Bacterial Meningitis in an Urban Area: Etiologic Study and Prognostic Factors

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Abstract

Objectives: The study of clinical features, diagnostic methods and prognostic factors of bacterial meningitis, in an urban area.

Patients and Methods: All patients admitted between June 2001 and July 2004 in the emergency departments of a few hospitals, with the diagnosis of bacterial meningitis were included. CSF and blood cultures were performed in every case. Phenotypic characterization of strains of *Streptococcus pneumoniae* and *Neisseria meningitidis* identified by culture were performed. In order to detect the three most common agents it was done a PCR assay in culture negative CSF samples.

Results: Bacterial meningitis was diagnosed in 201 patients. Etiologic definition was based on culture in 142 patients (70.6%), done by CSF PCR assay in 33 (16.4%) other patients and exclusively by latex agglutination test results in two cases. Thus, an etiologic diagnosis was established in 177 (88%) cases. Antigenic characterization showed a slight prevalence of *N. meningitidis* phenotype C:2b:P1; the *S. pneumoniae* serotype characterization showed that 43.8% of identified serotypes are not included in any of the available vaccines. Eighteen patients died (8.9%). The statistic analysis found that factors associated with an adverse outcome were age older than 50 years (OR 7.07; IC 95% 1.1–27.4), the presence of comorbidities (OR 3.3; IC 95% 1.1–9.6) and the occurrence of systemic complications (OR 5.8; IC 95% 2.1–16.0).

Conclusions: This epidemiologic pattern is similar to that found in other countries after the introduction of *Haemophilus influenzae* b conjugated vaccine. The association of culture and noncultural methods of diagnosis had a better performance in defining the etiology. Comparing to other series, in-patients mortality rate was lower (8.9%) than usually referred to, being considered unfavourable prognostic factors the age more than 50 years, the presence of comorbidities and of systemic complications.

Introduction

Bacterial meningitis is still associated with high mortality and neurological sequelae, even in developed countries. The severe forms of this disease are a medical emergency that can dramatically deteriorate to a severe form of intracranial hypertension, with consciousness depression or neurological deficits [1, 2]. While *Streptococcus pneumoniae* meningitis occurs in all age groups, *Neisseria meningitidis* is the first cause of meningitis in children, although it can also affect other age groups [3, 4].

Important modifications in the etiology of bacterial meningitis have been observed in the last decades, resulting from the development and availability of the new conjugated vaccines, designed after the antigenic definition of the most common etiologic agents: *Haemophilus influenzae* serotype b, *S. pneumoniae* and *N. meningitidis* serogroup C [5–7].

Otherwise, the prescription of pre-admission antibiotics has enhanced the interest of noncultural methods of diagnosis, as the case of molecular biology techniques, including real time PCR, which has a sensitivity and a specificity higher than 90% [8, 9].

Another important point was the emergence of antibiotic-resistant meningeal pathogens, particularly *S. pneumoniae*, with the increase of β-lactam multi-drug resistant isolates, although with a different geographic distribution. This is a growing problem in Europe that should not be forgotten when empiric treatment of bacterial meningitis is administered and enhances the need of a correct etiological diagnosis as well as a correct knowledge of local susceptibility profile to antibiotics [10, 11].

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As far as prevention is concerned, some advances during the last few years have to be acknowledged, with the introduction of conjugated vaccines against the most common agents. Epidemics by *N. meningitidis* of serogroup C has led to the introduction of the conjugated vaccine in the national immunization programs of several European countries [12, 13]. Recent publications of surveillance studies of meningococcal disease demonstrate the efficiency after 4 years of use of the conjugated vaccine in those countries, with a reduction in serogroup C incidence and a stable incidence of disease caused by serogroup B [14, 15]. Concerning pneumococcal disease prevention, some improvements have also been achieved, with the development of the conjugated vaccines [16]. In addition to their efficacy in preventing invasive disease, these conjugated vaccines also reduce the rate of nasopharyngeal carriage in children and could have the potential to decrease the transmission to non-immunized persons [17, 18]. Moreover, as antibiotic resistance is more commonly associated with the vaccine serotypes, vaccination can also have an impact on the emergence of resistant pneumococcal infections [14, 19].

In Portugal the conjugated vaccine to *H. influenzae* b has been included in the national immunization programme in 2000 and the conjugated vaccine to *N. meningitidis* serogroup C has been introduced in January of 2006. The heptavalent conjugated pneumococcal vaccine is commercially available in Portugal but has not been included in the immunization programme yet, although it is frequently prescribed, and studies to evaluate its efficacy are under way.

Our aim is the study of clinical presentation, diagnostic methods and prognostic factors of bacterial meningitis in an urban area.

Patients, Materials and Methods

From June 2001 to July 2004, 201 patients admitted in the emergency departments of the Oporto metropolitan area with diagnosis of acute meningitis were selected for study.

The cohort included children and adults with ages ranging from 1 month to 82 years of age (average age 26 years). One hundred and twenty (60%) patients were males.

Diagnosis of meningitis was based on an acute clinical syndrome with a less than a week's evolution, confirmed by a lumbar tap. Bacterial aetiology was considered when CSF cytosis exceeded 1,000 leukocytes/μl with proteins values higher than 50 mg/dl and glucose lower than 50% of blood values and/or if CSF cytosis lower than 1,000 but greater than 6 leukocytes/μl in the presence of a quick extending purpuric and/or petechial exantem. All CSF and blood specimens were cultured by microbiological standard methods and some samples were tested for capsular antigens. A CSF aliquot from the same sample collected for initial diagnosis was stored at –70 °C for subsequent study.

The selection of patients was done in emergency department and after this, patients' clinical and analytic data were collected based on medical records during hospitalization.

A definitive diagnosis of bacterial meningitis was based on agent identification from CSF and/or blood culture by standard methods, latex agglutination or by a PCR assay. Cases with a negative microbiologic investigation, but CSF leucocytes > 1,000/μl were considered as probable cases and also included in the study.

A PCR assay was applied to 86 CSF samples, 46 culture positive and 40 CSF culture negative specimens. The 36 culture-confirmed samples, for the three more common agents of bacterial meningitis included *S. pneumoniae* [20], *N. meningitidis* [13], and *H. influenzae* [3]. Other ten culture-positive CSF samples for *Staphylococcus epidermidis, Salmonella* spp., *Enterobacter* spp., *Enterococcus* spp. and *Klebsiella* spp. were included to test specificity.

The PCR assay was carried out using the primers sets of a real time PCR method as described by Corless: *ply* for *S. pneumoniae*, *ctrA* for *N. meningitidis* and *bexA* for *H. influenzae* [20] in culture-negative and culture-positive samples. Sybergreen detection was used instead of specific *N. meningitidis*, *S. pneumoniae* and *H. influenzae* probes and amplicon specificity verified with melting curve analysis.

Neisseria meningitidis ATCC 13090, *S. pneumoniae* ATCC 49619 and a clinical isolate of *H. influenzae* b were used as control strains. The bacterial suspensions were tested and used as positive control in each run.

In culture-positive cases, the isolates were stored at –70 °C in Brain-Heart medium added with 20% glycerol for antigenic characterization, using commercially available antisera (*Statens SerumInstitut – Copenhague*). Susceptibilities to the antibiotics commonly used were determined according to the "National Committee for Clinical Laboratory Standards" (NCCLSS) guidelines criteria [21].

A comorbid condition was considered whenever a chronic disease, an anatomic default, drug abuse or immunosuppressive treatment was present. HIV infected patients were excluded. An adverse outcome was defined by death. Proportions were compared using χ^2 test or Fisher's exact test, whenever appropriate. Crude OR and, respectively, 95% confidence intervals (95% CI) were used to measure the association between several factors and the likelihood of an unfavorable outcome. Logistic regression was use to estimate crude OR. For statistical analyses SPSS 12.0 was used.

Results

In this 3-year period bacterial meningitis was diagnosed in 201 patients. In 88 (43%) patients, at least one comorbidity was present: previous neurosurgery and pneumonia in 28 (14%) patients each, head trauma in 23 (11%), cerebrospinal leakage in 22 (11%), suppressed immune system condition in 19 (9%), diabetes in 17 (8%), acute otitis in 14 (7%), alcoholism in 10 (5%), ventriculoperitoneal shunt in 9 (4%), chronic otitis in 8 (4%), sinusitis in 7 (3%), cardiac failure in 6 (3%), splenectomy, drug abuse and chronic renal failure in two patients each and Celiac disease, Cushing syndrome, congenital heart disease in one patient each; in 32 (15.4%) antibiotics had been prescribed before admission. Meningitis was recurrent in 17 of the patients (8.4%).

Clinical findings observed on admission, by age group, are displayed in table 1. In 97 (48%) patients some form of neurological or/and systemic complication occurred

and in some cases more than one dysfunction. Mental status at admission was depressed (Glasgow Coma Scale < 15) in 170 (85%) patients, the score being less than 10 (GCS 3–10) in 77 (37%) and between 11 and 14 in 93 (45%). A normal mental status was observed in 18 (9%) patients; for 13 patients no information was registered. Seizures occurred in 38 (19%) patients, 30 (15%) had a focal motor deficit and 28 (14%) patients had cranial nerve palsies.

Table 1
Clinical findings at admission by age group.

	≤ 5 years	≥ 6 to ≤ 50 years	> 50 years	No (%)	p
Fever	70	86	39	195 (97)	0.864
Headaches	35[a,b]	80	30	145 (85)	0.011
Vomiting	45	56	21	122 (61)	0.436
Altered mental status (GCS < 15)	63	69	38	170 (85)	0.040
Nuchal rigidity	44[c]	72	30	146 (73)	0.032
Behavior manifestations	30	30	18	78[d] (50)	0.116
Fotophobia	12	19	2	33[e] (19)	0.011
Seizures	12	12	14	38 (19)	0.076
Cough	2	8	14	24 (12)	< 0.001
Rhinorrhea	4	7	2	13 (6)	0.810
Otorrhea	2	3	3	8 (4)	0.511
Hemorrhagic examthem	29	24	3	56 (28)	0.001
Mucocutaneous herpes	3	25	11	39 (19)	0.001
Motor deficit	6	11	13	30 (15)	0.008
Cranial nerve palsies	2	14	12	28 (14)	< 0.001
No (%)	73 (36)	88 (44)	40 (20)	201	

[a]Grunting in 8 infants; [b]Not applicable in 29 pts; [c]Bulbing fontanelle in 3 neonates and in other 9 infants (12); [d]Not applicable in 45 pts; [e]Not applicable in 31 pts

Systemic disorders were observed in 55 patients, with multi-organ dysfunction in 34 (17%): 33 (17%) had hypotension with shock in 27 (13%), coagulation disorders in 37 (18%), respiratory failure in 29 (14%) and acute renal failure in 20 (10%) cases. Hemorrhagic rash was observed in 49 (24%). One hundred and one (50%) patients required admission to the intensive care unit with 39 (19%) needing respiratory support.

The CSF study showed that cytosis varied from 10 to 31 360 cells/μl (4,631 ± 5,736), glucose from 0 mg/dl to 272 mg/dl (40 ± 37), and proteins from 10 mg/dl to 20,000 mg/dl (568 ± 2,291).

Bacteriologic Results

The Gram-stain, performed in only 64 CSF samples, was positive in 22 (33%). The latex agglutination test, performed in 103 of the CSF samples, was positive in 65 (63.1%), while identifying *N. meningitidis* in 41, *S. pneumoniae* in 21 and *H. influenzae* in 3. Culture identification was successful in 142 patients (70.6%): *S. pneumoniae* in 55, *N. meningitidis* in 51 and other agents in 36, including 4 out of the 5 cases of *H. influenzae* meningitis; the other 32 agents identified by cultural methods were: *Staphylococcus* spp. [13], *Klebsiella* spp. [3], *Pseudomonas* spp. [3], *Streptococcus agalactiae* [2], *Streptococcus viridans* [2], *Enterobacter* spp. [2], *Streptococcus pyogenes* [1], *Enterococcus faecium* [1], *Haemophilus parainfuenzae* [1], *E. coli* [1], *Salmonella* spp. [1], *Brucella* spp. [1] and *Listeria monocytogenes* [1]. In 59 cases (29.4%) no agent was identified by culture.

CSF PCR assay was applied to 86 samples, 40 with culture negative samples and 46 CSF specimens with culture-confirmed diagnosis. In culture-confirmed cases, PCR assay was done in CSF samples of *S. pneumoniae* [20], *N. meningitidis* [13], and other agents [13] including 3 isolates of *H. influenzae*. PCR DNA amplification was positive in all culture-confirmed samples for *S. pneumoniae* (100%) and for *N. meningitidis* (100%). In two of the three cases of *H. influenzae* the method failed this agent detection. PCR was negative in all samples with a positive culture to other agents.

PCR has allowed the identification of *N. meningitidis* in 17 CSF culture-negative samples and of *S. pneumoniae* in other 16. The PCR to *H. influenzae* was negative in all these assays. In seven samples no agent was identified by PCR assay. In one of them latex agglutination was positive for *N. meningitidis*. In two cases the diagnosis was based exclusively in a positive latex agglutination test result: *H. influenzae* and *N. meningitidis* in one each. In summary, an etiologic diagnosis could be established in 177 (88%) cases, by culture methods in 142 cases and by nonculture methods in 35 cases (Table 2). In 24 (12%) patients the aetiology could not be determined including the six samples with a negative PCR assay. In those samples in which an etiology was not defined, CSF was

Table 2
Etiologic diagnosis of bacterial meningitis and method of detection.

	Culture	Culture and PCR	Only PCR	Only latex test	Total
S. pneumoniae	35	20	16		71
N. meningitidis	38	13	17	1	69
H. influenzae	3	1		1	5
Other agents	32	0			32
Total	142 (70.6%)	35 (17.4%)	177 (88%)		

purulent in all of them, with a cytosis varying from 2,100 to10,880 cells/µl (3,545 ± 1,718).

The sensitivity of culture methods was 55.6% (IC 95%: 47.7–55.6) and 43.3% (IC 95%: 34.4–43.3), respectively, for *S. pneumoniae* and *N. meningitidis*, with a specificity of 100% (IC 95%: 95–100). For the same agents, the sensitivity of PCR compared to culture was 100% (IC 95%: 85.9–100), with a specificity of 77.8% and 78.5%, respectively.

All the patients received antibiotic therapy. Dexamethasone had been administered in 87 cases (42%), for 4 days. It was prescribed in 36 (51%) of the cases of *S. pneumoniae* meningitis, in 28 (41%) of the *N. meningitidis* meningitis and in the two cases of *H. influenzae* that occurred in children, in other 12 cases with no agent identified and in 9 cases with other identified agents.

The antigenic characterization of the *N. meningitidis* strains is given in table 2. The most common antigenic combination of serogroup C was "C:2b:P1.5,2" and "C:2b:P1.2,5. For serogroup B the serotype and subtype combinations were diverse, serotype 4 being the most frequent and serotype 2a being found in two strains. Serotype C:2b was present in 87% of the strains. No serotypes C: 2a were found (Table 3).

Characterization of *S. pneumoniae* serotypes was successful only in 32 (58%) cases. Identified serotypes, their distribution by age group and inclusion in the available vaccines are in the table 4.

Antibiotic susceptibility tests revealed that six (11%) isolates of *S. pneumoniae* were resistant to penicillin, three (14, 19A, 19A) had intermediate resistance (MIC ≥ 1 µg/ml) and other three (14, 19A, 23F) were highly resistant (MIC ≥ 2 µg/ml); four isolates (14, 23F and two 19A) were in addition resistant to cefotaxime (MIC ≥ 1–2 µg/ml) and one (23F) to levofloxacin (MIC ≥ 16 µg/ml).

The six patients infected with pneumococcal strains with high or intermediate grade resistance to penicillin recovered; three were treated with ceftriaxone and vancomycin, two with ceftriaxone and ampicillin and the remaining one with ceftriaxone.

All the isolates of *N. meningitidis* were susceptible to the antibiotics frequently used in its treatment and prophylaxis.

Eighteen patients died (8.9%). The time of death ranged between a few hours after admission to 22 days: *S. pneumoniae* was the etiologic agent in six cases, *Staphylococcus* spp. in 4, *N. meningitidis* in 3, *S. pyogenes*, *K. pneumoniae* and *E. coli* in a patient each. No agent was identified in the other two cases. Death occurred after day 14 in four of the cases (*S. pneumoniae* in two, *Staphylococcus* spp. and *K. pneumoniae* in one case each), associated with acute myocardic

Table 3
Sero-subtype by serogroups of *N. meningitidis*.

Serogroup/ serotype	C	B	W135	Y	Total
2b	n = 27 P1.2,5 (10) P1.5,2 (8) P1.5 (2) NST (1)	n = 1 NST (1)			n = 28 P1 (26) NST (2)
2a		n = 2 P1.5 (1) NST (1)	n = 3 P1.2,5		n = 5 P1(4) NST (1)
4	n = 1 P1.1 (1)	n = 9 P1.14 (2) P1.15 (2) P1.4 (1) P1.4,9 (1) NST (3)			n = 10 P1 (7) NST (3)
1		n = 7 P1.13 (1) P1.9 (1) NST (5)		NT P1.4	n = 8 P1 (3) NST (5)
Total	28	19	3	1	51

NST: non serotypable

Table 4
Serotype distribution by age group of *S. pneumoniae*.

Serogroup or serotype	≤ 2 years	> 2 to ≤ 5 years	> 5 to ≤ 18 years	> 18 to < 60 years	≥ 60 years	Total of strains
3				2[a,c]		2
4				1[a,b]	1[a,b]	2
6A		1		1	1	3[d]
6B				1[a,b]		1
6N				1		1[d]
9N				1[a]	1[a]	2
14				1[a,b]	1[a,b]	2
15C				1		1[d]
19A	2[a]			2[a]		4
21				1		1[d]
23B			1			1[d]
23F		3[a,b]		2[a,b]		5
24				1		1[d]
29		1		1	1	3[d]
33			1			1[d]
35					1	1[d]
35F			1			1[d]
Total	2	5	3	16	6	32

[a]Present in PPV23; [b]Present in 7V-PnC and in 9V-PnC; [c]Present in 11V-PnC; [d]Not contained in the PPV23 nor in any of the conjugated vaccines

Table 5
Unfavorable outcome and clinical and demographic variables.

Variables	Crude OR	IC 95%	p*
Sex			
Male	1		
Female	0.64	0.24–1.7	0.45
Age (years)			
< 5	1		0.029
5–50	1.36	0.31–5.86	
> 50	7.07	1.12–27.41	
Comorbidity			
No	1		
Yes	3.3	1.13–9.68	0.020
Agent[a]			
S. pneumoniae		1	
N. meningitidis	0.41	0.10–1.6	
Other agents	1.68	0.01–3.7	
No agent	0.43	0.54–5.1	
Neurological complications			
No	1		
Yes	1.69	0.60–4.79	0.38
Systemic complications			
No	1		
Yes	5.8	2.1–16.0	0.001
Shock			
No	1		
Yes	5.6	1.9–16.4	0.003
Renal failure			
No			
Yes	12.9	4.2–39.3	< 0.001
Coagulopaty			
No			
Yes	3.3	1.2–9.3	0.024
Respiratory failure			
No			
Yes	13.5	4.6–39.0	< 0.001

*p < 0.05; [a]Indifferent to identification method

infarction, pulmonary thromboembolism, multiorgan dysfunction and nosocomial pneumonia and intracranial hypertension and cerebral infarct. SAPS II varied from 9 to 81, with a mean value of 26 in survivors and 48 in patients who died.

Mortality was 8% in pneumococcal meningitis and 4% in meningococcal meningitis. To evaluate factors associated with an unfavorable outcome some variables were compared (Table 5). In a univariate analysis, age more than 50 years (OR 7.7; 95% CI 1.12–27.4), presence of a subjacent disease (OR 3.3; 95% CI 1.1–9.6) and the occurrence of systemic complications (OR 5.8; 95% CI 2.1–16.0) were associated with an adverse outcome. When we stratified this by agents, the neurological dysfunction was statistically associated with poor outcome in meningitis by *N. meningitidis* (OR 64; 95% CI 3.9–1034.3) and for the other agents no statistical association was found (*S. pneumoniae*: OR 0.54; 95% CI 0.09–3.04; other agents OR 0.40; 95% CI 0.04–3.8). A SAPS II value above 30 in the first 24 h was predictive of death (OR 7.8; 95% CI 2.2–26.4).

Discussion

This study includes 201 cases of bacterial meningitis selected in emergency department according to clinical picture and CSF study. Posterior data were collected from medical records during hospitalization, inducting some limitations in data performance. Anyway the most common symptoms were the classic ones: fever, headache, neck stiffness and change in mental status. We found some differences in clinical presentation by age group as shown in table 1. The study included only four neonates and the alerting symptoms were lethargy, grunting, and a bulbing fontanelle.

Diagnosis was defined by CSF and/or blood culture in 142 (70.6%) of the samples.

Thirty-two (16%) of the patients had taken antibiotics before diagnosis, but only ten of those had negative cultures. A noncultural identification can be due to several factors mainly previous antimicrobial therapy and including for CSF, the time lapse between the lumbar puncture and the smear of the samples. CSF requires immediate processing, otherwise the agents will die [22]. We think that this last point is relevant to our hospitals because it is frequent that samples are collected but not immediately processed.

This enhances the importance of Gram-stain immediately after CSF collection, because it can be very helpful in empiric antibiotic prescription when positive. The same was true for latex agglutination test, a method no longer currently used, but done at the beginning of the study in 103 of the samples, being positive in 65 assays. This test alone defined the aetiology of two additional cases, one *H. influenzae* and one *N. meningitidis*. In 59 (29.4%) of the cases no agent could be identified using cultural methods.

Unfortunately it was not possible to apply PCR assay in all negative cultures because some samples were lost. So, it was only possible to apply the assay to 40 (68%) negative CSF cultures with an agent definition in 33 of the samples: *N. meningitidis* in 17 and *S. pneumoniae* in 16. The application of PCR to culture-confirmed CSF samples of *S. pneumoniae* and *N. meningitidis* was positive in all of the cases (100%). In the three cases of *H. influenzae* the method failed to detect the agent in two, and although the low number of *H. influenzae* strains does not allow any conclusions, we think that for *H. influenzae* another target detecting capsulated and non-capsulated strains must be sought [23, 24]. If we consider culture the gold standard, the sensitivity of PCR for *S. pneumoniae* and *N. meningitidis*, was 100% (OR 95%: 85.9–100) for both of them with a specificity of 77.8% and 78.5%, respectively. PCR made possible an agent definition in about one quarter of the cases of *N. meningitis* and *S. pneumoniae* meningitis.

With the concurrent use of all these different methods, an etiologic diagnosis was obtained in 177 (88%) cases of meningitis.

Confirmation of the etiologic diagnosis is extremely important not only for a correct treatment, but also for a continuous knowledge of epidemiological evolution of meningitis agents. This can no longer be based on classic bacteriological methods and the use of real time PCR and sybergreen can be a versatile and easy way to achieve a better insight into this serious condition.

In Portugal, as in other European countries, *N. meningitis* was previously the most common etiology of meningitis, accounting for about 55% of the cases in all age groups, followed by *S. pneumoniae* (23%) and *H. influenzae* (12%) [25]. In the present study, *S. pneumoniae* was the most frequent agent, but with an incidence similar to that of *N. meningitidis*. We identified five cases of *H. influenzae* meningitis, but only two in 22-month-old and 2-year-old children, both serotype b. Both children had been previously inoculated with the Hib vaccine and no immunosuppressive condition was detected. As previously observed in other studies *S. pneumoniae* is nowadays the most frequent agent of bacterial meningitis and the mean age of incidence has shifted toward adult age [26]. Seventy-eight per cent of the cases of meningitis by *N. meningitidis* occurred in children younger than 15 years and 73.2% of *S. pneumoniae* meningitis occurred in adults.

In the present study antibiotic susceptibility tests for *S. pneumoniae* showed that all but six of the strains (11%) were susceptible to penicillin. These strains are included in the most common drug-resistant serotypes [16, 27].

Although the incidence of infections by *S. pneumoniae* resistant to penicillin and other β-lactam antibiotics varies geographically, resistance is increasing in Europe, particularly in France, Spain and in the Eastern European countries [11, 28, 29]. Currently it has not yet become a serious problem in Portugal, but a continuous vigilance is mandatory.

Conjugated vaccines for the two most common agents have been included in the immunization programs of many European countries more recently [30]. This enhances the importance of a correct epidemiologic local knowledge and of a sustained surveillance, namely after vaccine introduction.

Antigenic characterization of *S. pneumoniae* isolates included all age-groups, although with the predominance of adults (Table 3). Four out of the seven children younger than 5 years of age had received the commercially available 7V-PnC vaccine, but the strains responsible for their meningitis, 19A (n = 2), 6A and 29, are not included in this vaccine. The prevalence and serotype distribution in our investigation has shown that 14 (43.8%) of the identified strains are not included in the current formulation of PPV23; 68.8% are not included in the 7V-PnC and 62.5% are not present in the 11V-PnC vaccines, which is a lower rate of coverage than that observed in other European studies. This part of the study is limited and not conclusive as it was only possible to define serotypes in 32 (58%) of the 55 strains identified by culture because a technical problem occurred affecting some strains stored.

Another Portuguese study concerning serotype distribution from all *S. pneumoniae* isolates recovered between the years 1999 and 2002, revealed that the potential coverage of the 7V-PnC vaccine would be about 63% among infants [31]. So, in our population, conjugated and non-conjugated pneumococcal vaccines could have a lower potential for protection when compared to other European studies and a continuous surveillance program must go on.

As far as antigenic characterization of *N. meningitidis* is concerned, the more common phenotypes were the C:2b.P1,2,5 and C:2b.P1,5,2, a different result from that observed nowadays in the majority of European countries, where the incidence of phenotype C:2a.P1.5 predominates [32–34]. It was also interesting the finding of W135:2a:P1.5,2 strain in three patients, the same antigenic combination found in strains related to the Haj 2000 outbreak [35] and sporadically found in Europe. Thus, in spite of the proximity of our metropolitan area, situated in the north of Portugal, with European regions having a different epidemiological pattern, the epidemiologic pattern found in this study is still similar to what was observed in the last decade in neighboring countries. Now, that meningococcal C conjugated vaccine has been added to our immunization programme, it is important to maintain the surveillance to detect an eventual antigenic shift of *N. meningitidis* phenotypes.

According to guidelines published, dexamethasone 0.6 mg/kg/day in children and 15 mg/day–20 mg/day in adults, beginning either before or simultaneously with the first dose of antibiotic, would be recommended in 142 of the cases (in 71 *S. pneumoniae*, 69 *N. meningitidis* and in two *H. influenzae*). With all the knowledge about the efficacy of dexamethasone in adjuvant treatment of meningitis, it has been only prescribed in 46% of these cases, because in the emergency department antibiotics were administered before the diagnosis of meningitis, and after then dexamethasone was no more indicated [36]. In the randomized, double-blind, multicenter trial of adjuvant treatment with dexamethasone in adult meningitis, the mortality in dexamethasone group was 7% vs 15% in placebo group [37]. A recent review of randomized controlled trials to examine the efficacy and safety of adjuvant corticosteroid therapy in acute bacterial meningitis shows a mortality of 13.4% vs 16.1% [38].

The mortality rate in our study (8.9%) and the fact that more than 50% of the patients for whom dexamethasone would be indicated have not done it, do not allow for any conclusion about the use of steroids in our cohort. Mortality rate as per our study was lower when compared to other data from the literature, namely the mortality of *S. pneumoniae* meningitis (8%). Traditionally, we have a

lower mortality rate of bacterial meningitis than that referred to in literature and we cannot explain this difference. In a retrospective study we carried out some years ago, including 366 patients of all age groups, admitted in the ICU of our Infectious Disease Service between 1988 and 1997, 48 fatalities occurred (13%) [39].

The univariate statistical analysis done to evaluate predictive factors associated with prognosis (Table 5) show that age, the presence of at least a comorbidity and the occurrence of systemic complications were associated with an adverse outcome. Only in *N. meningitidis* meningitis neurological complications were statistically associated with a higher mortality. SAPS II score at admission was also predictive of a high risk of death [40].

A revision of literature studies about the prognostic factors in meningitis revealed that the majority of them are retrospective using different variables, which makes it difficult to compare conclusions. We found a European prospective study of *van de Beek* et al. [2] with the aim of evaluating prognostic aspects, in which the authors have concluded that risk factors for an unfavorable outcome were associated with the presence of a systemic compromise, a low level of consciousness and infection by *S. pneumoniae*. This study also highlights the high mortality due to of meningitis in developed countries.

Our study included all patients coming from the community and admitted in the emergency department where the Manchester triage system is now used and infectious diseases team is called later on, if necessary. So, the frequent administration of antibiotics before diagnosis of meningitis in critically ill patients was a limitation, thus impairing the later recommendation of corticosteroids in treatment support. We are conscious that the application of this protocol is difficult to overcome in this setting.

Acknowledgments

The present study was partially supported by a grant of the Calouste Gulbenkian Foundation.

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