Bordetella pertussis
Epidemiology and diagnostic tools in Belgium

Denis Piérard
**SYMPTOMS**
- Rhinorrhea, sneezing, +/- fever
- Cough
- Frequency, severity
- Distinctive paroxysms, post-tussive emesis, whoop

**LABORATORY FINDINGS**
- Lymphocytosis
- Chest radiograph

**DIAGNOSTIC TESTS**
- PCR
- Culture
- DFA
- Serology

<table>
<thead>
<tr>
<th></th>
<th>CATARRHAL STAGE</th>
<th>PAROXYSMAL STAGE</th>
<th>CONVALESCENT STAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apnea, bradycardia</td>
<td>+++</td>
<td>+++</td>
<td>+</td>
</tr>
<tr>
<td>Seizures, encephalopathy</td>
<td>++</td>
<td>+++</td>
<td>+</td>
</tr>
<tr>
<td>2° Pneumonia, atelectasis</td>
<td>+++</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Malnutrition, apathy</td>
<td>++</td>
<td>+++</td>
<td>+</td>
</tr>
</tbody>
</table>

© 2008 Elsevier Inc. Long et al: Principles and Practice of Pediatric Infectious Disease 3e.
Clinical manifestations

- Classical pertussis (mainly in unimmunized toddlers and children)
  - 3 stages, after incubation of 7 to 10 days (range 5 to 21 days)
    - Catarrhal: nondistinctive catarrhal symptoms
    - Paroxysms of cough with inspiring whoop and often post-tussive vomiting
    - Convalescent phase: , with recurrence of paroxysmal cough for up to 1 year

- Atypical pertussis
  - Infants: less inspiratory whoop but gagging, gasping, cyanosis, apnea and prolonged convalescent phase
  - Immunized children and adults: all stages foreshortened, often without paroxysms and without distinct stages
Clinical manifestations, atypical disease in babies
Severe and unrecognised: pertussis in UK infants

- Crowcroft et al. Arch Dis Child 2003; 88: 802-806
  Inclusion:
  → n=142 infants < 5 m, admission in PICU
  → resp failure – ALTE
  → systematically specimen collection for microbiology (incl *B pertussis*)
  Results:
  → 23% (n=33) pertussis confirmed (PCR, culture)
    - 33% co-infection with RSV
    - n=2 cases of fatality
  Conclusion:
  → severe pertussis is “atypical” and underdiagnosed
  → clinical diagnosis is difficult
  → RSV co-infection does not exclude pertussis and can aggravate clinical course
Clinical manifestations, atypical disease in oldies
A Pertussis Outbreak Associated with Social Isolation among Elderly Nuns in a Convent


- Outbreak in a Dutch convent in 1992, recognized at week 10; cohort study started at week 12. **Total: 66 cases / 75 nuns**
- Diagnosis: PCR, culture, significant change (3-fold) in IgG ab titres at weeks 9, 13 and 60 or single titre > 100 U/ml
- Incidence rates
  - 75 retired nuns (none vaccinated) IR = 60%
    - 9 with life-long career in convent IR = 100%
    - 66 with career outside convent IR = 55% P = 0.007
      - of which:
        - only in Dutch society IR = 74%
        - only in tropics IR = 35%
        - both IR = 17% P < 0.0001
  - 24 staff members (21 vaccinated) IR = 8%
- Mortality
  - 3/4 deaths were among nuns in age group 85-94 years
Clinical manifestations, atypical disease in oldies
A Pertussis Outbreak Associated with Social Isolation among Elderly Nuns in a Convent

Table 1. Relationship between pertussis incidence, age, and duration of isolation from society among 75 nuns with \( n = 66 \) and nuns without \( n = 9 \) a career outside the convent.

<table>
<thead>
<tr>
<th>Group</th>
<th>Pertussis incidence</th>
<th>Duration of cough among nuns with pertussis</th>
<th>No. of deaths among nuns with pertussis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. with pertussis/total</td>
<td>Percentage</td>
<td>( P ) for linear trend in proportions</td>
</tr>
<tr>
<td>Age group, years</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>55–64</td>
<td>8/16(^a)</td>
<td>50</td>
<td>(.31)</td>
</tr>
<tr>
<td>65–74</td>
<td>13/21</td>
<td>62</td>
<td></td>
</tr>
<tr>
<td>75–84</td>
<td>16/27</td>
<td>59</td>
<td></td>
</tr>
<tr>
<td>85–94</td>
<td>8/11(^a)</td>
<td>73</td>
<td>(.005)</td>
</tr>
<tr>
<td>Duration of isolation from society, (^c) years</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0–6</td>
<td>5/15</td>
<td>33</td>
<td></td>
</tr>
<tr>
<td>7–13</td>
<td>9/17</td>
<td>53</td>
<td></td>
</tr>
<tr>
<td>14–20</td>
<td>10/16</td>
<td>63</td>
<td></td>
</tr>
<tr>
<td>21–34</td>
<td>9/12</td>
<td>75</td>
<td></td>
</tr>
<tr>
<td>35–70</td>
<td>12/15</td>
<td>80</td>
<td></td>
</tr>
</tbody>
</table>

\(^a\) No significant difference between the incidence of pertussis in the youngest and oldest age group \( (P = .21) \), Fisher’s exact test.

\(^b\) No significant difference between the median duration of cough in the youngest and oldest age group \( (P = .10) \).

\(^c\) Duration of isolation of the 9 nuns without a career outside the convent and the duration of isolation since retirement of the 66 nuns with a career outside the convent.

Discussion: waning immunity due to social isolation in women born in 1898-1936, when 100% of population experienced pertussis before 15 years of age.
In the context of the Eupert-Labnet WP6 seroprevalence study (comparing sera from 14 European member states), 1500 anonymized leftover diagnostic samples were collected randomly during the second semester of 2012 by the laboratories of clinical biology of six participating Belgian centres, equally distributed between Flanders, Wallonia and Brussels Capital region. A total of 750 samples (125/centre) were selected from subjects in the age group 20-29 years and 750 samples (125/centre) from subjects in the age group 30-39 years.
Sixty-one (4%) sera were indicative of an infection in the past two years (between 50 and 100 IU/ml) and another sixty-one (4%) sera had anti-PT IgG antibodies reflecting acute infection (> 100 IU/ml).

These results highlight the presence of a *Bordetella pertussis* reservoir in the adult ‘healthy’ Belgian population.
Laboratory diagnosis

- **Symptoms**
  - Rhinorrhea, sneezing, +/- fever
  - Cough
  - Frequency, severity
  - Distinctive paroxysms, post-tussive emesis, whoop

- **Laboratory Findings**
  - Lymphocytosis
  - Chest radiograph

- **Diagnosis Tests**
  - PCR: 0 – 3 to 4 weeks
  - Culture
    - ++
  - DFA
    - ++
  - Serology
    - Serology > 3 weeks
Only Bordetella pertussis is included in the ECDC diagnosis of pertussis

- **Bordetella pertussis**
  Exclusively isolated from humans, as agent of whooping cough (*has also been isolated from alveolar macrophages and blood*)

- **Bordetella parapertussis**
  Pertussis-like disease (no pertussis for ECDC)
  (mild cases: 20% pertussis, 40% aspecific bronchitis, 40% asymptomatic)
  Also found in sheep

- **Bordetella holmesii**
  Rare cause of respiratory or not respiratory infections in humans, recently isolated from patients with pertussis-like disease

- **Bordetella bronchiseptica**
  Respiratory pathogen for and commensal of many animals, incl. dog, pig, cat & rabbit
  Humans: rare cases of pertussis-like disease, opportunistic in immuundeficient patients
Pertussis

Pertussis (Bordetella pertussis)

The case definition and classification is that stipulated by EU Commission Decision of 8 August 2012.

Clinical criteria:
Any person with a cough lasting at least two weeks and at least one of the following three:

- Paroxysms of coughing
- Inspiratory "whooping"
- Post-tussive vomiting
or

Any person diagnosed as pertussis by a physician

or

Apnoeic episodes in infants

Laboratory criteria:
At least one of the following three:

- Isolation of Bordetella pertussis from a clinical specimen
- Detection of Bordetella pertussis nucleic acid in a clinical specimen
- Bordetella pertussis specific antibody response

SeroLOGY results need to be interpreted according to the vaccination status

Epidemiological criteria:
Two labs

→ UZ Brussel
  - Diagnosis on respiratory samples (PCR; culture for strain typing)
  - Most useful for young children
  - Confirmation of diagnosis/strain identification for other labs

→ WIV-ISP
  - Serological diagnosis (anti-PT IgG; not applicable if vaccine dose < 1 year)
  - Mostly samples from adults, generally one-point > 3 weeks symptoms
| NRC pertussis |
| Most important data |
| - Duration of symptoms |
| - Vaccination |

**Most important data**

- Duration of symptoms
- Vaccination
Real-time PCR: which target for which species?

- Screening assay: IS481-IS1001
  - High sensitivity: high copy number
  - Low specificity

- Confirmation assay: IS1002-recA
  - Lower sensitivity
  - Specificity is high in combination with first assay

- At first other assays were only performed after positive result in screening
  - practical difficulties
  - performing both at the same time

<table>
<thead>
<tr>
<th></th>
<th>B. pertussis</th>
<th>B. parapertussis</th>
<th>B. holmesii</th>
<th>B. bronchiseptica</th>
</tr>
</thead>
<tbody>
<tr>
<td>IS481++</td>
<td>-</td>
<td>++</td>
<td>+/-</td>
<td></td>
</tr>
<tr>
<td>IS1001-</td>
<td>++</td>
<td>-</td>
<td>+/-</td>
<td></td>
</tr>
<tr>
<td>IS1002+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+/-</td>
</tr>
<tr>
<td>recA-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

PCR interpretation

- **Sensitivity**
  - IS481: 3 CFU/PCR
  - IS1001: 5 CFU/PCR
  - recA: 52 CFU/PCR
  - IS1002: +/- 40 CFU/PCR

- **Possible outcomes:**
  - Positive for *B. pertussis*
  - Positive for *B. parapertussis*
  - Positive for *B. holmesii*
  - Positive for *B. species*, probably *B. pertussis* if compatible with clinical information
  - Positive for *B. species*, probably *B. parapertussis* if compatible with clinical information
  - **Undetermined** (weak positive signal, which was not confirmed by repeating the test, sample should be considered as negative)
● 1395 cases of *B. pertussis* infection

→ Serology: 930 cases  
→ PCR: 456 cases  
→ Serology + PCR: 7 cases  
→ 2 strains for confirmation by culture

● 94 *B. parapertussis*

● 7 *B. holmesii*
B. pertussis cases reported by the NRC

© WIV-ISP | Data source: NRC
Total number of reported pertussis cases by semester by surveillance system, 2005-2015 (first semester), Belgium.
The incidence of the reported pertussis cases by the NRC per region per 100,000, 2014, Belgium (based on 2014 Belgian population data).
Geographical distribution of reported pertussis cases by borough, 2014, Belgium (NRC, the sentinel laboratory network and the mandatory notification).
Deaths due to _Bordetella (pertussis)_ per age group, 2000-2012, Belgium.

<table>
<thead>
<tr>
<th>Year</th>
<th>Number of recorded deaths</th>
<th>Age group (in years)</th>
<th>Number of recorded deaths</th>
<th>Age group (in years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2000</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>65-74</td>
</tr>
<tr>
<td>2001</td>
<td>0</td>
<td>/</td>
<td>0</td>
<td>/</td>
</tr>
<tr>
<td>2002</td>
<td>0</td>
<td>/</td>
<td>0</td>
<td>/</td>
</tr>
<tr>
<td>2003</td>
<td>0</td>
<td>/</td>
<td>0</td>
<td>/</td>
</tr>
<tr>
<td>2004</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>&lt;1</td>
</tr>
<tr>
<td>2005</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>45-64</td>
</tr>
<tr>
<td>2006</td>
<td>1</td>
<td>&lt;1</td>
<td>0</td>
<td>/</td>
</tr>
<tr>
<td>2007</td>
<td>0</td>
<td>/</td>
<td>0</td>
<td>/</td>
</tr>
<tr>
<td>2008</td>
<td>0</td>
<td>/</td>
<td>&lt;5</td>
<td>15-19</td>
</tr>
<tr>
<td>2009</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>&lt;1</td>
</tr>
<tr>
<td>2010</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>&lt;1</td>
</tr>
<tr>
<td>2011</td>
<td>2</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>&lt;1</td>
</tr>
<tr>
<td>2012</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;5</td>
<td>&lt;1</td>
</tr>
</tbody>
</table>
LE MICROBE DE LA COQUELUCHE

PAR LES DRs J. BORDET ET O. GENGOU
Avec la planche XXVIII.

(Travail de l'Institut Pasteur de Bruxelles.)