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Original Article

Invasive pneumococcal disease in Indian adults: 11 years' experience

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Abstract *Purpose:* To investigate the epidemiology of invasive pneumococcal disease (IPD), prevalent serotypes, and pattern of antimicrobial resistance (AMR) in Indian adults.
Methods: Prospective laboratory based surveillance of IPD was carried out in >18 years age group between January 2007 and July 2017, from a tertiary care hospital in South India. All *Streptococcus pneumoniae* culture positives from blood, CSF and sterile body fluids were characterized to identify the serotypes and AMR.
Results: A total of 408 IPD cases were characterized in this study. The overall case fatality rate in this study was 17.8% (95% confidence interval (CI): 14.1, 22.4). Pneumonia (39%), meningitis (24.3%), and septicaemia (18.4%) were the most common clinical conditions associated with IPD. Serotypes 1, 3, 5, 19F, 8, 14, 23F, 4, 19A and 6B were the predominant serotypes in this study. Penicillin non-susceptibility was low with 6.4%
Conclusion: Serotype data from this study helped in accurate estimation of pneumococcal conjugate vaccine-13 and pneumococcal polysaccharide vaccine-23 protective coverage against serotypes causing IPD in India as 58.7% (95% CI: 53.8, 63.4) and 67.4% (95% CI: 62.7, 71.8) respectively. Penicillin non-susceptibility in meningeal IPD cases is 27.4%. Empirical therapy for meningeal IPD must be cephalosporin in combination with vancomycin since cefotaxime non-susceptibility in meningeal IPD is 9.9%
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Introduction

Invasive diseases due to *Streptococcus pneumoniae*, continue to be a major global cause of morbidity and mortality, particularly in children <2 years, adults ≥65 years of age[1,2] and in population with certain co-morbid illnesses such as HIV infection and haematological malignancy of all age group.[3] While the widespread use of pneumococcal conjugate vaccines (PCVs) has resulted in a significant decline in the incidence of invasive pneumococcal disease (IPD) in children,[4] data on the usefulness of the pneumococcal polysaccharide vaccine (PPV-23) in adults remain limited. The administration of PCV-13/PPV-23 in individuals older than 65 years is recommended by the Geriatric Society of India,[5] and by the Expert Group of the Association of Physicians of India (API). The API also recommends PCV-13/PPV-23 in more than 18 years individuals who are at risk of acquiring pneumococcal diseases.[6] India has the lowest incidence of penicillin resistant *S. pneumoniae* amongst Asian nations, however increase in the emergence of resistant isolates in India in recent times is of major concern,[7] since it significantly contributes to treatment failure, prolonged hospital stay and poorer outcomes.

India lacks data on the trends of antimicrobial resistance (AMR) of IPD isolates and the national distribution of invasive pneumococcal serotypes that could aid in the revision of treatment and preventive policy guidelines. With the exception of a large decade long (1993–2008) multicentre Indian study reporting adult IPD,[8] there are relatively few studies focussed on the AMR and serotype profiles of IPD isolates on Indian adults and on possible changes in these patterns since 2008.[9–11]

The objectives of this present work was to study the prevalence of serotypes and AMR, describe the changing trend of AMR profile in adult IPD isolates over 11 years (January 2007–July 2017) and predict the range of PCV-13 and/or PPV-23 coverage in Indian adults.

Material and methods

Study design and sample population

This was a prospective, laboratory based study comprising of adult individuals aged more than 18 years, admitted to a 2500 bed tertiary care referral centre in South India between January 2007 and July 2017. Patients with clinical suspicion of IPD attending inpatient and outpatient services were enrolled and their sterile specimens (blood, CSF and sterile body fluids) were submitted for microbial identification of *S. pneumoniae*.

Case definition and clinical data collection

IPD is defined as *S. pneumoniae* identification from blood, CSF and any other sterile body fluid specimens. Clinical data from eligible subjects were prospectively collected using a systematic data abstraction form. Patient data included age, gender, clinical syndrome, mortality outcome and co-morbid illnesses. The study population was categorised into three groups as, very high risk, high risk and moderate risk, based on the risk for acquiring IPD due to the accompanying co-morbid illnesses, as described in a previous report.[12] Briefly individuals with anatomical or functional asplenia, HIV and individuals with medical conditions like leukaemia, lymphoma and bone marrow cancer are considered to be very high risk for pneumococcal diseases. Individuals with complement-defects, T-cell deficiency and neutropenia, chronic liver and kidney disease, individuals with >65 years, homeless individuals and drug addicts are meant to have high risk for acquiring IPD. Moderate risk group for acquiring IPD are individuals with diabetic mellitus, cardiovascular problems, chronic respiratory disease [e.g. chronic obstructive pulmonary disease (COPD)], neurological conditions like stroke, tetraplegia, multiple sclerosis and myasthenia gravis, or severe osteoporosis with kyphosis and Bechterew's disease and individuals who are under disease modifying anti-rheumatic drugs, and smokers.

Case definitions for meningeal and non-meningeal *S. pneumoniae* isolates

Pneumococcal meningeal diseases was defined as isolation of *S. pneumoniae* from a patients CSF or blood with clinical syndrome consistent with meningitis: CSF leucocytosis >100 cells/mm^3 or 10–100 cells/mm^3 either with decreased CSF glucose (<40 mg/dL) or elevated CSF protein (>100 mg/dL).[13] Pneumococcal non-meningeal diseases were defined as isolation of *S. pneumoniae* from blood or sterile body fluids from a patient without a meningitis clinical syndrome.[14]

Laboratory procedures

Clinical specimens (blood, CSF and other sterile body fluids) were collected from each enrolled patients with possible IPD depending clinical presentation. Blood specimens were processed on BacT/ALERT 3D system (semi-automated blood culture system from biomerieux, France) as per standard protocols.[15] CSF and other fluid specimens were processed within 1 h of receipt of specimens as described elsewhere.[15] *S. pneumoniae* was identified by routine phenotypic methods like bile solubility and optochin susceptibility tests[15] and further serotyped using co-agglutination technique[16] using antisera (Statens Serum Institute, Denmark). The serotypes were reconfirmed by sequential multiplex PCR. In brief, PCR was set up in a 25 μL reaction mix containing, 2× PCR Buffer (QIAGEN Multiplex PCR Kit (cat#206143)), 5 μL of template DNA, primers with specific concentrations and PCR cycling conditions as described previously.[17]

Antimicrobial susceptibility testing (AST)

AST was performed on 131 isolates (2007–2010) using the agar dilution method[18] and rest of 277 isolates (2011–July 2017) was tested using the VITEK system 2 (biomerieux, France). Minimum inhibitory concentration (MIC) breakpoint interpretations for antibiotics were done as per the Clinical and Laboratory Standards Institute recommendations (CLSI, 2017).[19] For meningeal isolates MIC breakpoints of susceptibility and resistance for penicillin are ≤0.06 and

≥0.12 μg/mL and for cefotaxime, susceptibility, intermediate susceptibility and resistance are ≤0.5, 1.0 and ≥2 μg/mL, respectively.[19] For non-meningeal isolates susceptibility, intermediate susceptibility and resistance for penicillin and cefotaxime were defined as ≤2, 4.0, ≥8 μg/mL and ≤1, 2.0, ≥4 μg/mL respectively as per CLSI recommendations.[19]

Statistical methods

Statistical analysis was performed using STATA 13.0 for Windows (StataCorp, College Station, TX, USA) software. All variables were summarized using frequency distribution with percentages. Association between demographic and clinical characteristics with case fatality was compared using χ^2 test or Fishers' exact test depending on the distribution of the data. The 95% CI was presented for each category of the demographic and clinical characteristics and was calculated using Wilson Score Method. Bar charts were used to visualize the categorical variables. All statistical tests were two sided and $P < 0.05$ defines significance.

Results

A total of 408 invasive isolates were recovered during the study period. Clinical data on IPD associated syndromes, associated co-morbid illnesses and outcomes were available for 91.7% (374/408), 81.6% (333/408) and 79.6% (325/408) of patients respectively (Table 1).

Clinical syndrome

Pneumonia and meningitis were the most common clinical syndromes associated with IPD, accounting for 39% (146/374) and 24.3% (91/374) of IPD cases. 18.4% (69/374) of IPD patients had clinical features suggestive of pneumococcal septicaemia with an unknown infective focus (Table 1).

Clinical outcomes

The overall case fatality rate was 17.8% (58/325; 95% confidence interval (CI): 14.1, 22.4). Based on clinical syndrome, case fatality rates were noted to be highest in pneumococcal septicaemia without a known focus of infection with 36.9% (24/65; 95% CI: 26.2, 49.1) followed by patients with pneumonia 19.3% (23/119; 95% CI: 13.2, 27.3) and IPD associated meningitis 9.6% (7/73; 95% CI: 4.7, 18.5). Outcomes in 7.1% (23/325; 95% CI: 4.8, 10.4) IPD patients could not be determined since they were discharged against medical advice. The case fatality rate was noted to be the highest in individuals aged ≥66 years with 34.9% (15/43; 95% CI: 22.4, 49.8) (Table 1).

Comorbidities

17.4% (58/333) cases were healthy before acquiring IPD. The percentage of individuals in very high risk, high risk

Table 1 Demographic, clinical presentation and fatal outcome of study subjects.

Demographic variables	N[a] = 408 (%)	Case fatality			P value[b]	Most common serotypes
		Died	No of cases with fatality information	% (95% CI)		
Age group						
19–65	352 (86.3)	43	282	15.2 (11.5, 19.9)		1, 3, 5, 19F, 8
≥66	56 (13.7)	15	43	34.9 (22.4, 49.8)	0.002	3, 14, 19F, 7F, 8
Sex						
Male	284 (69.6)	39	222	17.6 (13.1, 23.1)		1, 3, 8, 19F, 5
Female	124 (30.4)	19	103	18.4 (12.1, 27.0)	0.847	1, 5, 19F, 3, 6A
Clinical syndromes						
Pneumonia	146 (39.0)	23	119	19.3 (13.2, 27.3)		1, 3, 5, 19A, 19F
Meningitis	91 (24.3)	7	73	9.6 (4.7, 18.5)		6B, 3, 1, 6A, 19F
Septicaemia	69 (18.4)	24	65	36.9 (26.2, 49.1)		3, 1, 8, 5, 14
Empyema	43 (11.5)	3	34	8.8 (3.0, 23.0)		1, 5, 4, 19F, 3
Peritonitis	25 (6.7)	1	25	4.0 (0.7, 19.5)	<0.001[c]	23F, 3, 17F, 19F, 1
Co-morbidities						
No risk	58 (17.4)	4	56	7.1 (2.8, 17.0)		1, 5, 14, 3, 4
Very high risk	79 (23.7)	10	79	12.7 (7.0, 21.8)		6A, 9V, 4, 8, 3
High risk	135 (40.5)	27	117	23.1 (16.4, 31.5)		3, 19F, 8, 14, 23F
Moderate risk	61 (18.3)	13	60	21.7 (13.1, 33.6)	0.033	1, 3, 19F, 23F, 4
Serotype groups						
PCV-10	179 (44.5)	24	143	16.8 (11.5, 23.8)		1, 5 19F, 14, 23F
Extra 3 PCV 13	57 (14.2)	12	46	26.1 (15.6, 40.3)		3, 19A, 6A
Extra 11 PPV 23	45 (11.2)	5	39	12.8 (5.6, 26.7)	0.235	8, 12F, 9N, 11A,17F

[a] Total number of individuals with IPD (%).
[b] P value based on χ^2 test.
[c] P value based on Fisher's exact test.

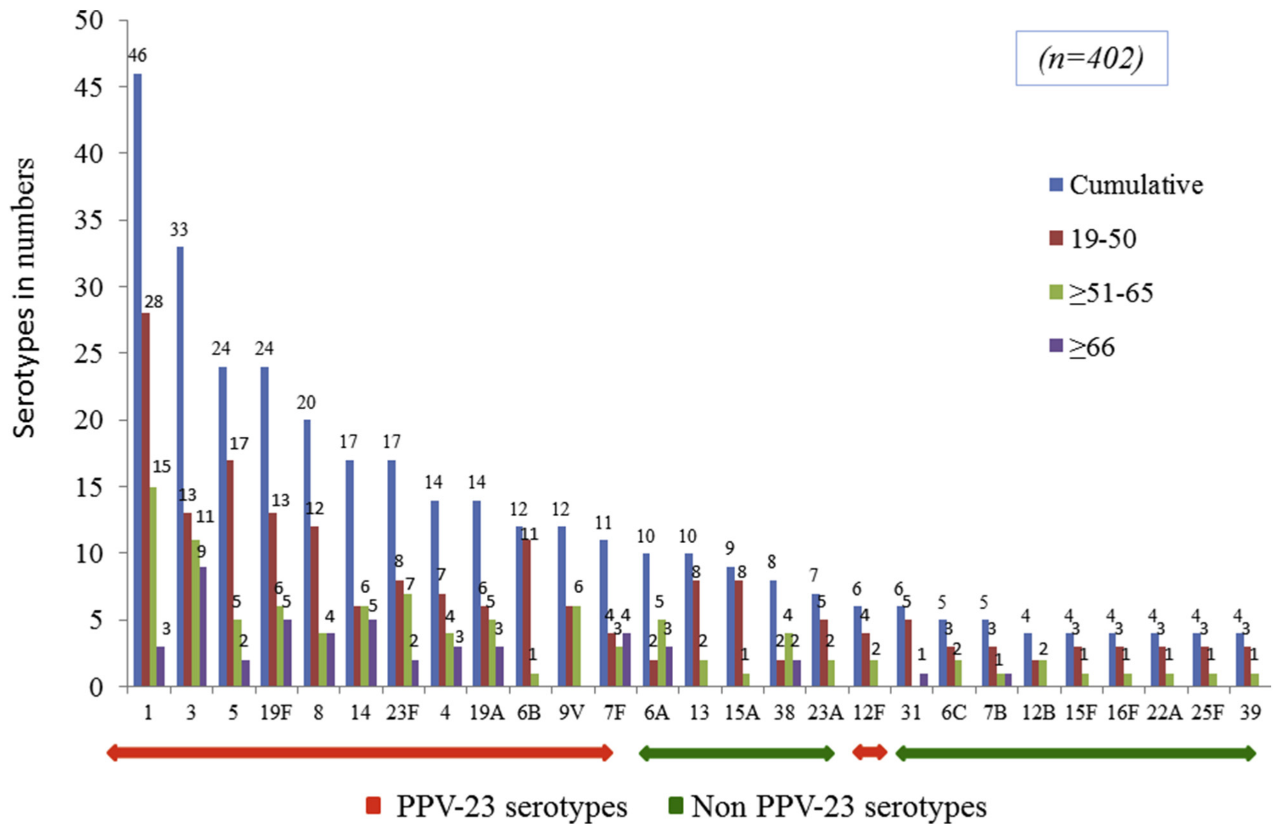

Figure 1. Serotype distribution in India in >18 years age group from January 2007 to July 2017. Data not shown in figure. Cumulative: Three isolates each of serotypes 9N, 10C, 11A, 17F, 18F, 23B, 33F, 35B and 45. Two isolates each of serotypes 2, 10A, 18A, 18C, 19B, 19C, 20, 21 and 34. One isolate each of serotypes 7A, 7C, 9B, 10B, 10F, 15B, 18B, 24, 28A, 28F, 29, 33B, 33C, 35A, 35F, 36, 37 and 48 and five isolates were non-typeable. 19–50: Three isolates of serotype 45. Two isolates each of serotypes 2, 9N, 10C, 11A, 19B, 23B and 35B, and one isolate each of serotypes 7A, 7C, 9B, 10A, 10B,17F, 18A, 18C, 18F, 19C, 20, 21, 28F, 29, 33C, 34 and 37 and three isolates were non-typeable. ≥51–65: Three isolates of serotype 33F, two isolates of serotype 17F, and one isolate each of serotypes 9N, 11A, 15B, 18B, 18C, 18F, 19C, 20, 21, 23B, 24, 33B, 34, 35B, 36, 48, and one isolate was non-typeable. ≥66: One isolates each of serotypes 10A, 10C, 10F, 18A, 18F, 28A, 35A and 35F and one non-typeable isolate.

and moderate risk groups for acquiring IPD was 23.7% (79/333), 40.5% (135/333), 18.3% (61/333) respectively (Table 1).

Serotype distribution

Serotypes 1, 3, 5, 19F, 8, 14, 23F, 4, 19A and 6B were the most common serotypes isolated in our study comprising 54.9% of the total study isolates. Fig. 1 illustrates the overall frequency of individual serotype distribution.

AST

In pneumococcal meningitis group, 27.4% and 9.9% isolates were non-susceptible to penicillin and cefotaxime respectively, while only <1% of non-meningeal pneumococcal isolates demonstrated penicillin and cefotaxime non-susceptibility. The overall non-susceptibility to erythromycin and co-trimoxazole was 27.2% and 93.8% respectively. Fig. 2 illustrates pneumococcal AST trends between January 2007 and July 2017.

Discussion

The existing recommendations for pneumococcal vaccines in Indian adults includes the use of PCV-13/PPV-23 in >65 years age group and in >18 years age group who are prone for pneumococcal diseases. However routine uses of these vaccines were limited largely due to inadequate data on pneumococcal disease burden and its serotype distribution. The data from this study are evident to consider pneumococcal vaccines in adults who are at risk of acquiring IPD.

IPD contributes significantly to infection related morbidity and mortality in India. Our study reports a case fatality rate of 34.9% in individuals aged ≥66 years, which indicates a rise in case fatality rate among older individuals compared to 1993–2008 report (26%)[8] is a matter of concern. The overall IPD associated case fatality rate in our study (17.8%) is however lower compared to previously reported Indian rates (25–30%)[8] and could be attributed to a general improvement in health care access in India in the past decade.

Patients with IPD presented with a variety of clinical syndromes, most common includes the respiratory and central nervous systems. It was interesting to note that

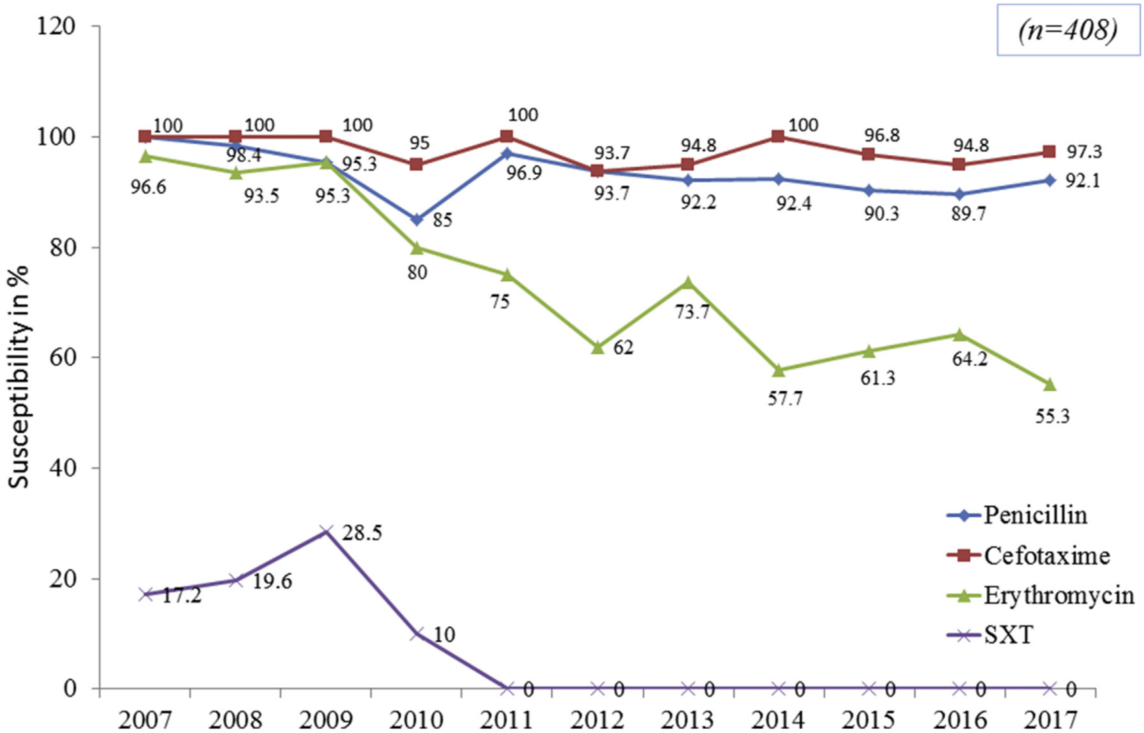

Figure 2. Antimicrobial susceptibility profile for the isolates in India in >18 years age group from January 2007 to July 2017.

there was significant increase in pneumococcal septicaemia cases during 2007–2017 with 18.4%, when compared to 9.4% of 1993–2008.[8] This could be due to the widespread use of inappropriate broad spectrum antimicrobials, especially fluoroquinolones and cephalosporins, without stringent adherence to regional antimicrobial therapy guidelines and infection control practices. This possibly results in the mystification of the primary infective focus at the time the patient is referred to a tertiary care centre for further management of septicaemia. Septicaemia patients with an unknown infective focus also had the highest case fatality rate (36.9%). This is in contrast to the earlier report on individuals with IPD wherein patients with meningitis had the highest case fatality rate (>35%).[8] However it is important to note that 8.2% (6/73; 95% CI: 3.8, 16.8) of meningitis patients in this study were discharged against medical advice and details of their outcomes were not available.

In this study penicillin non-susceptibility was 6.4% quite higher than the earlier report of 3% ($P < 0.001$).[8] However analysis within subgroup of pneumococcal meningeal and non-meningeal isolates revealed, 27.4% and 9.9% isolates from meningitis cases were non-susceptible to penicillin and cefotaxime respectively. Similar increase in non-susceptibility to penicillin and cefotaxime amongst pneumococcal meningeal isolates of children in the same period was also observed in our recent report.[20] The increase in penicillin non-susceptibility could be explained with increased use of amoxicillin for treating community acquired pneumonia[21] and also possibly because of higher penicillin non-susceptibility rates in neighbouring countries like Sri Lanka and other Asian countries. Pneumococcal resistant clones from these countries might evade Indian settings. Gopi et al. (2016) have shown Pneumococcal Molecular Epidemiology Network (PMEN) clones were already circulating in Indian invasive isolates.[22] This underlines molecular typing methods must be incorporated in surveillance systems to monitor resistance clonal spreads within and outside India. Erythromycin non-susceptibility rates in this study is 27.2%, comparing to <10% in the previous report from our centre.[8] This could be due to a longer elimination half-life of azithromycin expose to *S. pneumoniae* resulting in sub-inhibitory concentrations of antibiotics leading to antibiotic resistance.[23–25]

The most common ten serotypes observed in this study were 1, 3, 5, 19F, 8, 14, 23F, 4, 19A and 6B. These serotypes were accountable for 54.9% of study IPD cases. Remarkably the percentage of coverage by these ten prevalent serotypes was relatively less compared to our previous serogroup/type data. During 1993–2008, the ten most common serogroup/types were accountable for 69% of total IPD cases.[8] Also the most common serotype noted in this study, serotype 1 (11.4%) was reduced which was previously 22.9% of total IPD cases. Serogroups 7, 12 and 15 were amongst the ten most common serogroup/types in the earlier study, while serotypes 19F, 14 and 23F are the most prevalent serotypes in the present study.

Overall, PCV-13 and PPV-23 could possibly provide protective coverage against 58.7% (95% CI: 53.8, 63.4) and 67.4% (95%: 62.7, 71.8) of all serotypes responsible for IPD in India (Table 2) (Fig. 3). This is less when compared with our previous report wherein PPV-23 potentially provided protective coverage against 82% of serogroup/types causing IPD in the Indian adult population.[8] The apparent decline in serotype coverage could possibly be due to the availability of only serogroup data and not serotype data, unlike our

Table 2 Pneumococcal vaccine coverage in study participants.

	Age group (in yrs)				Overall (N = 402)		P value
	19–65 (N = 346)		≥66 (N = 56)				
	n	% (95% CI)	n	% (95% CI)	n	% (95% CI)	
PCV10	155	44.8 (39.6, 50.1)	24	42.9 (30.8, 55.9)	179	44.5 (39.7, 49.4)	0.7863
PCV13	197	56.9 (51.7, 62.0)	39	69.6 (56.7, 80.1)	236	58.7 (53.8, 63.4)	0.0732
PPV23	230	66.5 (61.3, 71.2)	41	73.2 (60.4, 83.0)	271	67.4 (62.7, 71.8)	0.3181

n represents vaccine serotypes. *N* represents cumulative serotypes.

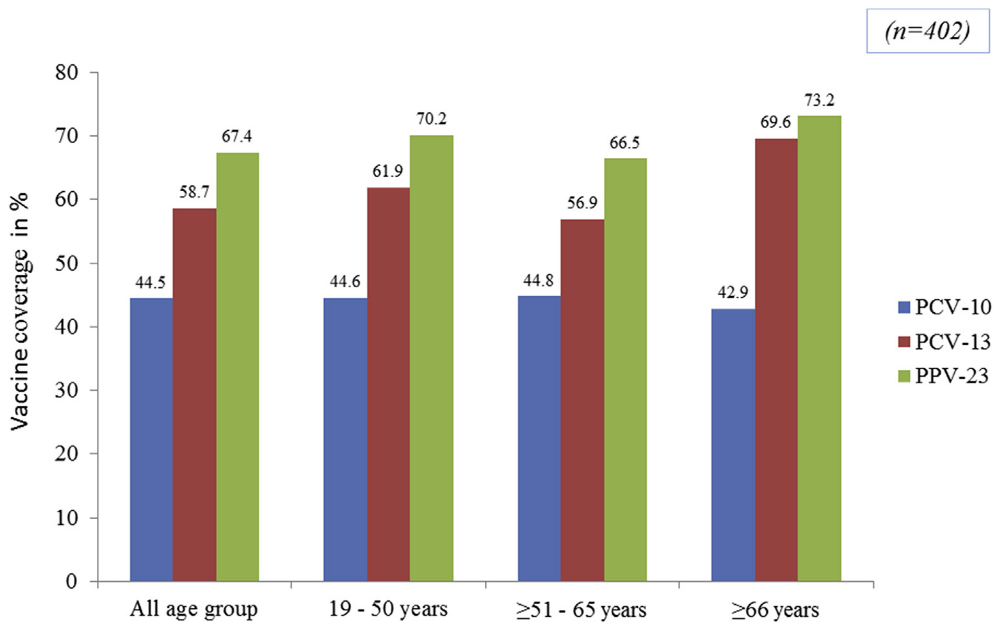

Figure 3. Expected PCV-13 and PPV-23 coverage for the IPD isolates in >18 years age group from January 2007 to July 2017.

study which used serotype data in addition to serogroup data. The presence of individual serotype data allows for an accurate estimation of the vaccine coverage against serotype causing IPD in India.

To conclude, this could be the first report on complete serotype data on pneumococcal vaccine coverage in Indian adults. PCV-13 and PPV-23 provide coverage of 58.7% and 67.4% for IPD respectively. Penicillin and cefotaxime non-susceptibility in meningeal IPD is increased gradually during past few years. This shows penicillin can be inefficient for pneumococcal meningitis and cephalosporin must be used. Since 9.9% cefotaxime non-susceptibility is seen in meningeal isolates, it is preferable to empirically start with vancomycin and cephalosporin.

Conflicts of interest

The authors declare that they have no conflict of interest.

Ethical approval

The study design and proposals were approved by the ethics committee and Institutional review board (IRB) of Christian Medical College, Vellore, South India.

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