Enferm Infecc Microbiol Clin. 2021;39(1):25–28

Enfermedades Infecciosas y Microbiología Clínica

www.elsevier.es/eimc

Original brief

Evolution of the distribution of *Streptococcus pneumoniae* serotypes isolated in pleural fluid in the Madrid Autonomous Community between the years 2007–2018☆

Juan Carlos Sanz[a,*], Sara de Miguel[b], María Ordobás[b], Luis García Comas[b]

[a] *Unidad de Microbiología Clínica, Laboratorio Regional de Salud Pública, Dirección General de Salud Pública, Comunidad de Madrid, Centro de Especialidades Médicas Vicente Soldevilla, CIBER de Epidemiología y Salud Pública (CIBERESP), Madrid, Spain*
[b] *Servicio de Epidemiología, Dirección General de Salud Pública, Comunidad de Madrid, Madrid, Spain*

ARTICLE INFO

Article history:
Received 5 January 2020
Accepted 8 February 2020
Available online 28 November 2020

Keywords:
Streptococcus pneumoniae
Serotypes
Pleural fluid

ABSTRACT

Introduction: The aim of this study was to describe the distribution of *Streptococcus pneumoniae* serotypes in isolates from pleural fluid in the Madrid Autonomous Community between the years 2007–2018.
Methods: Invasive pneumococcal disease strains isolated during the period 2007–2018 were studied. The frequency of serotypes from pleural fluid was compared with that observed in other samples.
Results: A total of 6,115 pneumococcal invasive isolates were processed. Of them, 182 (3%) were isolated from pleural fluid. A total of 70.9% of isolates belonged to some of the following six serotypes: 1, 3, 19A, 8, 7F and 5. The serotypes 3 and 8 increased significantly from 9.6% to 30.8%, and from 5.3% to 20.5%, respectively, over the periods 2007–2010 to 2015–2018.
Conclusions: Pneumococcal serotypes 3 and 8 are currently significant causes of infection of pleural fluid in our region.

Evolución de la distribución de serotipos de *Streptococcus pneumoniae* aislados en líquido pleural en la Comunidad de Madrid entre los años 2007 y 2018

Palabras clave:
Streptococcus pneumoniae
Serotipos
Líquido pleural

RESUMEN

Introducción: El objetivo de este estudio es describir la distribución de serotipos de *Streptococcus pneumoniae* en aislados de líquido pleural en la Comunidad de Madrid (CM) entre los años 2007–2018.
Métodos: Se estudiaron las cepas de episodios de enfermedad neumocócica invasora aisladas en la CM durante el periodo 2007–2018. La frecuencia de serotipos en líquido pleural se comparó con la observada en otras muestras.
Resultados: Se procesaron 6.115 cepas invasoras de neumococo. Ciento ochenta y dos (3%) se aislaron en muestras de líquido pleural. El 70,9% de los aislados pertenecía a alguno de los 6 siguientes serotipos: 1, 3, 19A, 8, 7F y 5. Los serotipos 3 y 8 aumentaron significativamente, pasando del 9,6% al 30,8%, y del 5,3% al 20,5%, respectivamente, entre los períodos 2007–2010 a 2015–2018.
Conclusión: Los serotipos 3 y 8 son actualmente causas importantes de infección del líquido pleural en nuestra área.

DOI of original article: https://doi.org/10.1016/j.eimc.2020.02.021
☆ Please cite this article as: Sanz JC, Miguel S, Ordobás M, García Comas L. Evolución de la distribución de serotipos de *Streptococcus pneumoniae* aislados en líquido pleural en la Comunidad de Madrid entre los años 2007 y 2018. Enferm Infecc Microbiol Clin. 2021;39:25–28.
* Corresponding author.
E-mail address: juan.sanz@salud.madrid.org (J.C. Sanz).

Table 1
The most common serotypes of *Streptococcus pneumoniae* isolated in pleural fluid in cases of invasive pneumococcal disease. Autonomous Community of Madrid 2007–2018.

	Origin of the strain			
	Typically sterile samples	Pleural fluid		
Serotype	No. of strains	n	(%)	χ^2
1	467	40	8.57	p <0.001
3	584	35	5.99	p <0.001
19A	535	21	3.93	NS
8	858	16	1.86	NS
7F	345	9	2.61	NS
5	143	8	5.59	NS

Introduction

Invasive pneumococcal disease (IPD) is microbiologically defined as detection of *Streptococcus pneumoniae* in typically sterile clinical samples. Although there are many *S. pneumoniae* serotypes, only a limited number of them cause most infections. Distribution of pneumococcal serotypes varies by disease, the type of clinical sample studied and geographical region. *S. pneumoniae* causes a broad spectrum of diseases and represents a significant cause of empyema.[1]

The greater or lesser tropism of particular strains for pleural fluid can cause the frequency with which different serotypes are detected to vary in cases of empyema. Serotypes isolated in pleural fluid may be different depending on vaccination policies. The 7-valent pneumococcal conjugate vaccine (PCV7) to protect against serotypes 4, 6B, 9V, 14, 18C, 19F and 23F was placed on the Spanish market in 2001. In 2006, the Autonomous Community of Madrid added this vaccine to its childhood vaccination schedule, and in 2010, it replaced it with the PCV13 vaccine, which added serotypes 1, 3, 5, 6A, 7F and 19A to the PCV7 vaccine. In 2012, the PCV13 vaccine was removed from that vaccination schedule and continued to be recommended on an individual basis. In 2015, the Autonomous Community of Madrid re-added PCV13 to its childhood vaccination schedule.[2]

IPD is a reportable disease in the Autonomous Community of Madrid, and the Laboratorio Regional de Salud Pública [Regional Laboratory of Public Health] acts as a reference centre for serotyping and monitoring antibiotic susceptibility. The objective of this study is to report changes in the distribution of serotypes in isolates from pleural fluid and their tropism for this type of sample in patients of all ages in the Autonomous Community of Madrid between 2007 and 2018.

Methods

Strains from episodes of IPD were examined in samples sent by public and private hospitals in the Autonomous Community of Madrid to the Laboratorio Regional de Salud Pública between 2007 and 2018. Serotyping was performed by means of latex agglutination (Pneumotest-Latex) and the Quellung reaction, using specific antisera (Statens Serum Institut, Copenhagen, Denmark). Determination of antibiotic susceptibility to penicillin, erythromycin and levofloxacin was done by ETEST (bioMérieux, France) according to EUCAST criteria for interpretation[3] (minimum inhibitory concentration [MIC] against penicillin ≤0.06 mg/l susceptible and >2 mg/l resistant; erythromycin ≤0.25 mg/l susceptible and >0.5 mg/l resistant and levofloxacin ≤0.001 mg/l susceptible and >2 mg/l resistant). The frequency of serotypes from pleural fluid samples was compared to that seen in other samples that are typically sterile (such as blood, cerebrospinal fluid, as citic fluid, synovial fluid, pericardial fluid, etc.) using the χ^2 test and the *odds ratio* (OR) with a 95% confidence interval. Changes over time (across the four-year periods of 2007–2010, 2011–2014 and 2015–2018) were analysed using the χ^2 test for linear trend.

Results

Of the 6,115 invasive pneumococcal strains that were processed, 182 (3%) were isolated in samples of pleural fluid (70 in children and 112 in adults). Of these strains, 95.6% were identified at the serotype level (27 different serotypes) and 4.4% were only identified at the serogroup level (3 different serogroups). In 70.8% of pleural fluid isolates, at least one of the following 6 serotypes were identified: 1 (22.0%), 3 (19.2%), 19A (11.5%), 8 (8.8%), 7F (4.9%) and 5 (4.4%). Among them, 2 serotypes appeared more frequently in pleural fluid than in other locations (p <0.001), with higher tropism for this location: 1 (OR: 3.63; 95% CI: 2.52–5.23) and 3 (OR: 2.33; 95% CI: 1.60–3.41). Table 1 shows the proportions of strains isolated in pleural fluid in the most common serotypes in this sample out of all cases of IPD. Serotypes 1, 3 and 5 were isolated in pleural fluid in more than 5% of all strains causing IPD. The total frequency of strains isolated in pleural fluid decreased over the course of the study period (p < 0.001) (Table 2). Among the 6 most frequent serotypes, 1, 5, 19A and 7F decreased (there were no isolates of serotypes 1, 5 or 7F and only one isolate of 19A from 2015 to 2018) and the reduction in the frequency (in terms of proportions) achieved statistical significance in the first 2. Conversely, serotypes 3 and 8 showed a statistically significant increase in frequency (reaching percentages in pleural fluid of 30.8% and 23.1%, respectively, in 2015–2018). Both serotypes were isolated, particularly in adults (for serotype 3, 21 strains were isolated in adults out of a total of 35; for serotype 8, 14 strains were isolated in adults out of a total of 16).

Among the 182 strains isolated in pleural fluid, 37 (20.3%) demonstrated intermediate susceptibility to penicillin and 3 (1.6%) were resistant to it; 2 (1.1%) presented intermediate susceptibility to erythromycin and 47 (25.8%) were resistant and, lastly, 8 (4.4%) were resistant to levofloxacin. Of the isolates of serotype 19A, 20 (95.2%) were not susceptible to penicillin (13 of them [61.9%] were isolated in 2007–2010 and only 1 [4.8%] was isolated in 2015–2018), and all were resistant to erythromycin. In serotype 8, 4 (25.0%) of the strains were resistant to erythromycin and 3 (18.8%) of the strains were resistant to levofloxacin.

Discussion

In the era prior to the use of conjugate vaccines (1997–2000), the serotypes covered by PCV7 accounted for half to one-third of isolates in pleural fluid in Spain. During the PCV7 post-marketing period (2001–2008), Spain saw a significant decrease in these serotypes in pleural fluid and an increase in others, such as 1, 3 and 19A,[4] which were not included in PCV7 but were included in

Table 2
Changes in the main serotypes of *Streptococcus pneumoniae* isolated in pleural fluid in cases of invasive pneumococcal disease. Autonomous Community of Madrid. 2007-2010, 2011-2014 and 2015-2018.

Serotypes		Total	2007–2010	2011–2014	2015–2018	*p*[a] trend
	n	182	94	49	39	
	%	3.0	3.8	3.2	1.9	<0.01
	OR[b]		1	0.82	0.48	
	n	40	32	8	0	
1	%	22.0	34.0	16.3	0.0	<0.01
	OR[c]		1	0.38	–	
	n	35	9	14	12	
3	%	19.2	9.6	28.6	30.8	<0.01
	OR[c]		1.0	3.8	4.2	
	n	21	14	6	1	
19A	%	11.5	14.9	12.2	2.6	0.054
	OR[c]		1	0.8	0.15	
	n	15	5	2	8	
8	%	8.2	5.3	4.1	23.1	<0.01
	OR[c]		1	0.76	5.34	
	n	9	5	4	0	
7F	%	4.0	5.3	8.2	0.0	NS
	OR[c]		1	1.58	–	
	n	8	8	0	0	
5	%	4.4	8.5	0.0	0.0	<0.05
	OR[c]		1	0	0	
	n	182	94	49	39	
Total	%	3.0	3.8	3.2	1.9	<0.01
	OR[c]		1	0.82	0.48	

[a] χ^2 of linear trend.
[b] *Odds ratio* with respect to the total number of serotypes causing invasive pneumococcal disease.
[c] *Odds ratio* with respect to the total number of serotypes isolated in pleural fluid.

PCV13 and seem to show a marked tropism for pleural tissue.[5] Other serotypes covered by PCV13, such as 5 and 7 F, have also been predominant in this type of infection.[6] In the pre-PCV13 era, the serotypes included in this vaccine represented the most common cause of paediatric parapneumonic effusion.[5,7] After PCV13 was introduced, a reduction in pleural fluid infection occurred with a drop in these vaccine serotypes and a rising trend among non-vaccine serotypes,[8] with there being no clear replacement at that time.[5]

This study showed a drop in levels of pneumococcal infection in pleural fluid since 2007. Six serotypes (1, 3, 19A, 8, 7F and 5) accounted for more than 3/4 of isolates in pleural fluid in the study period. The changes in serotypes 1 and 3 are of great significance, given that both showed higher tropism for this location. Serotype 1 presented a very marked decrease following the introduction of PCV13: it went from accounting for 1/3 of cases in 2007–2010 to accounting for no cases in 2015–2018. The significant decrease in this serotype in pleural fluid infections has also been seen in other studies.[8] Another declining serotype is serotype 5, which disappeared in 2011, perhaps due to cyclical epidemic factors and not merely as a direct result of the vaccine.[9] On the other hand, the increase in serotype 3 is striking, as it increased in frequency despite being included in the PCV13 vaccine. Some studies have suggested that PCV13 has limited efficacy against this serotype,[10] which could have accounted for this trend somehow.

Although, all things considered, a general decrease in levels of pneumococcal infection in pleural fluid has been seen since 2007, with a significant decrease in the frequency of serotype 1, the increase in serotypes 3 and 8 (with the 2 accounting for half of cases in 2015–2018) is an problem that deserves attention. These 2 serotypes have a high incidence in the Autonomous Community of Madrid, particularly among adult patients.[3] It should be noted that serotype 19A isolates in pleural fluid have a high rate of resistance to erythromycin and non-susceptibility to penicillin. This can make them more difficult to treat. Fortunately, the frequency of this serotype has been decreasing since PCV13 was introduced. That which is seen in pleural fluid is comparable to that which is seen in IPD in general, with a decrease associated with elimination of vaccine serotypes and an increase in other non-vaccine serotypes such as serotype 8.

Conflicts of interest

JCS has attended conferences funded by Pfizer.

References

1. Liese JG, Schoen C, van der Linden M, Lehmann L, Goettler D, Keller S, et al. Changes in the incidence and bacterial aetiology of paediatric parapneumonic pleural effusions/empyema in Germany, 2010-2017: a nationwide surveillance study. Clin Microbiol Infect. 2019;25:857–64.
2. Latasa Zamalloa P, Sanz Moreno JC, Ordobás Gavín M, Barranco Ordoñez MD, Insúa Marisquerena E, Gil de Miguel Á, et al. Evolución de la enfermedad neumocócica invasora y sus serotipos en la Comunidad de Madrid. Enferm Infecc Microbiol Clin. 2018;36:612–20.
3. The European Committee on Antimicrobial Susceptibility Testing. Breakpoint tables for interpretation of MICs and zone diameters. Version 9.0; 2019 http://www.eucast.org
4. Fenoll A, Aguilar L, Vicioso MD, Gimenez MJ, Robledo O, Granizo JJ, et al. Serotype distribution and susceptibility of *Streptococcus pneumoniae* isolates from pleural fluid in Spain from 1997 to 2008. Antimicrob Agents Chemother. 2010;54:5387–90.
5. Azzari C, Serranti D, Nieddu F, Moriondo M, Casini A, Lodi L, et al. Significant impact of pneumococcal conjugate vaccination on pediatric parapneumonic effusion: Italy 2006–2018. Vaccine. 2019;37:2704–11.
6. De Schutter I, Vergison A, Tuerlinckx D, Raes M, Smet J, Smeesters PR, et al. Pneumococcal etiology and serotype distribution in paediatric community-acquired pneumonia. PLoS One. 2014;9:e89013.
7. Slinger R, Hyde L, Moldovan I, Chan F, Pernica JM. Direct *Streptococcus pneumoniae* real-time PCR serotyping from pediatric parapneumonic effusions. BMC Pediatr. 2014;14:189.
8. Díaz-Conradi A, Hernández S, García-García JJ, Muñoz-Almagro C, Moraga-Llop F, Ciruela P, et al. Complicated pneumococcal pneumonia with pleural effusion or empyema in the 13-valent pneumococcal conjugate vaccine era. Pediatr Pulmonol. 2019;54:517–24.

9. Picazo J, Ruiz-Contreras J, Casado-Flores J, Negreira S, García-de-Miguel MJ, Hernández-Sampelayo T, et al. Expansion of serotype coverage in the universal pediatric vaccination calendar: short-term effects on age- and serotype-dependent incidence of invasive pneumococcal clinical presentations in Madrid, Spain. Clin Vaccine Immunol. 2013;20:1524–30.

10. Silva-Costa C, Brito MJ, Pinho MD, Friães A, Aguiar SI, Ramirez M, et al. Pediatric complicated pneumonia caused by *Streptococcus pneumoniae* serotype 3 in 13-valent pneumococcal conjugate vaccines, Portugal, 2010–2015. Emerg Infect Dis. 2018;24:1307–14.