Decline in antibiotic resistance and changes in the serotype distribution of *Streptococcus pneumoniae* isolates from children with acute otitis media; a 2001–2011 survey by the French Pneumococcal Network

M. Kempf[1], E. Varon[2], A. Lepoutre[3], A. Gravet[1], R. Baraduc[1], M. Brun[1], H. Chardon[1], J. Cremniter[1], J. Croizé[1], F. Dalmay[4], M.-C. Demachy[1], T. Fosse[1], C. Grelaud[1], T. Hadou[1], F. Hamdad[1], J.-L. Koeck[1], S. Luce[4], S. Mermond[1], I. Patry[1], A. Péchinot[1], J. Raymond[1], A. Ros[1], C. Segonds[1], B. Soullié[1], D. Tandé[1], M. Vergnaud[1], V. Vernet-Garnier[1], F. Wallet[1], L. Gutmann[2], M.-C. Ploy[1] and P. Lanotte[1]

1) *French pneumococcal surveillance network 'Observatoires Régionaux du Pneumocoque', CHU Limoges, Limoges,* 2) *Centre National de Référence des Pneumocoques (CNRP), AP-HP HEGP, Paris,* 3) *Institut de Veille Sanitaire (InVS), Saint Maurice and* 4) *CHU Limoges, UFRCB, Limoges, France*

Abstract

Streptococcus pneumoniae is an important cause of acute otitis media (AOM). The aim of this study was to evaluate trends in antibiotic resistance and circulating serotypes of pneumococci isolated from middle ear fluid of French children with AOM during the period 2001–2011, before and after the introduction of the PCV-7 (2003) and PCV-13 (2010) vaccines. Between 2001 and 2011 the French pneumococcal surveillance network analysed the antibiotic susceptibility of 6683 *S. pneumoniae* isolated from children with AOM, of which 1569 were serotyped. We observed a significant overall increase in antibiotic susceptibility. Respective resistance (I+R) rates in 2001 and 2011 were 76.9% and 57.3% for penicillin, 43.0% and 29.8% for amoxicillin, and 28.6% and 13.0% for cefotaxime. We also found a marked reduction in vaccine serotypes after PCV-7 implementation, from 63.0% in 2001 to 13.2% in 2011, while the incidence of the additional six serotypes included in PCV-13 increased during the same period, with a particularly high proportion of 19A isolates. The proportion of some non-PCV-13 serotypes also increased between 2001 and 2011, especially 15A and 23A. Before PCV-7 implementation, most (70.8%) penicillin non-susceptible pneumococci belonged to PCV-7 serotypes, whereas in 2011, 56.8% of penicillin non-susceptible pneumococci belonged to serotype 19A. Between 2001 and 2011, antibiotic resistance among pneumococci responsible for AOM in France fell markedly, and PCV-7 serotypes were replaced by non-PCV-7 serotypes, especially 19A. We are continuing to assess the impact of PCV-13, introduced in France in 2010, on pneumococcal serotype circulation and antibiotic resistance.

Keywords: Epidemiology, French regional observatories, pneumococcal vaccine, *Streptococcus pneumoniae*
Original Submission: 9 June 2014; **Revised Submission:** 25 July 2014; **Accepted:** 6 August 2014
Editor: I. Gyssens
Article published online: 12 October 2014

This work was partially presented at the ESPID Congress in 2013 (poster discussion No. C2-258)
Corresponding author: P. Lanotte, Service de Bactériologie-Virologie, Hôpital Bretonneau, CHRU de Tours, 2 boulevard Tonnellé, F-37044 Tours Cedex, France
E-mail: philippe.lanotte@univ-tours.fr

Introduction

Acute otitis media (AOM) is the most common bacterial infection in childhood and one of the most frequent indications for antibiotic treatment of young children [1]. Among the pathogens involved in this infection, *Streptococcus pneumoniae* is the most frequently isolated, together with *Haemophilus influenzae* [2]. In recent decades the emergence and spread of penicillin non-susceptible pneumococci (PNSP) has limited the therapeutic options in AOM, as penicillin non-susceptibility is associated with reduced activity of other oral β-lactams [3–5]. In France, a steady increase in the percentage of PNSP isolates in both invasive and non-invasive infections has been observed since the 1990s, reaching 55.4% overall in 2001 and 77% among paediatric AOM isolates [6]. In 2001 the French national health

http://dx.doi.org/10.1016/j.cmi.2014.08.009

insurance system initiated an extensive programme to avoid inappropriate antibiotic use among outpatients [6]. The heptavalent pneumococcal conjugate vaccine (PCV-7) was recommended for at-risk children under 2 years of age in 2002, and for all children in 2006 [7]. PCV-7 vaccine coverage increased slowly: at least one dose was received by 62% of children under 2 years old in 2006, rising to 69% in 2007, 85% in 2008 and 90% in 2009 [7]. PCV-13 gradually replaced PCV-7 over a 3-month period between June and early September 2010 [8]. In 2011, the full primary vaccination coverage rate was 91.7% among 9-month-old infants [Public Health Institute unpublished data, available at http://www.invs.sante.fr/Dossiers-thematiques/Maladies-infectieuses/Maladies-a-prevention-vaccinale/Couverture-vaccinale/Donnees/Pneumocoque].

The aim of this study was to evaluate, through the French pneumococcal surveillance network (Observatoires Régionaux du Pneumocoque, Centre National de Référence des Pneumocoques, Institut de Veille Sanitaire), changes in antibiotic resistance and circulating serotypes of *S. pneumoniae* isolated from middle ear fluid of children (0–16 years) with AOM over an 11-year period (2001–2011) following the introduction of PCV-7 and PCV-13.

Materials and Methods

Data and strain collection

Surveys were conducted every 2 years from January 2001 to December 2011 by the French pneumococcal network, which includes 396 public (75%) and private (25%) laboratories in 23 regions, as previously described [6]. All isolates from middle ear fluid of children aged 0–16 years were included in the study. Only one strain was considered per AOM episode and per child.

Susceptibility testing

Minimal inhibitory concentrations (MICs) of penicillin G, amoxicillin and cefotaxime were determined with the agar dilution method according to the guidelines of the Comité de l'Antibiogramme de la Société Française de Microbiologie (www.sfm.org). Three quality-control strains (R6 (WT), ATCC49619 and CNRP32475) provided by the French National Reference Centre for Pneumococci (CNRP) were included. Erythromycin and cotrimoxazole susceptibility was determined using the disk diffusion method, the VITEK2® kit or the ATB-Pneumo® kit. The results were interpreted according to 2009 French guidelines, with the following breakpoints: 0.064–2 mg/L for penicillin, and 0.5–2 mg/L for amoxicillin and cefotaxime. PNSP were defined as strains with a penicillin MIC >0.064 mg/L.

Serotyping

A subset of isolates was selected for serotyping by random systematic sampling. Serotyping was performed by the National Pneumococcal Reference Centre, using the capsular swelling method with commercial antisera (Statens Serum Institute, Copenhagen, Denmark). The panel of antisera covered the 92 known serotypes.

Statistical analysis

Data were monitored and validated with CAPTURE SYSTEM software. SAS software (version 9.1.3., SAS Institute, Cary, NC, USA) was used for statistical analysis, using the chi-square test for trends. Values of $p < 0.05$ were considered to denote significant differences.

Results

We studied 6683 isolates recovered from middle ear fluid of 6683 children with AOM. The annual number of pneumococcal isolates declined gradually during the study period, from 1694 in 2001 to 1378 in 2003, 1159 in 2005, 998 in 2007, 922 in 2009 and 560 in 2011. The children's mean age ± SD was 1.9 ± 2.0 years. A total of 2189 isolates (32.8%) were recovered from children less than 12 months old, and 2314 (34.6%) from children between 1 and 2 years old.

Antimicrobial resistance

Trends in resistance (I+R) to penicillin, amoxicillin, cefotaxime, erythromycin and cotrimoxazole among the 6683 isolates are shown in Table I. A significant decrease in overall antibiotic

TABLE I. Resistance of pneumococcal isolates from children's middle ear fluid to five antibiotics; France 2001–2011

	2001 (n = 1694)	2003 (n = 1378)	2005 (n = 1159)	2007 (n = 998)	2009 (n = 922)	2011 (n = 560)	p
PEN I+R, *n* (%)	1302 (76.9)	959 (69.6)	736 (63.5)	604 (60.5)	581 (63.0)	321 (57.3)	$<10^{-5}$
AMX I+R, *n* (%)	729 (43.0)	572 (41.5)	405 (34.9)	298 (29.9)	338 (36.7)	167 (29.8)	$<10^{-5}$
CTX I+R, *n* (%)	485 (28.6)	369 (26.8)	259 (22.3)	164 (16.4)	192 (20.8)	73 (13.0)	$<10^{-5}$
ERY[a] I+R, *n* (%)	1303 (77.6)	1016 (74.1)	708 (61.9)	585 (59.3)	541 (58.7)	310 (55.4)	$<10^{-5}$
SXT[b] I+R, *n* (%)	862 (53.8)	572 (47.7)	386 (39.3)	306 (31.7)	419 (45.4)	130 (23.2)	$<10^{-5}$

Abbreviations: AMX, amoxicillin; CTX, cefotaxime; ERY, erythromycin; PEN, penicillin G; SXT, cotrimoxazole.
Note: p values are from a χ^2-test for trends
[a]For erythromycin (ERY), the number and percentage of I+R isolates were calculated for 1680, 1370, 1143 and 986 isolates in 2001, 2003, 2005 and 2007, respectively
[b]For cotrimoxazole (SXT), the number and percentage of I+R isolates were calculated for 1602, 1200, 981 and 968 isolates in 2001, 2003, 2005 and 2007, respectively.