Invasive bacterial infections in a paediatric emergency department in the era of the heptavalent pneumococcal conjugate vaccine

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Objective **To describe the characteristics of patients diagnosed with invasive bacterial infections (IBIs) in a Paediatric Emergency Department (PED) following the introduction of the heptavalent pneumococcal conjugated vaccine (PCV7).**

Methods **Descriptive retrospective study of children under 14 years of age diagnosed with IBIs in a PED of a tertiary hospital between January 2008 and December 2009.**

Results **In this period we registered 123 396 episodes and 59 patients who were diagnosed with IBIs (22 patients under 1 year of age, 37.2%). Of these, 11 (18.6%) had some severe underlying condition and 38 (64.4%) were stable on arrival. The most common diagnoses were sepsis with/without meningitis (23, 38.9%) and bacteraemia (14, 23.7%), while the pathogens most frequently isolated were *Streptococcus pneumoniae* (23, 38.9%) and *Neisseria meningitidis* (18, 30.5%). Pathogens were isolated from blood in 57 patients and from the cerebrospinal fluid in eight (in these, the same bacterial species was isolated in the blood, except for two cases with *S. pneumoniae*). Of the pneumococci isolated, 80% corresponded to serotypes included in the 13-valent PCV13. In seven cases, pathogens were detected using only PCR analysis (*N. meningitidis*, four; *S. pneumoniae*, three). Twenty-five patients were admitted to the Paediatric Intensive Care Unit. No patient died but two had sequelae.**

Conclusion **In the era of PCV7, pneumococcus is the leading cause of IBI in PED. The introduction of PCV13 may lead to a very significant decrease in the IBI rate and meningococcus may become the leading cause of IBI. *European Journal of Emergency Medicine* 00:000–000**

European Journal of Emergency Medicine 2011, **00**:000–000

Keywords: invasive bacterial infection, pneumococcal conjugate vaccine, polymerase chain reaction

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Received 16 February 2011 **Accepted** 28 April 2011

Introduction

Fever is a common reason for children to be brought to Paediatric Emergency Departments (PEDs), and in most cases they have self-limiting viral infections. It can be difficult to identify those with the most severe infections, such as meningitis or occult bacteraemia, as patients arrive at hospital very early in the course of their illness at which point signs and symptoms of invasive bacterial infection (IBI) tend to be nonspecific. Given this, in some cases complementary tests to detect IBIs should be carried out in at-risk patients.

Classically, since the introduction of the conjugate vaccine against *Haemophilus influenzae b*, *Streptococcus pneumoniae* has been the most commonly isolated pathogen in IBIs in childhood. However, the introduction of the pneumococcal conjugated vaccine (PCV7) has caused changes in invasive pneumococcal diseases. There has been a significant decrease in the prevalence of invasive pneumococcal diseases among vaccinated populations [1–4], but, in contrast, in the last few years, serotype replacement has started to be reported [5–9]. PCV7 was introduced in the Basque Country in 2001 and, although it is not included in the list of child vaccinations paid for by the Public Health System, it is estimated that the rate of vaccination is 70%. Not being the only reason, the introduction of PCV7 has been accompanied by a replacement of the pneumococcal serotypes, and, in turn, new pneumococcal conjugate vaccines (including more serotypes) have been developed to prevent these IBIs.

The objective of this study is to describe the clinical and microbiological characteristics of patients diagnosed with IBIs in our PED following the introduction of the PCV7.

Methods

Study design

This is a descriptive retrospective study including patients under 14 years of age diagnosed with IBI attending the PED of a tertiary teaching hospital.

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Definitions

Invasive bacterial infection

IBI was defined as the isolation in blood or cerebrospinal fluid (CSF) of a bacterial pathogen, using bacterial culture or PCR techniques. The results were considered positive when the following microorganisms were isolated: *Neisseria meningitidis*, *S. pneumoniae*, *Escherichia coli*, *Staphylococcus aureus*, *Proteus mirabilis*, *Enterococcus faecium*, whereas coagulase-negative *Staphylococci* were considered the contaminant in healthy patients.

The blood culture media used were BACTEC PLUS aerobic/F (Becton Dickinson San Agustín del Guadalix, Spain). The CSF culture was performed according to conventional techniques.

For the PCR, the extraction of nucleic acids was performed using the QIAamp DNA Mini Kit (QIAGEN, Hilden, Germany) (blood) or EZ1 Virus Mini Kit (CSF) (QIAGEN Sample & Assay Technologies. Hilden, Germany). The molecular diagnosis of *N. meningitidis* and *S. pneumoniae* was made using real-time PCR with TaqMan probes (RealCycler MENE, Progenie molecular Valencia. España) in a SmartCycler System (Cepheid Europe, Maurens-Scopont, France), on EDTA-treated blood samples and/or CSF. The detection of *N. meningitidis* and *S. pneumoniae* was based on the amplification of fragments of the capsular transport protein and pneumolysin genes. The internal control was a heterologous DNA construct, containing regions homologous to the human *CFTR* gene and the pathogen.

Well-appearing children: Children were considered to appear well when assessed by a qualified nurse in the PED within half an hour of admission and by the emergency paediatrician using the Pediatric Assessment Triangle of the American Academy of Pediatrics [10]. For the general condition of a child to be classified as well, their appearance, work of breathing and circulation to the skin had to be normal and these data were noted in their medical records.

Case

Cases were established related to the day of the consultation on which the blood sample was taken for bacterial culture or PCR, the day on which the attending physician considered that the patient could have an IBI. Previous consultations were defined as any visits that had occurred in the preceding days, and reconsultations as visits made in the following days.

Management of febrile patients in our Emergency Department:

(1) Toxic-appearing children, all febrile patients with a toxic appearance are admitted for antibiotic treatment and supportive care.
(2) Well-appearing children, the management is related differently in the presence/absence of focality. The decision to obtain a blood culture from previously healthy patients with fever without a source and well appearance varies according to age and vaccine status. For all infants under 3 months of age with fever, complete blood cell count, C-reactive protein (CRP), procalcitonin (PCT) and blood culture are indicated. In older infants (3–24 months of age) blood tests are only routinely performed for those with a temperature of 39.5°C or more who have received fewer than two doses of the PCV7. Further, a chest radiograph may be considered depending on the blood test results, and we recommend obtaining it for a child under 24 months of age with fever without a source, if white blood cell (WBC) count was more than 20.000/mm^3. In babies between 3 and 24 months of age and with a temperature of more than 39°C, we recommend obtaining a urine sample for all baby girls under 24 months of age and baby boys under 12 months of age. PCR was obtained when there was a suspicion of sepsis or bacterial meningitis.

The parents or guardians of children managed as outpatients receive verbal and written instructions explaining that they should observe children at home and describing what should prompt them to bring the child back to hospital or to their general paediatrician.

System of data collection

The period of the study was from 1 January 2008 to 31 December 2009. Positive results in these tests were identified in the electronic records of the Microbiology Service of our hospital, with updated reports provided on a monthly basis. Data regarding the patient and the episode were obtained from the hospital admission records and subsequent electronic clinical records of the PED. The following data were collected: case number, date of the consultation, age, sex, personal history, vaccine status, duration of fever and maximum temperature, associated symptoms, temperature in the PED, general condition, findings from the Pediatric Assessment Triangle, physical examination, diagnosis at earlier consultation and number of days since this previous visit (when applicable), previous antibiotic therapy, vital signs (heart and respiratory rates, oxygen saturation and blood pressure), blood results, CSF, urine and/or other complementary tests, destination of the patient, approach taken after arrival of the culture results and treatment and progress.

Statistical analysis

Statistical analysis was carried out using the Statistical Package for the Social Sciences (SPSS version 17; IBM company, Chicago, Illinois, USA). Data were expressed as mean and standard deviations, and numbers and percentages for quantitative and categorical variables, respectively. Quantitative variables were compared using Student's *t*-tests, whereas categorical variables were analysed using

χ^2 and Fisher's exact tests. Results of P values of less than 0.05 were considered statistically significant.

The study was approved by the Research Committee of the Paediatric Emergency Department. Data collection and entry were carried out anonymously. Informed consent was not considered necessary, as no intervention was provided or withheld in association with this study.

Results

Epidemiology

During the study period, 123 396 cases were seen in the PED, for which 3392 blood cultures (2.75%), 391 CSF cultures (0.32%) and PCR analysis of blood and CSF (in 80 and 56 patients, respectively) were performed. Fifty-nine patients were diagnosed with IBI and their characteristics are listed in Table 1 and Fig. 1.

Of these patients, 11 (18.6%) had a relevant underlying disease, this being more common in children over 5 years of age (33.3%, three of nine; vs. 13.5%; eight of 50 in younger children), but the difference was not significant ($P = 0.35$).

Clinical signs and symptoms

More than half of the children (54.2%) were brought to hospital within 12 h of the onset of signs and symptoms. On arrival at the PED, 63% were stable (well appearance, no clinical sign of instability, normal blood pressure) and 23.7% were afebrile. There were no relevant findings on physical examination in 32.2%, whereas in the others petechiae–ecchymoses were the most common observations (17%).

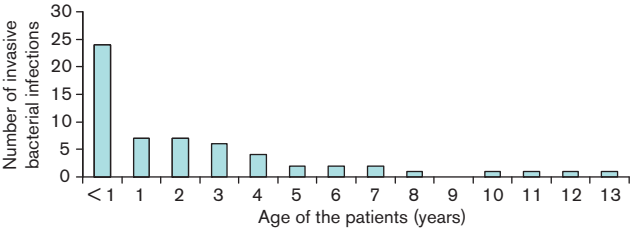
Of the 59 patients with IBI, seven had been seen previously in our PED, most of them having been diagnosed with fever without a source. Three of them were younger than 2 years of age. On their previous visits, all had been stable and their physical examinations normal, no complementary tests were requested, and they were managed as outpatients with instructions to be monitored at home. The second visit was prompted by the appearance of new signs, such as petechiae or malaise, or because they continued to have a high temperature. The definitive diagnoses of these children were: sepsis (three), pneumonia (two), meningitis (one) and bacteraemia (one). With the exception of the two children whose eventual diagnosis was pneumonia, all had come back to the hospital within 24 h. None of them had a relevant personal history or sequelae.

Table 1 **Characteristics of the children with invasive bacterial infections**

Period of the year	
October–March	43/59 (72.9%)
April–September	16/59 (27.1%)
Sex	
Male	35/59 (59.3%)
Female	24/59 (40.7%)
Personal medical history	
Respiratory	1/59 (1.7%)
Immunological	4/59 (6.8%)
Nephrological	4/59 (6.8%)
Others	2/59 (3.4%)
None	48/59 (81.4%)
PCV7	
None	29/59 (49.2%)
One dose	3/59 (5.1%)
Two doses	5/59 (8.5%)
Three doses	5/59 (8.5%)
Four doses	8/59 (13.6%)
Unknown	9/59 (15.3%)

PCV7, pneumococcal conjugated vaccine.

Fig. 1



Invasive bacterial infections related to the age of the patient.

Acute-phase reactants and cytochemical examination of cerebrospinal fluid

In our series, WBC count was obtained for 57 patients (24, 42.1%, had normal WBC count; five, 8.8%, less than 5000 leucocytes/μl; and 28, 49.1%, more than 15 000 leucocytes/μl), and 33 (56%) had more than 10 000 neutrophils/μl.

The CRP was obtained in 56 patients and was above 20 mg/l in 42 (75%). In 88.9% of the 36 patients in whom the PCT was obtained it was higher than 0.5 ng/dl.

Two patients had normal blood test results: a 3-year-old girl with an infection of the bottom of the mouth and bacteraemia due to *S. pyogenes* and a 16-month-old immunosuppressed child with bacteraemia due to *Pseudomonas aeruginosa*.

For five patients, a second blood test was taken within 24 h, and three of these produced abnormal results that had not been detected previously.

Samples of CSF were obtained from 15 patients, eight of which revealed pleocytosis and/or had a positive-gram stain.

Microbiology

The bacterial isolates are listed in Table 2.

In all the patients in whom a pathogen was isolated from the CSF, the same pathogen was also found in their blood, except for two in whom a pneumococcus was isolated only in the CSF.

In seven patients (11.8%), the pathogen was only identified by PCR: 11.5% of the pneumococcal and 20% of the meningococcal isolates. PCR was not obtained for all the patients (11 in blood, five in CSF and four in blood and CSF).

The serotypes of 20 cases of *S. pneumoniae* were identified; these and their relationship with the various pneumococcal conjugate vaccines are reported in Table 3.

Table 2 Bacterial isolates in blood and cerebrospinal fluid

Medium	Species	Number of isolates	Percentage
Blood		*N*=57	%
	S. pneumoniae	21	36.8
	N. meningitidis	18	31.5
	E. coli	5	8.7
	S. aureus	4	7.0
	S. agalactiae	2	3.5
	S. pyogenes	2	3.5
	P. aeruginosa	2	3.5
	E. faecium	1	1.7
	P. mirabilis	1	1.7
	K. pneumoniae	1	1.7
CSF		8	
	S. pneumoniae	5	62.5
	N. meningitides	2	25
	S. agalactiae	1	12.5

CSF, cerebrospinal fluid.

Table 3 Serotypes causative of the invasive pneumococcal infections

Serotype	*n*
In PCV7	
14	2
19F	1
In PCV10	
1	5
7F	2
In PCV13	
19A	6
Other serotypes	
10A	1
7	1
17	1
23B	1

PCV7, pneumococcal conjugated vaccine.

The three patients with pneumococcal serotypes included in the PCV7 had not received any doses of PCV7.

Diagnosis

The diagnoses and the characteristics of the 59 patients with IBIs are listed in Table 4. Most common diagnoses were sepsis and bacteremia.

Treatment and progression

A total of 43 patients (72.8%) were admitted, of which 25 were transferred to the Paediatric Intensive Care Unit. None of the patients died.

All the 16 patients who were discharged received antibiotic, except one who had a normal physical examination and did not show any alteration in the tests practiced. Of these 16 patients, 11 had a pneumococcal infection.

Two patients, both with *S. pneumoniae* serotype 7F, had sequelae: one necrotising pneumonia with a bronchopleural fistula and the other hydrocephalus, endocarditis and deafness.

Discussion

The pneumococcus is the main cause of IBI in our PED in the era of the PCV7. The second cause is the meningococcus that accounts for nearly one-third of the cases. Children are brought to hospital with very early onset of symptoms (mainly, fever) and tend to have nonspecific signs and symptoms.

Fever is a very common sign and reason for consultation in the PEDs [11] and it has always been a challenge for paediatricians to identify severe and potentially severe infections. To this end, a great deal of research has been conducted and protocols have been designed to support decisions concerning complementary tests. These studies stress the importance of the age, vaccine status, and in particular appearance when admitted to the Emergency Department [12]. Further, to correctly manage febrile paediatric patients, it is essential to have up-to-date information concerning the prevalence of pathogens causing IBI and to identify differences between geographical areas

Table 4 Diagnoses and characteristics of the 59 children with invasive bacterial infections

Definitive diagnosis	*n* (%)	Well appearance (%)	Duration of fever >12 h	Altered physical examination[a]	Altered WBC[b]	Altered CRP[c] (%)	Altered PCT[d] (%)
Sepsis	23 (38.9)	12 (52)	11	23/23 (100%)	14/22 (63.6%)	20/21 (95)	17/17 (100)
Bacteraemia	14 (23.7)	13 (92)	6	2/14 (14.6%)	7/13 (53.8%)	5/13 (38.5)	4/7 (57)
Pneumonia	8 (13.5)	2 (25)	7	7/8 (88.5%)	5/8 (62.5%)	7/8 (87.5)	1/1 (100)
Meningitis	6 (10.1)	2 (33)	5	5/6 (83.2%)	4/6 (66.6%)	4/6 (66.7)	4/5 (80)
Urinary tract infection	5 (8.4)	5 (100)	2	0/5	3/5 (60%)	4/5 (80)	5/5 (100)
Osteoarticular and soft tissues infections	3 (4.9)	3 (100)	2	3/3 (100%)	0/3	2/3 (66.7)	1/1 (100)
Total	59	37/59 (62.7)	33/59	40/59 (67.7%)	33/57 (57.8%)	42/56 (75)	32/36 (88.8)

Data are expressed as numbers and percentages.
CRP, C-reactive protein; PCT, procalcitonin; WBC, white blood cell.
[a]Presence of any focality or petechiae.
[b]Patients with a number of leucocytes lower than 5000/mm^3 or higher than 15 000/mm^3. WBC was obtained in 57 patients.
[c]C-reactive protein values higher than 20 mg/l. CRP was obtained in 56 patients.
[d]Procalcitonin values higher than 0.5 ng/ml. PCT was obtained in 36 patients.

[13]. Classically, *H. influenzae* has been the main causative agent of this type of infection [14–16]. However, the introduction of a vaccine against this pathogen has significantly decreased its prevalence, *S. pneumoniae* subsequently becoming the main causative agent. The introduction of the PCV7 in our region has changed the pattern of IBI in children, decreasing the prevalence of pneumococcal bacteraemia in infants checked for occult bacteremia (1.62–0.69%) [1]. In particular, children who have received any doses of PCV7 have a lower risk of suffering from invasive pneumococcal infections [12,17–19]. In fact, in this study, all the patients with invasive pneumococcal infections due to a serotype included in the PCV7 had not received any doses of PCV7. However, most of the serotypes that we isolate nowadays are not included in the PCV7 therefore, as observed by other researchers [20–22], to have the highest degree of protection, children need to have received a PCV containing the most prevalent serotypes, currently including the serotype 19A. Indeed, this serotype is nowadays the main causative agent in invasive pneumococcal infection in children [23].

In our series, *S. pneumoniae* was the most common causative agent of IBI followed by *N. meningitidis*. The conjugate vaccine against 13 serotypes including 19A, PCV13, was introduced in our region soon after the end of this study. It seems logical to suppose that within the next years *N. meningitidis* will become the main agent involved in IBI in children. It is extraordinarily important to determine whether this is true, as the various potential complementary tests have very different degrees of effectiveness in identifying pneumococcal or meningococcal infections.

The typical profile of a child brought to the PED with an IBI is that of a previously healthy infant with recent-onset fever, nonspecific symptomatology and a good appearance on arrival. This means that it is difficult to differentiate between such patients on the basis of their clinical picture, making it necessary to perform complementary tests in high-risk children and recognize the importance of a close observation of such patients. In addition, personal medical history is very important in febrile children. In our series, a large proportion of patients had a relevant underlying condition and, as postulated by other researchers, this was much common in patients more than 5 years of age than younger ones, although in our study there was no significant difference probably because of our sample size [6].

Potentially useful complementary tests include WBC and neutrophil counts, and measurement of acute-phase reactants, such as CRP and PCT. Although in most cases abnormal results are found in such tests, there is no single parameter that enables us to identify all cases of IBI. Nevertheless, we note that PCT seemed to be among the more useful indicators, as has been found by other researchers, especially in the case of meningococcal infections [24]. Furthermore, sometimes blood tests should be repeated in PEDs, as infection parameters can change [25].

Treatment on an outpatient basis must be accompanied by an explanation to the family of warning signs that should prompt them to bring the child back to hospital. This explanation is given verbally in our department by the physician and in writing on discharge. These explanations help to enhance family involvement, improving their contribution to the management of the child's condition, and can decrease the number of complications due to delays in seeking medical attention. In our series, most children were brought to the hospital within hours of the onset of warning signs. None of them had notable complications.

With regard to the identification of the causative agent, a blood culture is still the gold standard. Nevertheless, in a relatively high proportion of our patients the pathogen was only isolated by PCR. It could be useful to adopt this technique in selected patients in emergency departments to enable such children to be diagnosed rapidly [26,27].

This study has certain limitations. The data were collected retrospectively. However, the fact that all cases seen in our department are recorded electronically, including medical histories and notes concerning progression and follow-up of patients, makes the collection process consistent. Further, the fact that this was not a multicentre study may limit the extent to which our findings can be extrapolated to other areas. In particular, there may be microbiological differences between countries that must be taken into account. Although we found patients in whom the microbiological diagnosis was reached only by using PCR techniques, we were unable to assess what results this technique would have produced in the febrile patients for whom only blood culture was performed. PCR techniques give very important information much faster than cultures and it seems clear that their systematic adoption in certain groups of patients is necessary in Paediatric Emergency Units. Finally, a larger sample size might have made it easier to reach conclusions for subgroups of patients, although this was not the main objective of this study.

To conclude, pneumococcus is currently the main causative agent of IBI seen in our PED, although the introduction of new pneumococcal conjugate vaccines may cause the meningococcus to become the most prevalent agent, with the implications this has for the management of patients with suspicion of IBI on arrival at hospital, related to the different value of acute phase reactants to identify the children with a serious bacterial infection among those admitted with fever to the emergency department.

Acknowledgements

The authors of this manuscript have no conflicts of interest to report. There is no financial support to disclose with respect to this study.

References

1 Benito J, Mintegi S, Pocheville I, Sánchez J, Azcunaga B, Capapé S. Pneumococcal bacteremia among infants with fever without known source before and after introduction or pneumococcal conjugate vaccine in the Basque Country of Spain. *Pediatr Infect Dis J* 2007; **26**:667–671.

2 Robinson KA, Baughman W, Rothrock G, Barrett NL, Pass M, Lexau C, *et al.*; for the Active Bacterial Core Surveillance (ABCs)/Emerging Infections Program Network. Epidemiology of invasive *Streptococcus pneumoniae* infections in the United States, 1995–1998. Opportunities for prevention in the conjugate vaccine era. *JAMA* 2001; **285**:1729–1735.

3 Feikin DR, Klugman KP. Historical changes in pneumococcal serogroup distribution: implications for the era of pneumococcal conjugate vaccines. *CID* 2002; **35**:547–555.

4 Aristegui J, Bernaola E, Pocheville I, García C, Arranz L, Durán G, *et al.* Reduction in pediatric invasive pneumococcal disease in the Basque Country and Navarre, Spain, after introduction of the heptavalent pneumococcal conjugate vaccine. *Eur J Clin Microbiol Infect Dis* 2007; **26**:303–310.

5 Singleton RJ, Hennessy TW, Bulkow LR, Hammitt LL, Zulz T, Hurlburt DA, *et al.* Invasive pneumococcal disease caused by nonvaccine serotypes among Alaska native children with high levels of 7-valent pneumococcal conjugate vaccine coverage. *JAMA* 2007; **297**:1784–1792.

6 Hsu K, Shea KM, Stevenson AE, Pelton SI; members of the Massachusetts Department of Public Health. Changing serotypes causing childhood invasive pneumococcal disease. *Pediatr Infect Dis J* 2010; **29**:289–293.

7 González BE, Hulten KG, Lamberth L, Kaplan SL, Mason EO; US Pediatric Multicenter Pneumococcal Surveillance Group. *Streptococcus pneumoniae* serogroups 15 and 33. An increasing cause of pneumococcal infections in children in the United States after the introduction of the pneumococcal 7-valent conjugate vaccine. *Pediatr Infect Dis J* 2006; **25**:301–305.

8 Hanage WP. Serotype replacement in invasive pneumococcal disease: where do we go from here? *J Infect Dis* 2007; **196**:1282–1284.

9 Hicks LA, Harrison LH, Flannery B, Hadler JL, Schaffner W, Craig AS, *et al.* Incidence of pneumococcal disease due to non-pneumococcal conjugate vaccine (PCV7) serotypes in the United States during the era of widespread PCV7 vaccination, 1998–2004. *J Infect Dis* 2007; **196**:1346–1354.

10 Dieckmann RA, Brownstein D, Gausche-Hill M. The pediatric assessment triangle: a novel approach for the rapid evaluation of children. *Pediatr Emerg Care* 2010; **26**:312–315.

11 Mintegi S, Benito J, García S, Corrales A, Bartolomé MJ, Trebolazabala N. Demanda y asistencia en un servicio de urgencias hospitalario. *An Pediatr (Barc)* 2004; **61**:156–161.

12 Mintegi S, Benito J, Sánchez J, Azkunaga B, Iturralde I, García S. Predictors of occult bacteremia in young febrile children in the era of heptavalent pneumococcal conjugated vaccine. *Eur J Emerg Med* 2009; **16**:199–205.

13 Lee GM, Fleisher GR, Harper MB. Management of febrile children in the age of conjugate pneumococcal vaccine: a cost-effectiveness analysis. *Pediatrics* 2001; **108**:835–844.

14 Campos J, Aracil B. Regreso de la infección por Haemophilus influenzae b. *An Pediatr (Barc)* 2003; **59**:425–428.

15 Pushparajah K, Ramnarayan P, Maniyar A, Paget R, Britto J. Continued threat of Haemophilus influenzae type B disease in the UK. *Lancet* 2003; **361**:90.

16 Rijkers GT, Vermeer-de Bondt P, Spanjaard L, Breukels MA, Sanders EAM. Return of Haemophilus influenzae type b infections. *Lancet* 2003; **361**:1563–1564.

17 Stoll ML, Rubin LG. Incidence of occult bacteremia among highly febrile young children in the era of the pneumococcal conjugate vaccine. A study from a Children's hospital emergency department and Urgent care center. *Arch Pediatr Adolesc Med* 2004; **158**:671–675.

18 Carstairs KL, Tanen DA, Johnson AS, Kailes SB, Riffenburgh RH. Pneumococcal bacteremia in febrile infants presenting to the emergency department before and after the introduction of the heptavalent pneumococcal vaccine. *Ann Emerg Med* 2007; **49**:772–777.

19 Wilkinson M, Bulloch B, Smith M. Prevalence of occult bacteremia in children aged 3–36 months presenting to the emergency department with fever in the postpneumococcal conjugate vaccine era. *Acad Emerg Med* 2009; **16**:220–225.

20 Hsu HE, Shutt KA, Moore MR, Beall BW, Bennett NM, Craig AS, *et al.* Effect of pneumococcal conjugate vaccine on pneumococcal meningitis. *N Engl J Med* 2009; **360**:244–256.

21 Levy C, Varon E, Bingen E, Lecuyer A, Boucherat M, Cohen R. PneumococcaL meningitis in French children before and after the introduction of pneumococcal conjugate vaccine. *Pediatr Infect Dis J* 2011; **30**:168–170.

22 Casado-Flores J, Rodrigo C, Aristegui J, Martinón JM, Fenoll A, Mendez C. Decline in pneumococcal meningitis in Spain after introduction of the heptavalent pneumococcal conjugate vaccine. *Pediatr Infect Dis J* 2008; **27**:1020–1022.

23 Kaplan SL, Barson WJ, Lin PL, Stovall SH, Bradley JS, Tan TQ, *et al.* Serotype 19A is the most common serotype causing invasive pneumococcal infections in children. *Pediatrics* 2010; **125**:429–436.

24 Carrol ED, Newland P, Riordan FAI, Thomson APJ, Curtis N, Hart CA. Procalcitonin as a diagnostic marker of meningococcal disease in children presenting with fever and a rash. *Arch Dis Child* 2002; **86**:282–285.

25 Bressan S, Andreola B, Cattelan F, Zangardi T, Perilongo G, Da Dalt L. Predicting severe bacterial infections in well-appearing febrile neonates: laboratory markers accuracy and duration of fever. *Pediatr Infect Dis J* 2010; **29**:227–232.

26 Klouche M, Schröder U. Rapid methods for diagnosis of bloodstream infections. *Clin Chem Lab Med* 2008; **46**:888–908.

27 Peters RP, van Agtmael MA, Danner SA, Savelkoul PH, Vandenbroucke-Grauls CM. New developments in the diagnosis of bloodstream infections. *Lancet Infect Dis* 2004; **4**:751–760.