and that adaptation allowed the *B. pertussis* population to restore its high circulation rate. This assumption predicts a change in the makeup of the *B. pertussis* population after the introduction of vaccination, a phenomenon that has indeed been observed in The Netherlands.

### Changes in *B. pertussis* in a Highly Vaccinated Population

In some countries with highly vaccinated populations such as Australia (5), Canada (6), and The Netherlands (7) (Figure 1), pertussis has reemerged. Such a phenomenon may have been caused by changes in the accuracy of notifications, decreases in vaccine coverage, or changes in vaccine quality. These possibilities have been excluded for The Netherlands (7), and we have proposed another possible cause: adaptation of *B. pertussis*

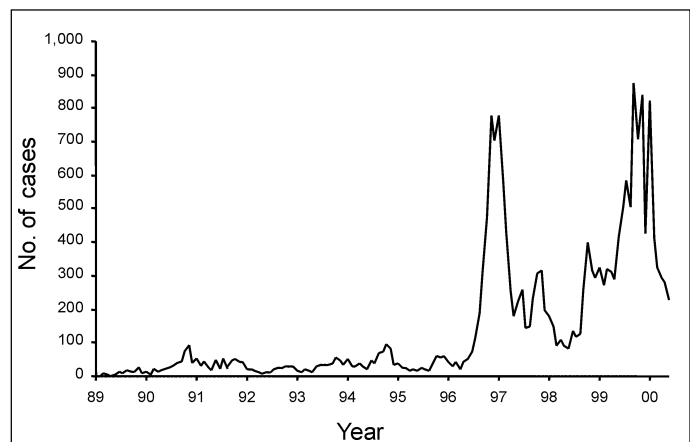


Figure 1. Notifications of pertussis in the Netherlands. Source: National Institute of Public Health and the Environment



countries with a long history of pertussis vaccination, such as Finland and the United States (17,18), and also in Italy, where vaccine coverage has varied considerably.

### Discussion

Is polymorphism in pertactin and pertussis toxin driven by host immunity, or is it the result of random fixation due to genetic drift? The latter possibility is highly unlikely since essentially all DNA mutations we detected in the pertactin and pertussis toxin genes were nonconservative. In contrast, random genetic drift is characterized by a high degree of conservative mutations in protein coding regions (20). Further, the polymorphic regions interact directly with the immune system. The polymorphic region of pertactin induces a protective immune response (unpublished data). One of the polymorphic residues in PtxS1 has been implicated in binding to the T-cell receptor (14). Finally, the fact that the same temporal trends in allele frequencies are observed in geographically distinct regions such as Finland, the United States, and The Netherlands argues against random genetic drift. It is possible that the polymorphic loci we have identified are linked to other, as yet unknown, polymorphic loci that increase fitness of strains in vaccinated populations (hitchhiking).

Strains carrying nonvaccine-type pertactin or pertussis toxin variants were not found in the prevaccination era. Although the number of strains analyzed from this period was limited, these data suggest that the nonvaccine-type variants are not able to displace the vaccine-type strains in unvaccinated populations (i.e., they have a lower fitness level, or reproductive rate, in unvaccinated communities). Alternatively, the nonvaccine-type strains may have evolved relatively recently. Consistent with the first hypothesis, we have observed that nonvaccine-type strains are less fit in naive mice than vaccine-type strains. In immune mice the difference in fitness between the two types of strains was much less pronounced (unpublished data). Thus vaccination has acted to shift the competitive balance between strains.

An important question to address is whether adaptation of the *B. pertussis* population has affected vaccine efficacy, i.e., contributed to the reemergence of *B. pertussis*. Animal experiments have indicated that variation in pertactin affects vaccine efficacy (unpublished data). Further, we found vaccine-type pertactin variants less frequently among vaccinated persons than among unvaccinated persons, which would be expected if the vaccine protects differentially against strains with distinct pertactin types (8). However, the extent to which polymorphism affects vaccine efficacy is probably dependent on the vaccine used. It is conceivable that the increase in fitness associated with nonvaccine types of pertactin and pertussis toxin in vaccinated populations is substantial enough to drive expansion of strains carrying these protein variants but that the effect is too small to result in a measurable drop in vaccine efficacy. Further studies are required to assess the effect of the observed adaptations on the efficacy of pertussis vaccines. In this period, when whole-cell vaccines are being replaced by acellular vaccines in many countries, continued strain surveillance is of paramount importance.

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Dr. Mooi is at the National Institute of Public Health and the Environment, The Netherlands, and the Eijkman-Winkler Laboratory of the University of Utrecht. His current interests are molecular epidemiology and evolution of *Bordetella* spp.

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