SEROTYPES AND ANTIMICROBIAL RESISTANCE AMONG NON-INVASIVE PNEUMOCOCCI IN NEW ZEALAND, 2008

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September 2009

FW09087

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ACKNOWLEDGEMENTS

Dr Susan Taylor, Bruce Dove and other staff, Diagnostic Medical Laboratory, Auckland, and Dr Rosemary Ikram and laboratory staff, Medlab South, Christchurch, who referred isolates and collected patient data for the survey.

Julie Morgan, Invasive Pathogens Laboratory, and Rosemary Woodhouse, Antibiotic Reference Laboratory, ESR, for serotyping and antimicrobial susceptibility testing.

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SUMMARY

On 1 June 2008, the 7-valent pneumococcal conjugate vaccine (PCV-7), Prevenar®, was added to the New Zealand immunisation schedule. Overseas experience has shown that this vaccine not only has an impact on the incidence of invasive pneumococcal disease (IPD) but also on other pneumococcal infections. While ESR has systematically monitored serotypes and antimicrobial susceptibility among *Streptococcus pneumoniae* causing IPD in New Zealand, there has been no routine monitoring of non-invasive *S. pneumoniae*.

The aim of this survey was to provide baseline information on the serotypes and antimicrobial susceptibility of non-invasive pneumococci in New Zealand prior to the addition of pneumococcal vaccination to the New Zealand immunisation schedule. This baseline information should facilitate future assessments of the impact of childhood pneumococcal vaccination on all pneumococcal infections.

Between April and September 2008, non-invasive *S. pneumoniae* isolates were referred to ESR from Diagnostic Medical Laboratory, Auckland (DML), and Medlab South, Christchurch (MLS). The aim was to collect approximately 200 non-repeat isolates from both laboratories, comprising 50 isolates from ears, 50 from eyes, 50 from sputum and 50 from any other sites. At ESR, the isolates were serotyped by the capsular antigen reaction. Penicillin, cefotaxime and moxifloxacin minimum inhibitory concentrations (MICs) were determined by Etest. Chloramphenicol, co-trimoxazole, erythromycin, tetracycline and vancomycin susceptibilities were determined by disc susceptibility testing. The serotype distribution and antimicrobial susceptibility among the non-invasive pneumococci included in this survey was compared with that among pneumococci causing invasive disease in 2008. The data on the isolates from invasive disease was sourced from ESR's laboratory-based surveillance of IPD.

A total of 354 non-invasive pneumococcal isolates were collected for the survey: 200 from DML and 154 from MLS. Serotype 19F and non-typable isolates were prevalent, accounting for 21.2% and 18.4% of the isolates, respectively. Non-typable isolates were associated with pneumococci isolated from the eye, with 75.4% of non-typable isolates being from this site.

Compared with the serotypes causing IPD in 2008, non-typable and serotype 19F isolates were significantly more prevalent among non-invasive pneumococci, and serotypes 1, 4 and 14 were more prevalent among invasive isolates. The same comparison for patients ≤1 year of age again showed non-typable and serotype 19F isolates were significantly more prevalent among non-invasive pneumococci, but serotypes 4, 6B and 14 were more prevalent among invasive isolates.

Forty-eight percent of all the non-invasive pneumococci were one of the serotypes included in PCV-7. This coverage increased to 61.1% for isolates from patients ≤1 year old. The proportion of invasive pneumococci in 2008 due to a PCV-7 serotype was greater at 57.1% for isolates from all patients and 83.3% for those from patients ≤1 year of age.

continued

SUMMARY *continued*

Using the Clinical Laboratory Standards Institute's (CLSI's) MIC interpretation for oral penicillin treatment, 17.2% of isolates were penicillin resistant and a further 15.3% had intermediate resistance. Resistance rates to the other antimicrobials were: cefotaxime, 5.9% (based on CLSI non-meningitis interpretation); chloramphenicol, 2.5%; clindamycin, 13.3%; co-trimoxazole, 33.6%; erythromycin, 22.9%; and tetracycline, 19.8%. All isolates were susceptible to moxifloxacin and vancomycin.

Multidrug-resistance to penicillin and at least three other antibiotics was identified in 12.7% of isolates. While resistance rates were generally higher among isolates from Christchurch, the only significant difference in resistance between isolates from the two centres was higher chloramphenicol resistance in Auckland. Resistance was generally more prevalent among pneumococci from patients ≥65 years of age, infants ≤1 year of age, and isolates from ears and sputum. Serotype 19F accounted for the majority of resistant isolates, and, among the 75 serotype 19F isolates included in the survey, 52.0% were multidrug resistant.

A comparison of resistance rates among the non-invasive pneumococci included in this survey and invasive pneumococci isolated in 2008 showed that resistance to penicillin, cefotaxime, clindamycin, erythromycin, tetracycline and multidrug resistance was significantly more prevalent among non-invasive pneumococci.

The results of this survey indicate that, due to the serotypes associated with non-invasive pneumococcal infections, childhood pneumococcal vaccination would be expected to have a smaller impact on non-invasive infections than invasive disease. However, due to most of the resistance among non-invasive pneumococci being associated with serotype 19F, one of the types in PCV-7 and the most common serotype among non-invasive pneumococci, vaccination should have a marked impact on resistance among non-invasive pneumococci.

RECOMMENDATIONS

1 To monitor the impact of childhood pneumococcal vaccination on non-invasive pneumococci, this survey, with a greater geographical collection base, should be repeated within 3 years of the 1 June 2008 addition of pneumococcal vaccination to the New Zealand immunisation schedule.

2 To monitor the impact of vaccination on the prevalence and spectrum of non-invasive pneumococcal infections, there should also be clinical-based surveillance of these infections.

1. INTRODUCTION

For many years, ESR has systematically monitored serotypes and antimicrobial susceptibility among *Streptococcus pneumoniae* causing invasive pneumococcal disease (IPD) in New Zealand. Diagnostic microbiology laboratories have been asked to refer all invasive *S. pneumoniae* isolates to ESR for this surveillance.

Based on this surveillance, information on the epidemiology of IPD, serotypes and antimicrobial susceptibility has been published periodically.[1,2,3,4,5,6] In addition, reports on the antimicrobial susceptibility of isolates from IPD cases have been published annually on ESR's surveillance website at http://www.surv.esr.cri.nz/antimicrobial/streptococcus_pneumoniae.php.

On 1 June 2008, the 7-valent pneumococcal conjugate vaccine (PCV-7), Prevenar®, was added to the New Zealand immunisation schedule. Overseas experience has shown that this vaccine not only has an impact on the incidence of IPD but also on other pneumococcal infections in both the vaccinated and unvaccinated population.[7,8]

There has been no routine monitoring of serotypes and antimicrobial susceptibility among non-invasive *S. pneumoniae* in New Zealand. In March 2008, with the pending introduction of PCV-7, the Ministry of Health's Pneumococcal Surveillance Advisory Group recommended that data on serotypes and susceptibility among non-invasive pneumococci, circulating in New Zealand prior to the introduction of PCV-7, should be collected. Such data will be a necessary prerequisite to fully assess the impact of vaccination on all pneumococcal infections.

Therefore from early April 2008, a sample of non-invasive pneumococci was collected from two large community laboratories and referred to ESR for serotyping and susceptibility testing.

2. METHODS

2.1. Bacterial isolates

Non-invasive *S. pneumoniae* isolates were referred to ESR from Diagnostic Medical Laboratory, Auckland (DML), and Medlab South, Christchurch (MLS). The aim was to collect approximately 200 non-repeat isolates from both laboratories. Ideally the isolates from each laboratory were to comprise 50 from ears, 50 from eyes, 50 from sputum and 50 from any other sites.

The following information was supplied with the isolates:

1 patient age and sex;

2 date specimen (from which *S. pneumoniae* was isolated) was taken; and

3 site of specimen.

2.2. Confirmation and serotyping

Referred isolates were confirmed as *S. pneumoniae* using optochin testing, demonstration of alpha-haemolysis on blood agar, and the bile solubility test.

Confirmed isolates were serotyped by the capsular antigen reaction (Neufeld test), using the Danish system of nomenclature and sera obtained from the Statens Serum Institut.[9]

2.3. Antimicrobial susceptibility testing

The penicillin, cefotaxime and moxifloxacin minimum inhibitory concentrations (MICs) were determined by Etest (AB Biodisk, Solna, Sweden), using Mueller-Hinton agar with 5% sheep blood and incubation for 16-20 hours in 5% CO_2. Chloramphenicol, co-trimoxazole, erythromycin, tetracycline and vancomycin susceptibilities were determined by the Clinical and Laboratory Standards Institute's (CLSI's) disc susceptibility testing method.[10] Inducible clindamycin resistance was detected by the D-zone test.[10]

All MICs and zone diameters were interpreted according to the 2008 CLSI standard.[11] In this standard, the interpretive criteria for pneumococcal penicillin MICs were redefined, with the introduction of different criteria for the parenteral treatment of meningitis, the parenteral treatment of non-meningitis infections, and the oral treatment of non-meningitis infections. Different cefotaxime interpretive standards for meningitis and non-meningitis infections were introduced in 2002.

In this report, when associations between penicillin or cefotaxime resistance and patient demographics, geographical distribution or serotypes have been made, the penicillin oral treatment and the cefotaxime non-meningitis interpretive standards were used.

Multidrug resistance was defined as resistance to three antibiotics in addition to penicillin. For the purposes of this definition, the penicillin oral treatment and the cefotaxime non-meningitis interpretive standards were used.

2.4. Data analysis

The serotype distribution and antimicrobial susceptibility among the non-invasive pneumococci included in this survey was compared with that among pneumococci causing invasive disease in 2008. The data on the serotype distribution and antimicrobial susceptibility among isolates from invasive disease was sourced from ESR's laboratory-based surveillance of IPD.[6]

Statistical analyses were performed with SAS software v.9.1 (SAS Institute Inc, Cary, NC, USA). The chi-square test or Fisher's exact test, as appropriate, were used to determine the significance of any observed differences. An associated P value ≤ 0.05 was used to indicate that a difference was significant.

3. RESULTS

3.1. Isolates

The number and source of the non-invasive pneumococcal isolates included in the survey are shown in Table 1.

The isolates from DML were from specimens taken between 4 May and 23 July 2008, and those from MLS were from specimens taken between 10 April and 24 September 2008.

Table 1. Source of non-invasive *S. pneumoniae* isolates

	Number (%) of isolates				
	Site				
Laboratory	**ear**	**eye**	**sputum**	**other**	**all sites**
Diagnostic Medical Laboratory, Auckland	50	52	66[1]	32	200
Medlab South, Christchurch	52	43	43	16	154
Total	102 (28.8)	95 (26.8)	109 (30.8)	48[2] (13.6)	354

1 Includes 40 isolates from the throat.
2 36 (75%) of these 48 isolates were from nasal sites.

3.2. Patient demographics

The age and sex distribution of the patients from whom the non-invasive pneumococci were isolated is shown Table 2.

There were some differences in the age distribution of the patients in Auckland and those in Christchurch. A greater proportion of the Christchurch patients were <1 year of age (21.7% vs 12.5% of Auckland patients) and ≥65 years of age (17.1% vs 6.0%). A greater proportion of the Auckland patients were in the 2-14 year age group (40.5% vs 21.1%).

Table 2. Age and sex of patients from whom non-invasive *S. pneumoniae* isolated[1]

Age group (years)	Female	Male	All patients	
	Number	Number	Number	Percent
<1	28	30	58	16.5
1	23	26	50	14.2
2-4	30	28	58	16.5
5-14	30	25	55	15.6
15-24	10	6	16	4.6
25-34	9	7	16	4.6
35-44	16	7	23	6.5
45-54	10	7	17	4.8
55-64	13	8	21	6.0
65-74	9	11	20	5.7
75-84	6	8	14	4.0
≥85	3	1	4	1.1
All ages	187	164	354	100

1 Age and sex not known for 2 patients and sex not known for 1 further patient.

3.3. Serotypes

Table 3. Serotypes among non-invasive *S. pneumoniae* by patient age group

Serotype[2]	Proportion (%) of non-invasive *S. pneumoniae* within the age group (years)[1] due to the serotype:				
	≤1 (n=108)	**2-4** (n=58)	**5-64** (n=148)	**≥65** (n=38)	**All ages** (n=354)
Serotypes in PCV-7:					
4	0.0	0.0	0.7	0.0	0.3
6B	8.3	12.1	2.7	5.3	6.2
9V	3.7	3.5	5.4	5.3	4.5
14	11.1	10.3	2.7	7.9	7.1
18C	3.7	0.0	2.7	2.6	2.5
19F	23.2	24.1	14.9	34.2	21.2
23F	11.1	5.2	3.4	5.3	6.2
Total for PCV-7 serotypes	**61.1**	**55.2**	**32.4**	**60.5**	**48.0**
Additional serotypes in PCV-10:					
1	1.9	3.5	2.7	2.6	2.5
5	0.0	0.0	0.0	0.0	0.0
7F	0.0	0.0	0.0	0.0	0.0
Total for PCV-10 serotypes	**63.0**	**58.6**	**35.1**	**63.2**	**50.6**
Additional serotypes in PCV-13:					
3	2.8	5.2	12.8	5.3	7.9
6A	4.6	5.2	2.7	7.9	4.2
19A	9.3	5.2	3.4	0.0	5.1
Total for PCV-13 serotypes	**79.6**	**74.1**	**54.1**	**76.3**	**67.8**
Non-PCV serotypes:					
10A	1.9	0.0	1.4	2.6	1.4
11A	2.8	1.7	3.4	0.0	2.5
15B	0.9	0.0	2.0	2.6	1.4
22F	0.9	3.5	2.0	0.0	1.7
23A	0.0	0.0	2.0	2.6	1.1
29	0.9	1.7	2.0	0.0	1.4
non-typable	11.1	19.0	25.0	13.2	18.4
other types	1.9	0.0	8.1	2.6	4.2

1 Age not known for 2 patients.

2 All the serotypes included in the 7-valent pneumococcal conjugate vaccine (PCV-7), the 10-valent conjugate vaccine (PCV-10) and the 13-valent conjugate vaccine (PCV-13) are included in the table and listed according to their inclusion in each of the vaccines. The specific non-PCV serotypes included in the table are those that accounted for ≥1% of isolates. The non-PCV serotypes that accounted for <1% of isolates are grouped under 'other types'.

Table 3 shows the serotypes of the non-invasive pneumococci included in the survey according to the age of the patients from whom the pneumococci were isolated. The table also indicates the proportions of these non-invasive pneumococci that were one of serotypes included in PCV-7, the 10-valent pneumococcal conjugate vaccine (PCV-10), and the 13-valent pneumococcal conjugate vaccine (PCV-13).

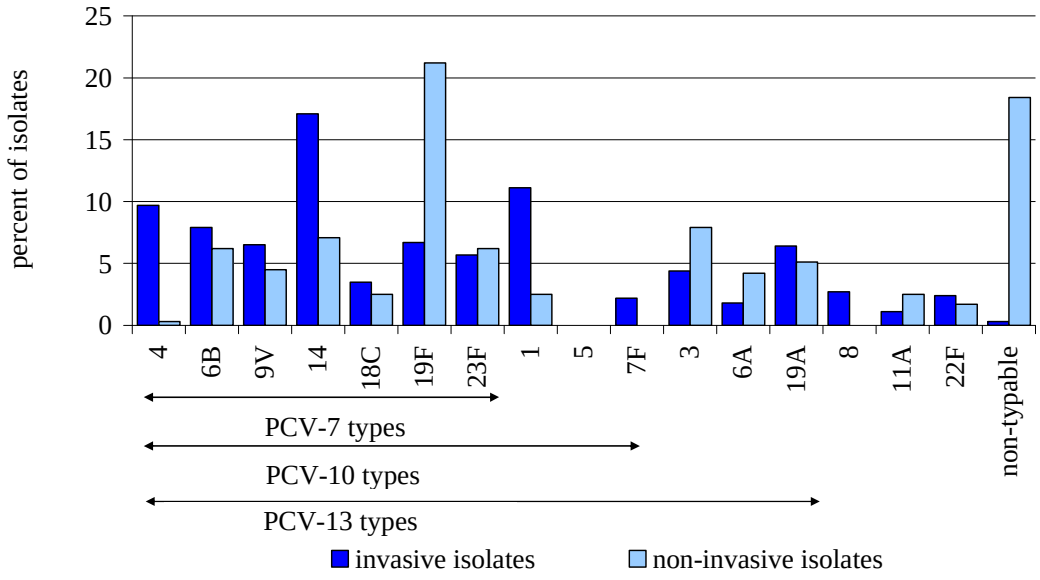
The serotype distribution was notable for the predominance of serotype 19F and non-typable isolates (Table 3). The proportion that were serotype 19F or non-typable was similar among isolates from Auckland and Christchurch: serotype 19F accounted for 21.5% and 20.8% of isolates from Auckland and Christchurch, respectively, and non-typable strains accounted for 18.5% and 18.2% of isolates from Auckland and Christchurch, respectively.

Non-typable isolates were associated with pneumococci isolated from the eye, with 75.4% of non-typable isolates being from this site and non-typable isolates accounting for 51.6% of isolates from the eye. No other associations between serotypes and site were obvious.

3.3.1. Comparison with serotypes causing invasive disease

Figure 1 presents a comparison of the serotypes among the non-invasive pneumococci included in this survey with the types among invasive pneumococci in 2008. Non-typable and serotype 19F isolates were significantly more prevalent among non-invasive pneumococci, and serotypes 1, 4 and 14 were more prevalent among invasive isolates.

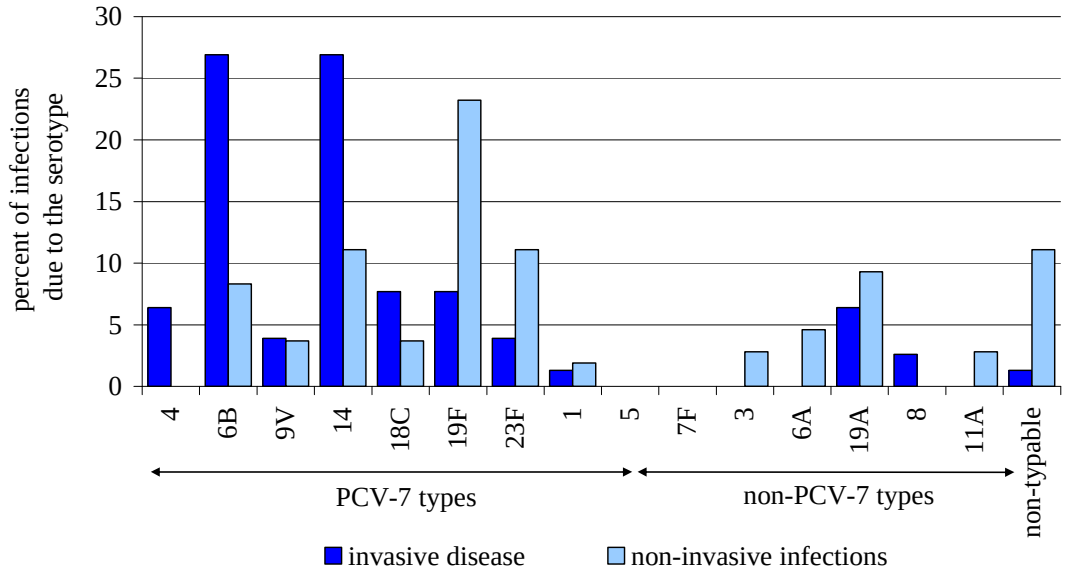
Figure 1. Serotypes among invasive and non-invasive pneumococci from all patients, 2008



The serotypes shown are those in the PCV-13 vaccine and any others that accounted for ≥2% of either invasive isolates or non-invasive isolates.

Figure 2 shows a similar comparison to Figure 1, but is confined to isolates from patients ≤1 year of age. Again non-typable and serotype 19F isolates were significantly more prevalent among non-invasive pneumococci. Serotypes 4, 6B and 14 were more prevalent among invasive isolates.

Figure 2. Serotypes causing invasive and non-invasive pneumococcal infections, among patients ≤1 year old, 2008



Serotypes shown are those in the PCV-13 vaccine and any others that accounted for ≥2% of either invasive isolates or non-invasive isolates in patients ≤1 year of age.

Table 4 compares the proportion of non-invasive and invasive pneumococci isolated from patients in the different age groups that were due to one of the serotypes included in PCV-7, PCV-10, PCV-13 and the 23-valent polysaccharide vaccine (PPV-23). In all age groups, there was better coverage of invasive isolates than non-invasive isolates, although, except for PPV-23, the differences were small in the ≥65 year age group.

Table 4. Comparison of the potential coverage of non-invasive and invasive *S. pneumoniae* by pneumococcal conjugate vaccines and the polysaccharide vaccine by patient age group, 2008

Vaccine[1]	Proportion (%) of *S. pneumoniae* within the age group (years)[2] due to serotypes in the vaccine:				
	≤1	2-4	5-64	≥65	All ages
PCV-7					
non-invasive isolates	61.1	55.2	32.4	60.5	48.0
invasive isolates	83.3	70.6	44.0	63.0	57.1
PCV-10					
non-invasive isolates	63.0	58.6	35.1	63.2	50.6
invasive isolates	84.6	82.4	67.4	67.8	70.5
PCV-13					
non-invasive isolates	79.6	74.1	54.1	76.3	67.8
invasive isolates	91.0	94.1	82.1	79.7	83.0
PPV-23					
non-invasive isolates	81.5	74.1	61.5	76.3	71.5
invasive isolates	96.2	94.1	94.2	91.6	93.5

1 PCV-7 includes serotypes 4, 6B, 9V, 14, 18C, 19F and 23F; PCV-10 includes serotypes 1, 4, 5, 6B, 7F, 9V, 14, 18C, 19F and 23F; PCV-13 includes serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F and 23F; PPV-23 includes serotypes 1, 2, 3, 4, 5, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 17F, 18C, 19A, 19F, 20, 22F, 23F and 33F.

2 Age not known for 2 patients.

3.4. Antimicrobial susceptibility

Table 5 shows the antimicrobial susceptibility of the non-invasive pneumococcal isolates included in this survey. In this table, the penicillin and cefotaxime MICs have been interpreted according to all of the CLSI interpretive criteria for these two antibiotics.

Table 5. Antimicrobial susceptibility among non-invasive *S. pneumoniae*

	Interpretive standards			Percent		
	S[1]	I[1]	R[1]			
	MIC (mg/L)			S	I	R
penicillin						
meningitis	≤0.06	-	≥0.12	67.5	-	32.5
non-meningitis	≤2	4	≥8	93.5	6.2	0.3
oral treatment	≤0.06	0.12-1	≥2	67.5	15.3	17.2
cefotaxime						
meningitis	≤0.5	1	≥2	75.1	11.9	13.0
non-meningitis	≤1	2	≥4	87.0	7.1	5.9
moxifloxacin	≤1	2	≥4	100	0.0	0.0
	Zone diameter (mm)					
chloramphenicol	≥21	-	≤20	97.5	-	2.5
clindamycin[2]	≥19	16-18	≤15	86.4	0.3	13.3
co-trimoxazole	≥19	16-18	≤15	63.3	3.1	33.6
erythromycin	≥21	16-20	≤15	76.6	0.6	22.9
tetracycline	≥23	19-22	≤18	78.5	1.7	19.8
vancomycin	≥17	-	-	100	-	-

1 S, susceptible; I, intermediate; R, resistant.
2 The percentages intermediate and resistant are for constitutive clindamycin resistance. A further 4 isolates (1.1%) had inducible clindamycin resistance.

13.8% of isolates had combined penicillin (oral treatment interpretation) and erythromycin resistance. 12.7% were multidrug resistant, that is, resistant to penicillin (oral treatment interpretation) and at least three additional antibiotics. The most common resistance patterns among these multidrug-resistant isolates were penicillin, co-trimoxazole, erythromycin and tetracycline resistance (51.1% of the multidrug-resistant isolates) and penicillin, cefotaxime, co-trimoxazole, erythromycin and tetracycline resistance (37.8% of the multidrug-resistant isolates).

The only significant difference in the prevalence of susceptibility among isolates from Auckland compared with those from Christchurch was higher chloramphenicol resistance in isolates from Auckland (4.0% vs 0.7%). However, while not significant at the 95% probability level, penicillin resistance was higher among isolates from Christchurch (oral treatment interpretation: 20.8% vs 14.5%, P=0.1209), as was cefotaxime resistance (non-meningitis interpretation: 8.4% vs 4.0%, P=0.0795), erythromycin resistance (27.3% vs 19.5%, P=0.0844), and multidrug resistance (15.6% vs 10.5%, P=0.1545).

Table 6 shows the prevalence of resistance in the different patient age groups. Cefotaxime, clindamycin, co-trimoxazole, erythromycin, penicillin and multidrug resistance was significantly more prevalent among pneumococci from patients ≥65 years of age. Co-trimoxazole, penicillin and multidrug resistance was significantly higher among isolates from infants ≤1 year of age than among patients 2-64 years old (ie, those in the 2-4 and 5-64 year age groups).

Table 6. Antimicrobial resistance among non-invasive *S. pneumoniae* by patient age group[1, 2]

	Percent resistance				
	≤1 year (n=108)	**2-4 years** (n=58)	**5-64 years** (n=148)	**≥65 years** (n=38)	**All ages** (n=354)
cefotaxime[3]	3.7	3.5	4.7	21.1	5.9
chloramphenicol	0.9	5.2	2.7	2.6	2.5
clindamycin[4]	13.0	12.1	10.1	26.3	13.3
co-trimoxazole	39.8	36.2	24.3	47.4	33.6
erythromycin	26.9	17.2	18.2	36.8	22.9
penicillin[5]	23.2	12.1	9.5	36.8	17.2
tetracycline	24.1	13.8	16.2	29.0	19.8
multidrug resistance	15.7	10.3	7.4	26.3	12.7

1 All isolates were susceptible to moxifloxacin and vancomycin.
2 Age not known for 2 cases.
3 Based on cefotaxime non-meningitis interpretation.
4 Constitutive clindamycin resistance.
5 Based on penicillin oral treatment interpretation.

Table 7 shows the prevalence of resistance among isolates according to the site of isolation. Generally resistance was highest among pneumococci isolated from ears and sputum.

Table 7. Antimicrobial resistance among non-invasive *S. pneumoniae* by site

	Percent resistance				
	ear (n=102)	**eye** (n=95)	**sputum** (n=109)	**other** (n=48)	**All sites** (n=354)
cefotaxime[1]	3.9	3.2	11.9	2.1	5.9
chloramphenicol	2.0	2.1	3.7	2.1	2.5
clindamycin[2]	8.8	13.7	17.4	12.5	13.3
co-trimoxazole	37.3	27.4	37.6	29.2	33.6
erythromycin	25.5	20.0	25.7	16.7	22.9
penicillin[3]	21.6	11.6	20.2	12.5	17.2
tetracycline	22.6	16.8	22.9	12.5	19.8
multidrug resistance	15.7	7.4	16.5	8.3	12.7

1 Based on cefotaxime non-meningitis interpretation.
2 Constitutive clindamycin resistance.
3 Based on penicillin oral treatment interpretation.

Serotype 19F accounted for the majority of the penicillin-resistant isolates, and almost all cefotaxime-resistant and multidrug-resistant isolates (Table 8). As a consequence, the majority of the penicillin-, cefotaxime- and multidrug-resistant isolates were one of the serotypes included in PCV-7. There was more serotype variation among the isolates with intermediate resistance to penicillin and cefotaxime.

Among the total 75 serotype 19F isolates, 39 (52.0%) were multidrug resistant, with the most common resistance patterns being penicillin, co-trimoxazole, erythromycin and tetracycline resistance (26.7% of the serotype 19F isolates) and penicillin, cefotaxime, co-trimoxazole, erythromycin and tetracycline resistance (22.7% of the serotype 19F isolates).

Table 8. Serotypes among penicillin and cefotaxime resistant and intermediate, and multidrug resistant, non-invasive *S. pneumoniae*

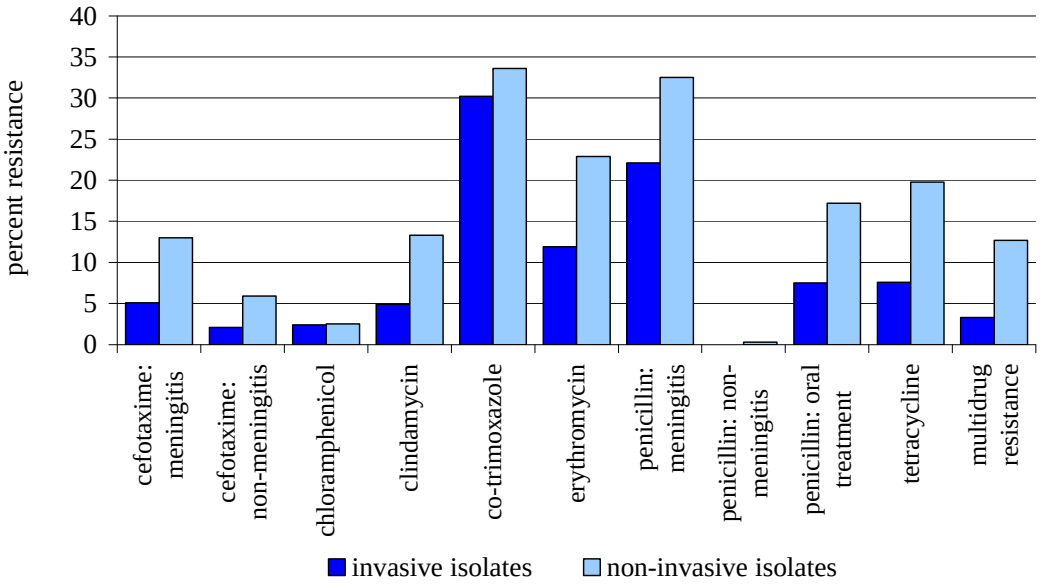
Serotype	Number (%[1]) isolates				
	penicillin		cefotaxime		multidrug resistant[4] (n=45)
	intermediate[2] (n=54)	resistant[2] (n=61)	intermediate[3] (n=25)	resistant[3] (n=21)	
Serotypes in PCV-7:[5]					
6B	5 (9.3)	2 (3.3)	1 (4.0)	0	1 (2.2)
9V	12 (22.2)	3 (4.9)	0	0	0
14	1 (1.9)	6 (9.8)	4 (16.0)	0	1 (2.2)
19F	11 (20.4)	44 (72.1)	16 (64.0)	20 (95.2)	39 (86.7)
23F	3 (5.6)	3 (4.9)	3 (12.0)	0	3 (6.7)
Total for PCV-7 serotypes	**32 (59.3)**	**58 (95.1)**	**24 (96.0)**	**20 (95.2)**	**44 (97.8)**
Additional serotypes in PCV-10:[6]	0	0	0	0	0
Total for PCV-10 serotypes	**32 (59.3)**	**58 (95.1)**	**24 (96.0)**	**20 (95.2)**	**44 (97.8)**
Additional serotypes in PCV-13:[7]					
3	0	1 (1.6)	0	1 (4.8)	1 (2.2)
19A	3 (5.6)	1 (1.6)	1 (4.0)	0	0
Total for PCV-13 serotypes	**35 (64.8)**	**60 (98.4)**	**25 (100)**	**21 (100)**	**45 (100)**
Non-PCV serotypes:					
19 non-typable	1 (1.9)	0	0	0	0
23A	1 (1.9)	0	0	0	0
29	3 (5.6)	1 (1.6)	0	0	0
non-typable	14 (25.9)	0	0	0	0

1 Percentage of the intermediate, resistant or multidrug-resistant isolates.
2 Based on penicillin oral treatment interpretations.
3 Based on cefotaxime non-meningitis interpretations.
4 Resistant to penicillin (oral treatment interpretation) and three additional antibiotics.
5 There were no penicillin- or cefotaxime-resistant or intermediate isolates of serotypes 4 or 18C which are also included in PCV-7.
6 There were no penicillin- or cefotaxime-resistant or intermediate isolates of serotypes 1, 5 and 7F which are the additional serotypes included in PCV-10.
7 There were no penicillin- or cefotaxime-resistant or intermediate isolates of serotype 6A which is also included in PCV-13.

3.4.1. Comparison with antimicrobial susceptibility among isolates causing invasive disease

Resistance to all antibiotics and multidrug resistance was more common among non-invasive pneumococci than pneumococci from IPD cases in 2008 (Figure 3). The differences were significant for cefotaxime (both with the meningitis and non-meningitis interpretations), clindamycin, erythromycin, penicillin (with the meningitis and oral treatment interpretations), tetracycline and multidrug resistance.

Figure 3. Resistance among invasive and non-invasive pneumococci, 2008



As observed for the non-invasive isolates included in this survey, chloramphenicol resistance was also more prevalent among invasive pneumococci isolated in Auckland in 2008 than among isolates from Christchurch (5.1% vs 0.0%), but the difference was not significant at the 95% probability level (P=01276). There were no significant differences in resistance to any of the other antibiotics among the invasive isolates from Auckland compared with those from Christchurch.

4. DISCUSSION

As expected, many countries have reported dramatic reductions in IPD rates following the introduction of childhood immunisation with pneumococcal conjugate vaccine. In addition to a reduction in IPD among children belonging to age groups eligible for vaccine, there have been two additional benefits from this immunisation. First, there have been reductions in IPD in other age groups through herd immunity.[12,13] These decreases in disease among unvaccinated individuals are assumed to be due to reduced exposure to pneumococci because conjugate vaccines prevent nasopharyngeal colonisation with vaccine types. Second, there have been reductions in rates of non-invasive pneumococcal infections, in particular, community-acquired pneumonia and acute otitis media.[7,8]

The aim of this survey was to provide baseline information on the serotypes and antimicrobial susceptibility of non-invasive pneumococci in New Zealand prior to the addition of pneumococcal vaccination to the New Zealand immunisation schedule. This baseline information should facilitate future assessments of the impact of childhood pneumococcal vaccination on all pneumococcal infections. To our knowledge, this is the first study of serotypes among non-invasive pneumococci in New Zealand.

Non-typable and serotype 19F isolates were most prevalent among the non-invasive pneumococci included in this survey, although three-quarters of the non-typable isolates were from eye sites. Similar results have been reported from other countries not using pneumococcal conjugate vaccines. A Chinese study reported a predominance of non-typable and serogroup 19 (serotyping not performed) pneumococci in nasopharyngeal specimens from children with upper respiratory infections between the years 2000 and 2005.[14] Serotype 19F was also the most prevalent serotype among pneumococci isolated in 2006 and 2007 from children with acute otitis media in Japan.[15]

Our results indicate that PCV-7 would provide less coverage against the serotypes causing non-invasive infections than those responsible for IPD. Forty-eight percent of the non-invasive isolates from patients of all ages were a PCV-7 type versus 57.1% of isolates from invasive disease in 2008 (Table 4). This margin was even greater among the younger age groups: 61.1% vs 83.3% for patients ≤1 year of age. The greater coverage of invasive disease is not surprising as PCV-7 is formulated to maximise coverage of IPD. The coverage rates for non-invasive pneumococci are similar to those found in several other studies summarised in a review by Bogaert et al.[16]

After non-typable isolates, serotype 3 was the most common non-PCV-7 type, when patients of all ages were considered, and serotype 19A was the most common non-PCV-7 type in the youngest age group (≤1 year olds). Both these serotypes have been associated with serious pneumococcal infections and their incidence has been reported to have increased following the introduction of PCV-7.[17] Serotype 19A is already a relatively frequent cause of IPD in New Zealand.[6] Serotype 3 has been associated with an increased case-fatality rate,[18] and pneumococcal necrotising pneumonia.[19]

Resistance to almost all antibiotics tested and multidrug resistance was significantly higher among the non-invasive pneumococci than pneumococci from IPD in 2008. Similar differences have often been reported.[20,21,22] Among the possible reasons for this are differences in the serotypes causing invasive and non-invasive pneumococcal infections and the selective pressure of frequent antibiotic use to treat upper respiratory tract infections. In this study, serotype 19F was the prevalent serotype, accounting for 21.2% of all non-invasive isolates and 52.0% of these isolates were multidrug resistant. In contrast, serotype 14 was the most prevalent serotype among isolates from IPD cases in 2008, but was only infrequently (1.9%) multidrug resistant. Serotype 19F also had the highest rate (28.6%) of multidrug resistance among invasive isolates in 2008, but this serotype only accounted for 6.7% of IPD cases. It is notable that a significantly greater proportion of the non-invasive isolates of serotype 19F than the invasive isolates were multidrug resistant (52.0% vs 28.6%).

While not statistically significant, resistance to several antibiotics, including penicillin and cefotaxime, was higher among isolates from patients in Christchurch. However, a greater proportion of the Christchurch patients were in the youngest (<1 year old) and older age groups (≥65 years), and resistance was generally highest at these age-group extremes. An earlier study

of non-invasive pneumococci, collected in several centres throughout New Zealand over a 6-month period in 1997, also recorded a higher rate of penicillin and cefotaxime resistance in Christchurch compared with all other centres.[23]

Rates of penicillin resistance and non-susceptibility (resistance + intermediate resistance) in New Zealand appear to be similar to those in at least some other countries. The latest available data from Australia recorded 31% penicillin non-susceptibility (MIC ≥0.12 mg/L) among non-invasive pneumococci in 2005 - similar to our rate of 32.5%.[22] A Beijing study of nasopharyngeal isolates from children reported 31.5% penicillin non-susceptibility for the 2004-05 period.[14] However, a Japanese study reported a somewhat higher rate of 37.7% penicillin non-susceptibility.[15] Both the Chinese and Japanese studies also reported that serotype 19F (or serogroup 19 in the case of the Chinese study) was the most prevalent type among penicillin-resistant pneumococci.

This study had some limitations. First, the timing of the survey was less than optimal, as the period required to collect the target number of isolates extended to the end of July in Auckland and the end of September in Christchurch - several months beyond the 1 June 2008 introduction of the vaccine. PCV-7 has a reported effectiveness of 73% after 1 dose in infants ≤7 months of age, which rises to 96% after two doses.[24] Therefore, during the later stages of the isolate collection period, the vaccine could already have been having some, albeit small, impact on pneumococcal infections in infants eligible for vaccine and possibly other age groups through herd immunity.

Second, the collection of isolates occurred during the late autumn and winter months. This may have biased the results towards higher than average levels of resistance, as resistance in *S. pneumoniae* can be cyclical with increasing resistance in the winter months.[25]

Third, the survey is limited to some extent by the fact that it only included isolates from two laboratories, however, these laboratories serve large population bases in geographically distinct areas. Given the late decision to initiate the survey, this approach was considered the most practical to ensure the earliest possible start to isolate collection.

The results of this survey indicate that, due to the serotypes associated with non-invasive pneumococcal infections, childhood pneumococcal vaccination would be expected to have a smaller impact on non-invasive infections than invasive disease. However, due to most of the resistance among non-invasive pneumococci being associated with serotype 19F, one of the types in PCV-7 and the most common serotype among non-invasive pneumococci, vaccination should have a marked impact on resistance among non-invasive pneumococci.

To monitor the impact of childhood pneumococcal vaccination on pneumococci causing non-invasive infections, this survey, with a greater geographical collection base, should be repeated within 3 years of the 1 June 2008 addition of pneumococcal vaccination to the New Zealand immunisation schedule. Ideally such laboratory-based monitoring of the impact of vaccination on the serotypes and antimicrobial susceptibility among non-invasive pneumococci should be complemented with clinical-based surveillance of trends in the prevalence and spectrum of non-invasive pneumococcal infections.

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