Report of Typing & Antimicrobial Susceptibilities of Isolates Causing Invasive Pneumococcal Disease in Ireland, 2011-2013

1. Background

Streptococcus pneumoniae is a major cause of life-threatening infections such as meningitis and bloodstream infection, i.e. invasive pneumococcal disease (IPD) [1]. Pneumococcal pneumonia is also a leading cause of death in children worldwide, with approximately one million deaths in children under 5 years of age occurring annually. The population groups at highest risk of pneumococcal infection are young children and the elderly [2].

S. pneumoniae owes its success as a pathogen in part to the diversity of the circulating capsular serotypes. The recent identification of new serotypes, 6C and 6D, serologically cross reactive with the 6A and 6B population, respectively [3,4], and the identification of a new subtype (11E) within the 11A population [5], suggests that at least 94 immunologically distinct serotypes exist, based on the chemical composition of the polysaccharide capsule [2, 4]. Immune responses elicited by current pneumococcal vaccines are directed towards the polysaccharide capsule [6].

In April 2007 the National IPD Typing Project was re-established as a collaboration between the Royal College of Surgeons in Ireland (RCSI), the Children's University Hospital, Temple St. and the Health Protection Surveillance Centre (HPSC) to provide reference laboratory support for the investigation of IPD in advance of the introduction of the 7-valent pneumococcal conjugate vaccine (PCV7). Currently, funding is provided by the HPSC on a year-to-year basis, supplemented by some support from commercial

sources, but with no guarantee of ongoing funding to ensure continuation of this very important work. The laboratory was recently accredited by Clinical Pathology Accreditation (CPA, UK) and it also participates in the IBD-Labnet external quality assurance scheme for *S. pneumoniae,* which is under the auspices of the European Centre for Disease Control.

In September 2008, PCV7 was introduced to the Irish infant immunisation schedule at 2, 6 and 12 months of age. In December 2010 PCV7 was replaced by the 13-valent pneumococcal conjugate vaccine (PCV13), which contains serotype 1, 3, 5, 7F, 6A and 19A in addition to the PCV7 serotypes. The objective of the surveillance programme is to monitor the serotype distribution of invasive *S. pneumoniae* isolates in Ireland and to ascertain the impact these vaccines have on the burden of IPD in this country. This report describes updated data up and including 2013.

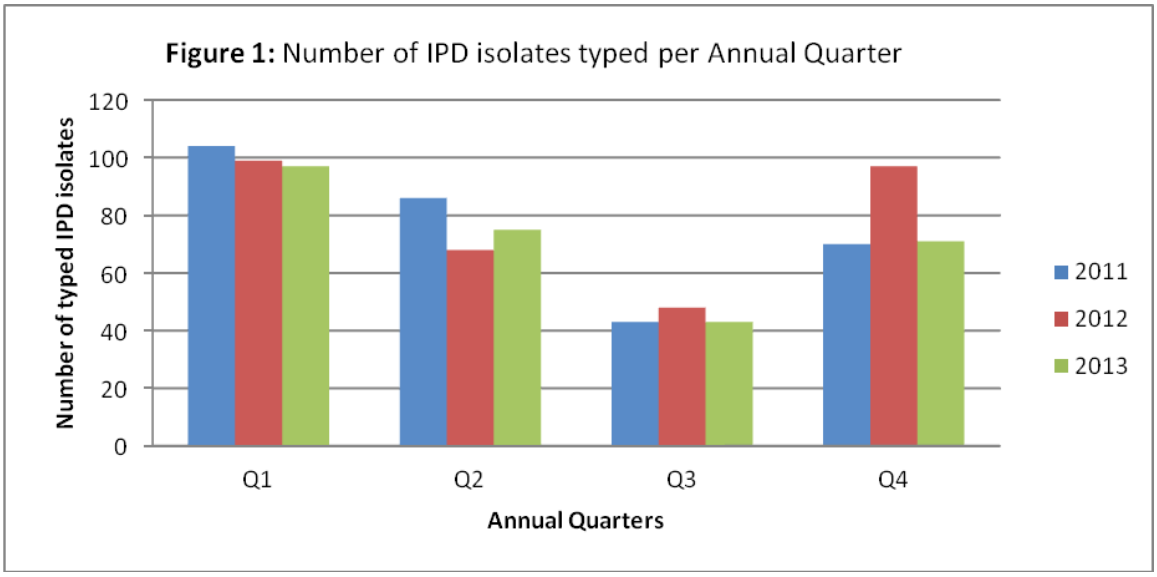
Invasive *S. pneumonia*e isolates are forwarded by clinical microbiology laboratories to RCSI Education and Research Centre, Beaumont Hospital and/or the Epidemiology and Molecular Biology Unit (EMBU) in Temple Street Children's Hospital for typing. Typing is performed using a combination of serological co-agglutination using antisera from the Statens Serum Institute (DK-2300 Copenhagen S, Denmark) and multiplex PCR, as previously described [7]. Penicillin susceptibility is assessed using the E-test method (bio Mérieux, Marcy l'Etoile, France), and results interpreted following Clinical Laboratory Standards Institute (CLSI), USA, guidelines M100-S23 [8]. For epidemiological purposes, reduced susceptibility to penicillin is defined using the CLSI breakpoints as MIC≥0.12mg/L [8].

2. Overview of Surveillance Data

Fewer invasive pneumococcal disease (IPD) isolates were received for serotyping in 2013 (*n*=305), in comparison to 2012 (*n*=314) and 2011 (*n*= 308). In 2013, 19 (6.2%) of the isolates were not culturable on receipt (no growth of the organism from the sample or *Streptococcus pneumoniae* not indentified in culture), compared to 2 and 5 in 2012 and 2011, respectively. The number of isolates typed were 286 in 2013, in comparison to 312

and 303 in 2012 and 2011, respectively. The isolates typed and categorized per quarter are displayed in **Figure 1**.

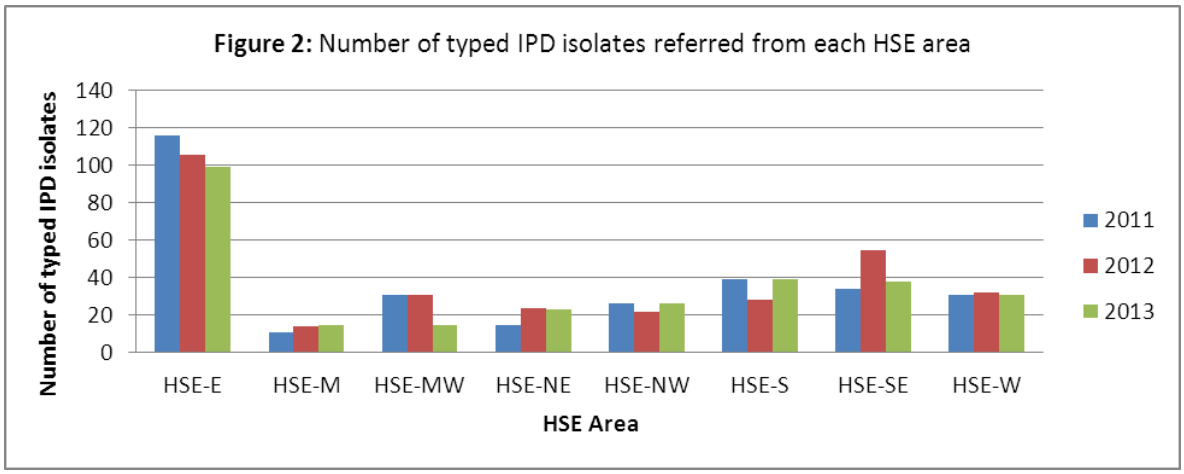
Figure 1. Number of IPD isolates typed per quarter (2011-2013)



The highest number of isolates were received in quarter 1 (January – March) and quarter 4 (October – November) of both years, most likely reflecting seasonal variation in IPD incidence (**Figure 1**). Significantly fewer isolates were received in quarter 3 than all other quarters. The largest proportion of IPD-related typed isolates were recovered from blood samples for 2013 (*n*=277/286, 96.9%) and 2012 (*n*=303/312, 97.1%). There was no significant difference between the number of IPD isolates typed from male and female patients in the past 3 years (2011-2013).

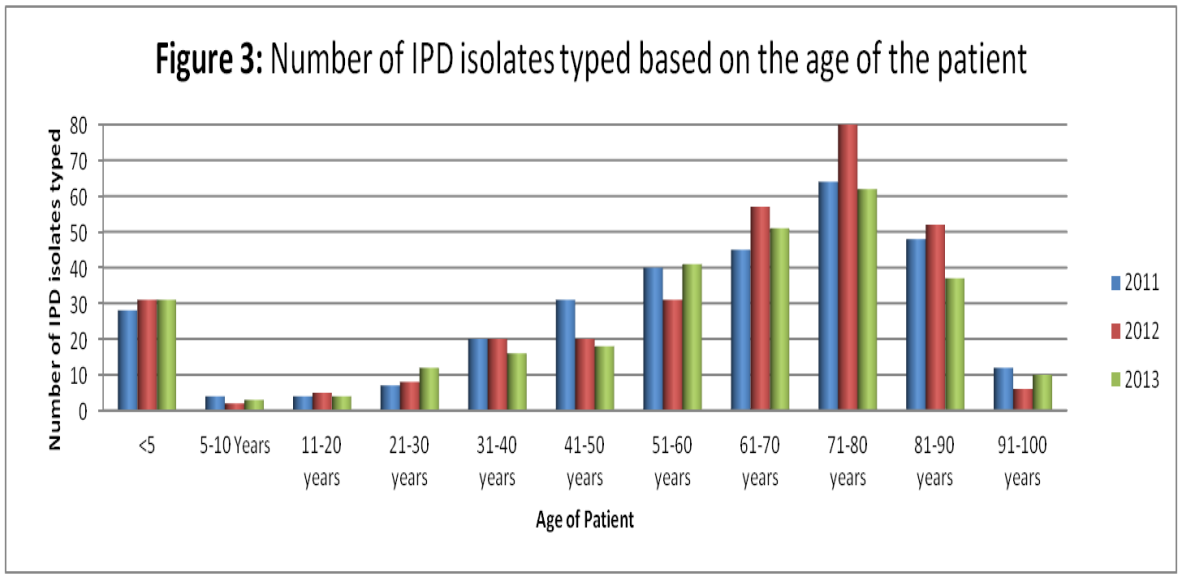
The number of IPD isolates referred to the laboratory for typing in each HSE area is shown in **Figure 2**. There is no apparent trend over the 3 years. The number of isolates referred from each HSE area is most likely associated with the population in each area, as more isolates were received from the HSE areas - East, South-East and South.

Figure 2. Number of IPD isolates referred from each HSE area (2011-2013)



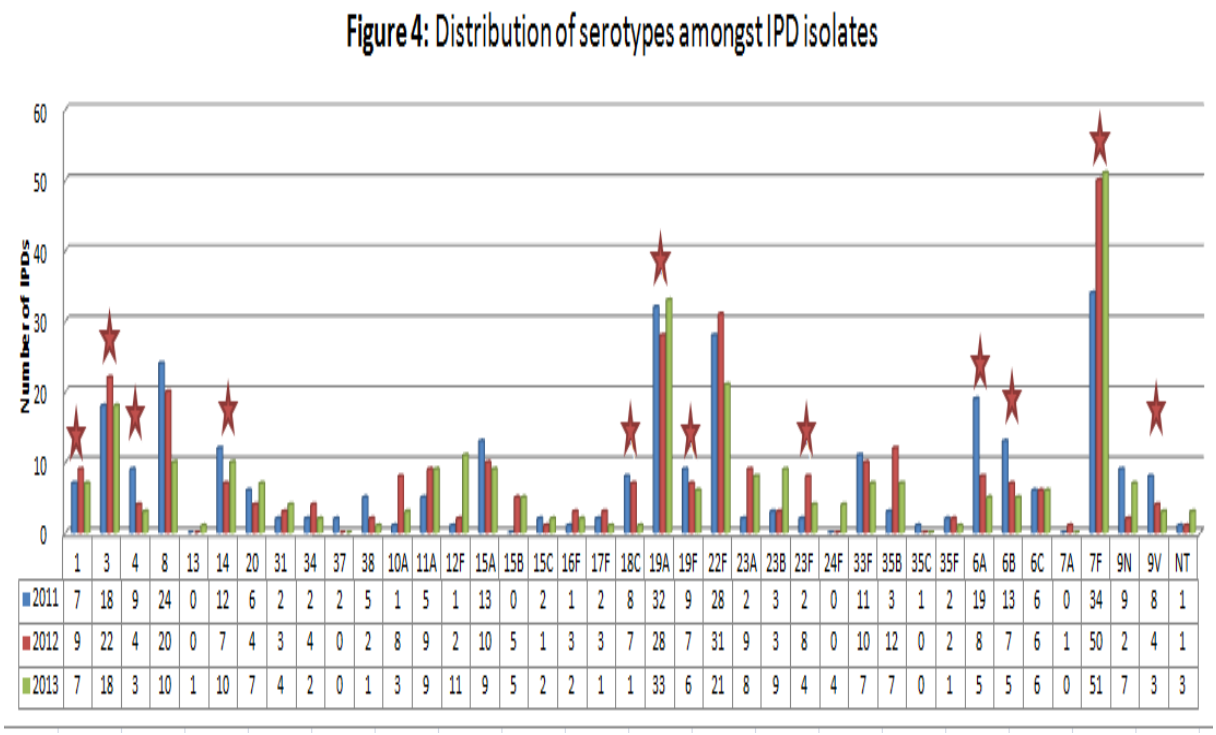
There was a slight increase in the number of isolates referred from children under 5 years of age in 2013 and 2012 (*n*=31 cases) in comparison to 28 isolates received in 2011 (**Figure 3)**. As in 2011 and 2012, the number of IPD's reported in elderly (aged 71-80 *n*=62) remains high, in comparison to young children (<5 years old), *n*=31 in 2013.

Figure 3. Number of IPD isolates based on the age of the patient (2011-2013)



As displayed in **Figure 4**, the four serotypes most commonly identified in 2013 were 7F, 19A, 22F and 3, similar to 2012 when 7F, 22F, 19A and 3 were the most common. There were no significant increases in the frequency of any particular serotypes identified in 2013 in comparison to 2012 or between 2013 and 2011. The frequency of some serotypes (37, 4, 6A, 6B, 9N) fell in 2012 and 2013, in comparison to 2011. Serotypes 4, 6A and 6B are included in PCV13, which may suggest that vaccination may have contributed to a reduction in the frequency of theses serotypes associated with IPD. However, serotypes 37 and 9N are not included in PCV13 but the frequency of these serotypes also fell.

Figure 4. Distribution of serotypes amongst IPD typed isolates (2011-2013)



	1	3	4	8	13	14	20	31	34	37	38	10A	11A	12F	15A	15B	15C	16F	17F	18C	19A	19F	22F	23A	23B	23F	24F	33F	35B	35C	35F	6A	6B	6C	7A	7F	9N	9V	NT
2011	7	18	9	24	0	12	6	2	2	2	5	1	5	1	13	0	2	1	2	8	32	9	28	2	3	2	0	11	3	1	2	19	13	6	0	34	9	8	1
2012	9	22	4	20	0	7	4	3	4	0	2	8	9	2	10	5	1	3	3	7	28	7	31	9	3	8	0	10	12	0	2	8	7	6	1	50	2	4	1
2013	7	18	3	10	1	10	7	4	2	0	1	3	9	11	9	5	2	2	1	1	33	6	21	8	9	4	4	7	7	0	1	5	5	6	0	51	7	3	3

Indicative of serotype covered in PCV13

Figure 5 displays the distribution of serotypes covered in the PCV13 vaccine according to patient age. for the years 2011-2013. The results for the category ≤5 years old indicate that there was relatively no change in the number of serotypes 1, 2, 4, and 6A. There were no 6B types isolated in 2012-2013 in comparison to 2 in 2011. There was an increase in the

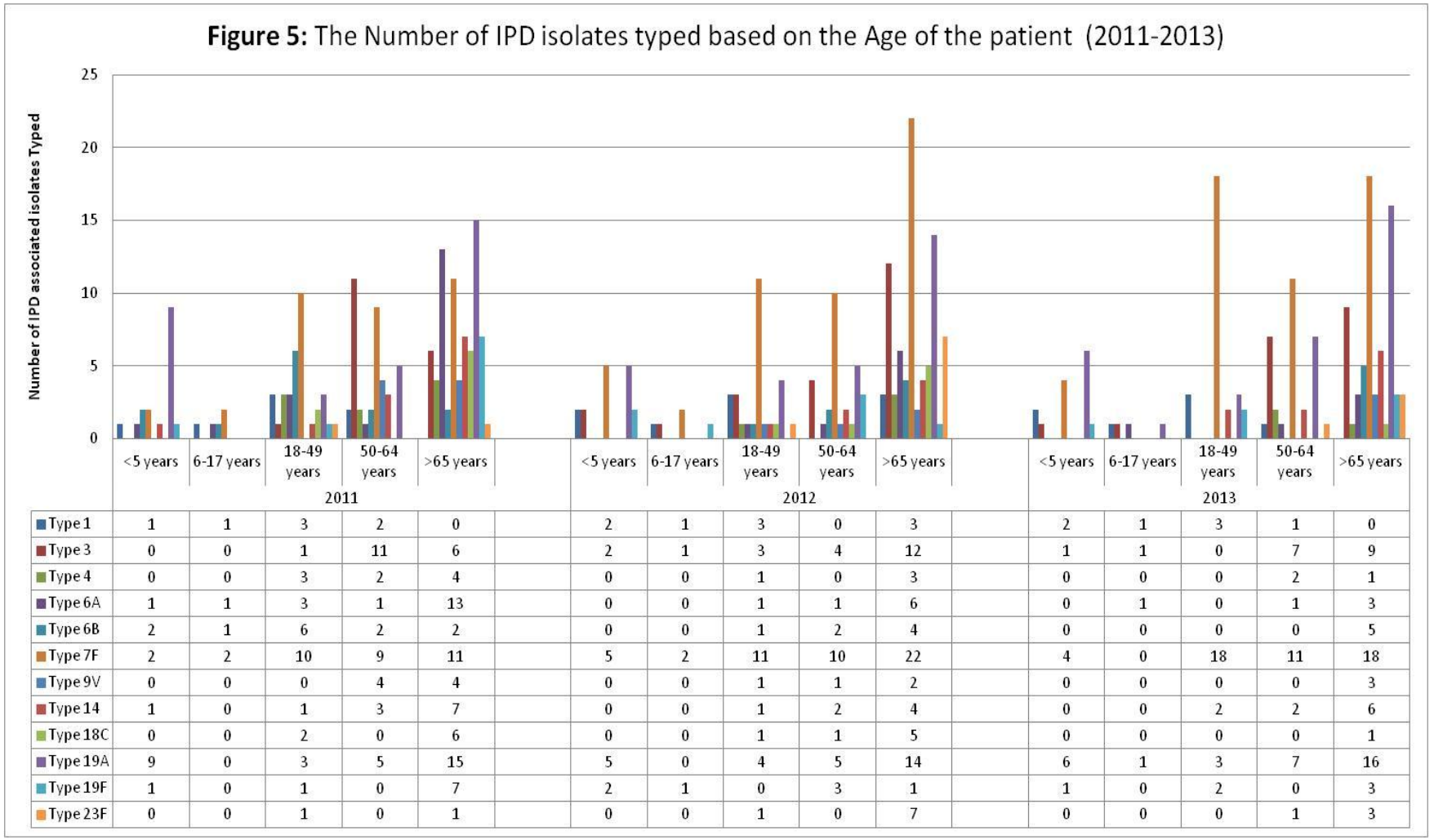
number of 7F serotypes from 2 in 2011 to 5 and 4 in 2012 and 2013, respectively. This increase was also seen in most other age categories, particularly in the age category 18-48 years old (2011 *n*=10, 2012 *n*=11, 2013 *n*=18) and the ≥65 years old category 2011 *n*=11, 2012 *n*=22, 2013 *n*=18). There was a notable reduction in the number of the serotype 6A in the age category >65 years old from 2011 (*n*=13) to 6 in 2012 and 3 in 2013. Serotype 6A is also included in the PPV23 vaccine (recommended for the elderly in Ireland). There were only marginal changes in the distribution of the other serotypes, i.e. 9V, 14, 18C, 19A, 19F and 23F.

The number of isolates with reduced susceptibility to penicillin (**Table 1**) (MIC>0.12mg/L) was lower in 2013 (18.88%, *n*=54/286) than 2012 (16.98%, *n*=53/312) and 2011 (19.14%, *n*=58/303). However, this rate is still high in comparison to other EU countries.

Table 1. The number of IPD isolates with reduced susceptibility to penicillin

	Penicillin Susceptible MIC <0.12 mg/L	**Penicillin** Reduced Susceptibility MIC >0.12 mg/L	**Total**
2011	245	58	**303**
2012	259	53	**312**
2013	232	54	**286**

Figure 5. The number of IPD isolates typed (covered in the vaccine) based on patient age (2011-2013)



	2011					2012					2013				
	<5 years	6-17 years	18-49 years	50-64 years	>65 years	<5 years	6-17 years	18-49 years	50-64 years	>65 years	<5 years	6-17 years	18-49 years	50-64 years	>65 years
Type 1	1	1	3	2	0	2	1	3	0	3	2	1	3	1	0
Type 3	0	0	1	11	6	2	1	3	4	12	1	1	0	7	9
Type 4	0	0	3	2	4	0	0	1	0	3	0	0	0	2	1
Type 6A	1	1	3	1	13	0	0	1	1	6	0	1	0	1	3
Type 6B	2	1	6	2	2	0	0	1	2	4	0	0	0	0	5
Type 7F	2	2	10	9	11	5	2	11	10	22	4	0	18	11	18
Type 9V	0	0	0	4	4	0	0	1	1	2	0	0	0	0	3
Type 14	1	0	1	3	7	0	0	1	2	4	0	0	2	2	6
Type 18C	0	0	2	0	6	0	0	1	1	5	0	0	0	0	1
Type 19A	9	0	3	5	15	5	0	4	5	14	6	1	3	7	16
Type 19F	1	0	1	0	7	2	1	0	3	1	1	0	2	0	3
Type 23F	0	0	1	0	1	0	0	1	0	7	0	0	0	1	3

3. Implications and the future

Continued surveillance of serotypes causing IPD is necessary to: monitor the epidemiology of IPD in Ireland, assess the effectiveness of the national vaccination programme in Ireland, detect the presence of non-vaccine serotypes and the emergence of replacement serotypes. Also, Ireland has a relatively high incidence of invasive isolates non-susceptible to penicillin and we need to monitor if efforts to improve antimicrobial prescribing positively impact on this.

There are other research studies underway assessing the potential impact of conjugated pneumococcal vaccines on non-invasive disease. On-going Irish data on IPD will greatly inform national health policy relating to use of conjugated vaccines in the Irish population and will be considered as new vaccines with extended valency become available.

Finally, there is an urgent need to firmly and permanently establish this important surveillance programme, which is integral to national IPD surveillance, and is part of a European initiative to monitor the effectiveness of conjugate vaccines among EU member states. Currently funding for this activity is supported by the HPSC on a year-to-year basis only, with no designated national funding. In the absence of long term assured funding and recognition that this is a national service, there are risks to the continuity of the service, and long term planning and quality improvement initiatives will be difficult to implement.

Additional information

Additional data on the impact of PCV on the burden of IPD in Ireland is available on the HPSC website, see http://www.hpsc.ie/hpsc/A-Z/VaccinePreventable/PneumococcalDisease/PostersPresentations/File,4292,en.pdf

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5. References

1. Scott JA, *et al*. Serogroup-specific epidemiology of *Streptococcus pneumoniae*: associations with age, sex, and geography in 7,000 episodes of invasive disease. *Clin Infect Dis* 1996; 22: 973-81.

2. Hausdorff WP, Feikin DR, Klugman KP. Epidemiological differences among pneumococcal serotypes. *Lancet Infect Dis* 2005; 5: 83-93.

3. Jin P, *et al*. First report of putative *Streptococcus pneumoniae* serotype 6D among nasopharyngeal isolates from Fijian children. *J Infect Dis* 2009; 200: 1375-80.

4. Park IH, *et al*. Discovery of a new capsular serotype (6C) within serogroup 6 of *Streptococcus pneumoniae*. *J Clin Microbiol* 2007; 45:1225-33.

5. Yu J, *et al*. A rapid pneumococcal serotyping system based on monoclonal antibodies and PCR. *J Med Microbiol* 2008; 57: 171-8.

6. Goldblatt D, *et al*. Antibody responses to nasopharyngeal carriage of *Streptococcus pneumoniae* in adults: a longitudinal household study. *J Infect Dis* 2005; 192 :387-93.

7. Vickers I, O'Flanagan D, Cafferkey M, Humphreys H. Multiplex PCR to determine *Streptococcus pneumoniae* serotypes causing otitis media in the Republic of Ireland with further characterisation of antimicrobial susceptibilities and genotypes. Eur J Clin Microbiol Infect Dis 2011; 30:447-53.

8. Clinical Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing; twenty-third information supplement. CLSI document M100-S23. Wayne, PA: Clinical Laboratory Standards Institute; 2013.

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