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Original Article

Microbiological and clinical characteristics of Streptococcus pneumoniae serotype 3 infection and risk factors for severe outcome: A multicenter observational study

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Sequence type

Abstract *Background/purpose:* Serotype 3 has persisted to be an important cause of invasive pneumococcal disease in adults in the post-vaccine era. We aimed to investigate clinical and microbiological characteristics of *Streptococcus pneumoniae* serotype 3 infection in Taiwan and identify the risk factors associated with severe clinical outcome.
Methods: A multicenter observational study was conducted to analyze serotype 3 isolates collected between 2012 and 2021. Demographics, comorbidities, and risk categories were statistically compared with clinical outcome. Antimicrobial susceptibility testing and multilocus sequence typing were performed.
Results: A total of 146 isolates were collected, including 12 isolates regarded as colonizers. Among 134 infected cases, 54 (40.3%) were aged 65 and older. Mortality was significantly associated with diabetes mellitus, immunosuppression, immunodeficiency, high-risk status, and older age. Susceptibility rates were high to levofloxacin (98.9%), moxifloxacin (100%), vancomycin (100%), and ceftriaxone (97.3%). 25.3% (37/146) of the isolates showed intermediate susceptibility and 0.7% (1/146) showed resistance to penicillin. ST180 was the dominant sequence type. ST13 and ST9625 isolates were less susceptible to penicillin and ceftriaxone.

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Conclusions: Serotype 3 infection showed a high mortality rate, especially in patients with older ages and comorbidities. Although the incidence rates decreased during the COVID-19 pandemic, serotype 3 remained as an important cause of infection after the implementation of PCV13. Developing a more effective vaccine against serotype 3 and monitoring the antimicrobial-resistant sequence types are necessary.

Introduction

Streptococcus pneumoniae, the most important bacterial respiratory pathogen in humans, causes a range of mucosal and invasive infections in both children and adults. It is a significant cause of morbidity and mortality worldwide.[1] *S. pneumoniae* has more than 95 identified capsular serotypes which are the targets of current vaccines.[2] Although the introduction of universal immunization programs significantly reduced the number of infections due to vaccine serotypes, serotype 3 remains to be an important cause of pneumococcal disease in many countries.[3] Taiwan introduced the PCV13 catch-up program in 2013 and implemented the full national immunization program in 2015. However, serotype 3 has persisted to be an important cause of invasive pneumococcal disease in adults in the post-vaccine era.[4]

Distinct capsular synthase mechanism, profuse production of a thick capsule, and its unique interaction with the bacterial surface are considered the mechanism of vaccine escape of serotype 3.[2] Additionally, serotype 3 has been shown to be associated with more severe clinical manifestations and an increased fatality rate compared to other serotypes.[5] The impact of comorbidities on clinical outcomes and clinical characteristics of serotype 3 infections in children and adults remain unclear. In this study, we aimed to investigate the characteristics of *S. pneumoniae* serotype 3 isolated between 2012 and 2021 and determined the prognostic factors associated with severe clinical outcome.

Methods

Study design

Clinical information extracted from medical record systems included confirmed diagnosis, history of comorbid conditions, tobacco smoking history, alcoholism, length of hospital stay, development of complications, need of intubation or operation, ICU admission, readmission, and mortality within 30 days of attendance. Sepsis, septic shock, pleural effusion, empyema, acute respiratory failure, acute renal injury, and abnormal liver functions were recorded as complications.

Patients were divided into 3 groups according to risk status definitions of the U.S. Advisory Committee on Immunization Practices (ACIP).[6] Existing history of malignancy, asplenia, immunodeficiency, immunosuppression, chronic renal failure, solid organ transplantation, cerebrospinal fluid leak, cochlear implantation, or HIV was defined as high-risk status; for individuals without immunosuppressed or immunocompromised status, history of diabetes mellitus, chronic lung disease, chronic liver disease, chronic heart disease, alcoholism or smoking was defined as at-risk status; immunocompetent individuals without any of these comorbidities were defined as normal risk status (Supplementary Table 1).

Patients and isolates

This multicenter observational study approved by the Institutional Review Board of Chang Gung Memorial Hospital (approval number 202100795B0A3) was conducted on *S. pneumoniae* serotype 3 isolates cultured between January 1, 2012 and December 31, 2021. Patients were from four hospitals located in Linkou, Chiayi, Kaohsiung, and Keelung in Taiwan. For duplicating isolates from the same individual, only the first episode of serotype 3 isolation was included. Recovery of an isolate from a normally sterile site (blood, cerebrospinal fluid, pleural fluid) was considered as invasive pneumococcal disease. Recovery of an isolate from a non-sterile site (sputum, bronchial washing, middle ear fluid, vaginal secretions, sinus discharge, corneal culture) was considered as non-invasive disease. Isolation of *S. pneumoniae* without any clinical signs or symptoms of infection was considered as colonization.

Demographic and clinical data were collected from electronic medical records. Demographic information included age and sex; clinical information included confirmed diagnosis, history of comorbidities, developed complications during hospitalization and mortality. Patients were divided into normal risk, at-risk and high-risk groups based on their comorbidities according to the ACIP risk status definitions (Supplementary Table 1).

Antimicrobial susceptibility testing

Antimicrobial susceptibility to levofloxacin, moxifloxacin, vancomycin, penicillin, and ceftriaxone was determined using the disk diffusion method. The minimum inhibitory concentration (MIC) of penicillin and ceftriaxone was determined using the E-test (bioMérieux, Marcy l'Etoile, France). Penicillin and ceftriaxone susceptibility classified as susceptible (S; penicillin, $\leq$0.06 mg/L; ceftriaxone, $\leq$0.5 mg/L), intermediate (I; penicillin, 0.12—2 mg/L; ceftriaxone, 1 mg/L), and resistant (R; penicillin, $>$2 mg/L; ceftriaxone, $>$1 mg/L) according to CLSI standards.[7]

Serotyping and multilocus sequence typing

Isolates were serotyped by polymerase chain reaction (PCR) and by using antisera obtained from the Statens Serum Institut (Copenhagen).[8] Multilocus sequence typing (MLST) was performed by PCR-sequencing of 7 housekeeping genes (*aroE*, *gdh*, *gki*, *recP*, *spi*, *xpt*, and *ddl*). The sequences were compared with the information provided in the MLST database (http://pubmlst.org/spneumoniae/).

Statistical analysis

Categorical data were expressed as numbers (%). Continuous data were expressed as median (Interquartile range (IQR), 25th percentile to 75th percentile), except for MICs that were provided as geometric mean (range). The annual incidence of serotype 3 isolated cases was calculated and expressed as the number of cases/10,000 admissions.

The relationship between the outcomes and the attributable factors was evaluated by Chi-square test or Fisher's exact test for categorical variables, and Mann-Whitney U test or Kruskal-Wallis test for continuous variables. P-value <0.05 was considered as statistically significant. Analyses were carried out using IBM SPSS Statistics Version 23.

Results

Patients and isolates

A total of 146 nonduplicating isolates of *S. pneumoniae* serotype 3 were included during the 10 years; 15 were isolated in 2012, 15 in 2013, 12 in 2014, 21 in 2015, 24 in 2016, 29 in 2017, 14 in 2018, 12 in 2019, and 4 in 2020. There was no serotype 3 isolation in 2021. Invasive isolates accounted for 13.7% (20/146) of all serotype 3 isolates in this period. The annual incidence of total and invasive serotype 3 during the study period is shown in Fig. 1.

Most isolates were cultured from sputum (59.6%), pus (16.4%), and blood (12.3%). 12 were considered as colonized cases and excluded from the clinical outcome analyses (Supplementary Table 2). Information regarding bacterial and viral coinfection can be found in Supplementary Tables 3 and 4 Within the 132 infected cases, 14.2% of patients were children and 40.3% were aged >65 years. The median ages of children and adults were 7.33 and 64, respectively.

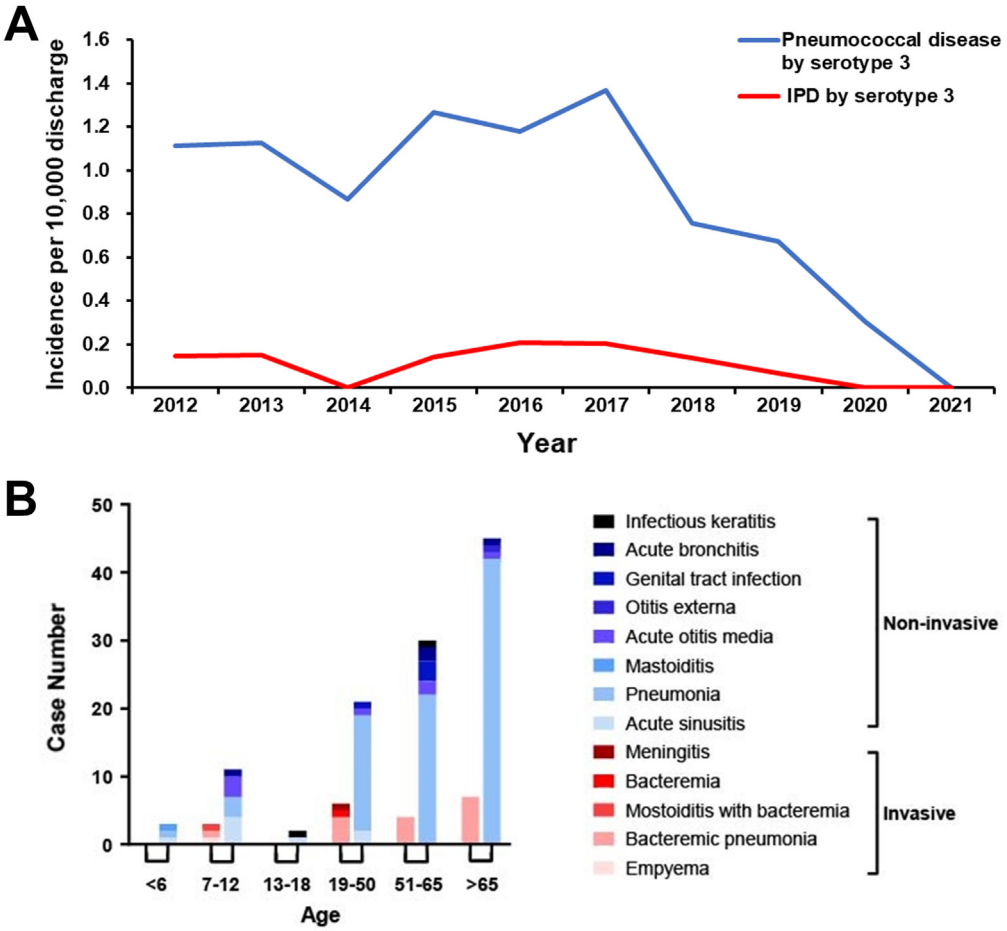

Figure 1. Age distribution and incidence rate of serotype 3 related infections. Annual incidence rate and age distribution of total and invasive pneumococcal disease caused by *Streptococcus pneumoniae* serotype 3. Annual incidence rate per 10,000 discharges in 2012—2021 (A) and age distribution and confirmed diagnoses of invasive and non-invasive pneumococcal disease (B) are demonstrated.

The male to female ratio was 1.73 (85/49). The number of invasive and non-invasive clinical diagnoses in different age groups is shown in Fig. 1. 67.2% of patients were hospitalized, whereas 32.8% were treated in outpatient clinics or in the emergency room.

The relationship between demographic and clinical factors and the severe clinical outcome was analyzed (Table 1). Gender and smoking status were not associated with the development of complication, need of intubation, ICU admission, and mortality. Whereas complication, ICU admission, and mortality were significantly different between patients in normal risk and high-risk status categories. Age was only associated with mortality which was significantly different between patients aged 18–65 years and >65 years. Among chronic diseases, diabetes mellitus was associated with complication, need of intubation, ICU admission, and mortality. Chronic kidney disease was found to be associated with complication and ICU admission. Immunosuppression and immunodeficiency were shown to be associated with only mortality. Detailed information regarding chronic diseases can be found in Supplementary Table 5. Within the study population, overall mortality was 9.7% (13 of 134), with 35% in patients with invasive disease (7 of 20).

Readmission and length of ICU stay were not found to be associated with any attributable factors (Table 2). Length of hospital stay was significantly different only between patients in at-risk and high-risk categories.

Multilocus sequence typing and antimicrobial susceptibility

There were 10 different sequence types (STs) among 146 isolates. The main sequence type was ST180 (90.4%, 132/146). Other STs included ST13 (n = 3), ST12902 (n = 3), ST505 (n = 2), and one isolates from each of ST458, ST9625, ST9628, ST12449, ST13739, ST15519. There was no significant statistical difference between STs in terms of invasiveness (p = 0.89). Majority of non-ST180 isolates were collected in 2016 and 2017 (Fig. 2).

0.7% (1/146) of isolates were resistant to penicillin and ceftriaxone. 25.3% (37/146) and 2.1% (3/146) of isolates have intermediate susceptibility to penicillin and ceftriaxone, respectively. Among isolates tested for vancomycin, levofloxacin, and moxifloxacin, there were no resistant isolates. Detailed information is shown in Supplementary Table 6.

Among 3 ST13, one had resistance to penicillin and intermediate susceptibility to ceftriaxone, one had resistance to ceftriaxone and intermediate susceptibility to penicillin, and the other had intermediate susceptibility to both antimicrobials. Similarly, ST9625 and ST9628 isolates showed intermediate susceptibility to penicillin (Fig. 3).

Discussion

We performed this longitudinal and observational multicenter study to estimate the potential prognostic factors of *S. pneumoniae* serotype 3 infections, and to monitor the sequence types and antimicrobial susceptibility between 2012 and 2021. Annual incidence rate of total serotype 3 and IPD cases demonstrated fluctuations between 2012 and 2017; however, it has begun to decline after 2017 with a more prominent decrease after 2019. The adoption of non-pharmaceutical interventions to prevent the spreading of COVID-19 in Taiwan appeared to be the reason of the rapid decline of serotype 3 cases after 2019 as shown in a recent study.[9] Notably, the incidence rate of serotype 3 increased from 2016 to 2017, despite the implementation of PCV13 in 2015. Similarly, a previous study in Taiwan showed that

Table 1 Different clinical outcomes of *Streptococcus pneumoniae* serotype 3 infections in relation to attributable factors of patients.

Factor	Outcomes								
	Number of cases	Complication		Intubation /operation		ICU admission		Mortality	
	n	n (%)	p value	n (%)	p value	n (%)	p value	n (%)	p value
All individuals	134	46 (34.3)		35 (26.1)		30 (22.4)		13 (9.7)	
Age, y			0.057		0.25		0.099		0.015 (18–65 vs. >65)
<18	19	5 (26.3)		7 (36.8)		4 (21.1)		–	
18-65	61	16 (26.2)		12 (19.7)		9 (14.8)		3 (4.9)	
>65	54	25 (46.3)		16 (29.6)		17 (31.5)		10 (18.5)	
Sex			0.76		0.46		0.68		0.77
Female	49	16 (32.7)		11 (22.4)		10 (20.4)		4 (8.2)	
Male	85	30 (35.3)		24 (28.2)		20 (23.5)		9 (10.6)	
Risk category					0.23				
No risk	47	9 (19.1)		11 (23.4)		7 (14.9)		1 (2.1)	
At risk	65	23 (35.4)	0.18 (No/At)	15 (23.1)		13 (20.0)	0.48 (No/At)	6 (9.2)	0.24 (No/At)
High risk	22	14 (63.6)	0.001 (No/High)	9 (40.9)		10 (45.5)	0.018 (No/High)	6 (27.3)	0.010 (No/High)
Cigarette Smoking			0.36		0.13		0.07		0.62
Never smoked	86	32 (37.2)		25 (29.1)		21 (24.4)		10 (11.6)	
Current Smoker	26	6 (23.1)		3 (11.5)		2 (7.7)		1 (3.8)	
Former Smoker	20	8 (40.0)		7 (35.0)		7 (35.0)		2 (10.0)	

Table 2 Length of hospital stay and length of intensive care unit stay in relation to attributable factors of patients.

Factor	Outcome							
	Inpatient cases	Hospital Stay (days)		Readmission		ICU admission	ICU stay (days)	
	n	median (IQR)	p value	n (%)	p value	n	Median (IQR)	p value
All individuals	90	8.50 (6–19)		7 (7.8)		30	6 (4–16.75)	
Age, y								
<18	9	19 (7.5–25)	0.094	–	1	4	12.5 (4.25–17.75)	0.51
18-65	39	8 (4–14)		3 (7.7)		9	11 (4.5–20)	
>65	42	8 (6–20.5)		4 (9.5)		17	5 (4–13)	
Sex								
Female	30	9.5 (7–23.25)	0.38	1 (3.3)	0.42	10	5 (4–13.5)	0.32
Male	60	8.5 (5–18.75)		6 (10)		20	9 (5–18)	
Risk category								
No risk	19	11 (6–19)		1 (5.3)	0.28	7	8 (3–18)	0.95
At risk	51	7 (5–11)	0.009	6 (11.8)		13	8 (5–15.25)	
High risk	20	22 (11.25–32.5)	(At/High)	–		10	5 (3.5–20)	
Cigarette Smoking								
Never smoked	55	9 (6–23)	0.53	2 (3.6)	0.08	21	6 (5–17.75)	0.92
Current Smoker	20	8 (6–13.25)		2 (10)		2	8.5	
Former Smoker	15	8 (5–27)		3 (20)		7	10 (2.5–26.5)	

serotype 3 was one of the most common serotypes for IPD in adults in the post-PCV13 era.[4] These results supported the hypothesis of less impact of PCV13 on serotype 3, compared to other vaccine serotypes as reported in other countries.[10–12]

While the majority (75.4%) had pneumococcal pneumonia, there was one case of meningitis among 132 included cases. We found that 14.9% of isolates caused invasive disease, with a similar rate in children (15.8%) and adults (14.8%). The increase in IPD and pneumococcal infections due to serotype 3 among old age suggest that serotype 3 is more likely to infect elderly people with comorbidities than young healthy adults.[13]

As our main outcome, the mortality rate of serotype 3 infection was 9.7% with a significantly higher rate (18.5%) in patients aged older than 65 years. Of the affected patients, 67.2% were hospitalized and 34.3% developed complications, such as sepsis, shock, pleural effusions, empyema, or ICU admission (22.4%). A previous study in Scotland reported a high mortality rate, and three serogroups, including serogroup 1, 3, and 16, were associated with mortality.[14] Similarly, a study from Germany stated that serotype 3 was a frequent cause of parapneumonic effusions and empyema among adults.[15] In Spain they reported that serotype 3 was more likely to cause septic shock compared to other serotypes.[13] In Colombia, they reported that major adverse cardiovascular events were more frequently seen in patients with serotype 3 IPD.[16] In this study we went beyond previous reports by analyzing the possible risk factors contributing to these serious clinical outcomes.

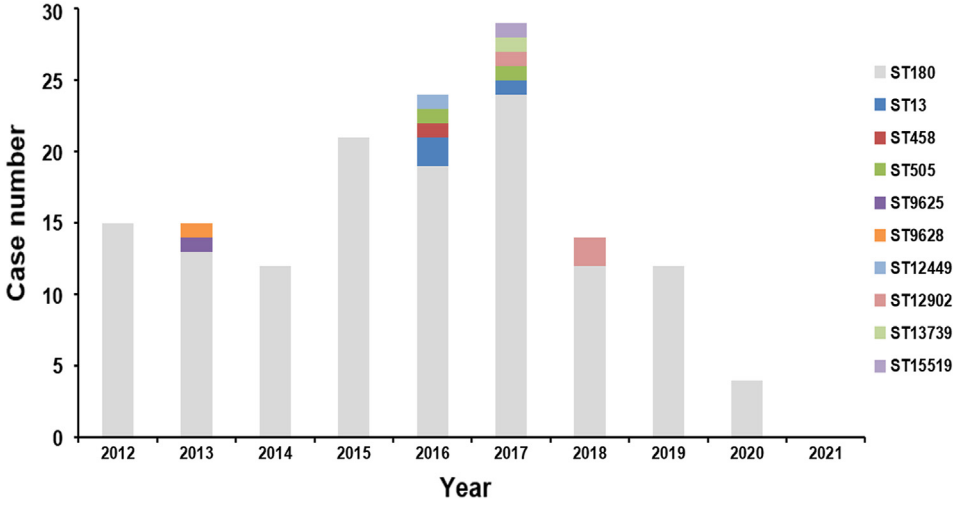

Figure 2. Annual distribution of sequence types. *Streptococcus pneumoniae* serotype 3 sequence types isolated between 2012 and 2021.

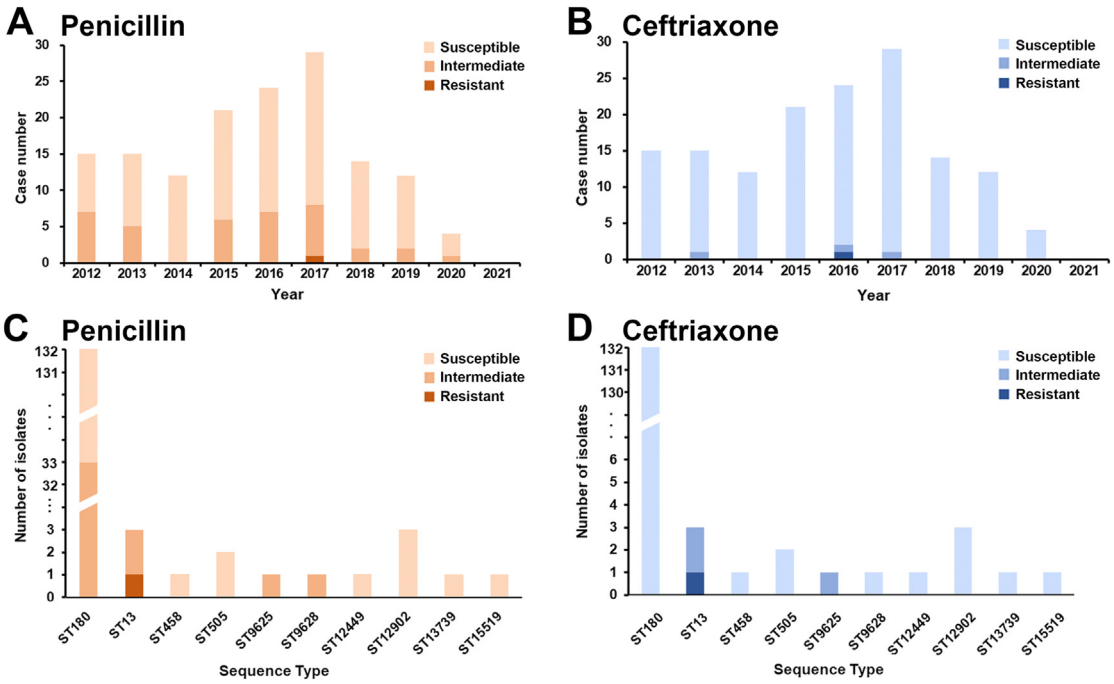

Figure 3. Antimicrobial susceptibility of serotype 3 isolates. Antimicrobial susceptibility of different sequence types of *Streptococcus pneumoniae* serotype 3. Penicillin (A) and ceftriaxone (B) susceptibility in 2012–2021 and penicillin (C) and ceftriaxone (D) susceptibility of different sequence types of *S. pneumoniae* serotype 3 isolates are demonstrated.

In addition to older age, the risk status of patients was found to be associated with increased risk of death. At-risk and high-risk status significantly increased the risk of developing complications, ICU admission, and mortality compared to normal risk status. Among the comorbidities that determined the risk status of patients, diabetes mellitus and chronic kidney disease were found to increase the risk of developing serious outcomes. Furthermore, diabetes mellitus, malignancy, and immunodeficiency were associated with increased risk of mortality in serotype 3 infections. Interestingly, although chronic lung disease was the most common comorbidity among patients, this study located no association between chronic lung disease and severe outcomes. In contrary to these findings, a recent study stated that chronic kidney disease was associated with increased risk of mortality, whereas diabetes mellitus was not associated with risk of death in pneumonia patients.[17] In another study, chronic pulmonary disease was a negative prognostic factor for bacteremic pneumonia-related mortality.[18] Although it is unclear that these differences could be attributed to serotype 3, the confounding effect of other comorbidities of patients might be the reason for the discrepancy between studies.

There was no difference in length of ICU stay and readmission between categories, and only at-risk and high-risk status groups were found to differ in the length of hospital stay. Shorter length of hospital and ICU stay of patients that expired in the hospital can be an explanation for these findings.

Among the isolates, the most common sequence type was ST180 (90.4%) followed by ST13, ST12902, and ST505. This finding is consistent with most of the global studies that ST180 was reported as the main sequence type.[3,19] However, ST505 was reported as a dominant sequence type among children in a recent study from China,[20] but ST505 was uncommon in Taiwan.

In previous studies, serotype 3 was highly susceptible to ceftriaxone, levofloxacin, moxifloxacin, and vancomycin.[21–22] However, isolates showed less susceptibility to penicillin. Importantly, all 3 isolates of ST13 showed either resistance or intermediate susceptibility to penicillin and ceftriaxone. ST9625 had intermediate susceptibility to both penicillin and ceftriaxone. In the review of the literature, no data was found to suggest this finding. However, ST242 and ST271 were found penicillin-resistant in two previous studies that they revealed that capsular switching had occurred between different *S. pneumoniae* serotypes by performing genome mapping on serotype 3.[23,24] Therefore, we hypothesized that our finding might be a result of capsular switching.

Our study has several strengths. We included all serotype 3 isolates from 4 hospitals in Taiwan. We had a relatively large sample size to analyze the microbiological and clinical characteristics of serotype 3. The study period of 10 years included both pre and post PCV13 era. As to limitation, we did not have the information of the immunization status of patients. Inclusion of vaccination status could provide better understanding of the effectiveness of PVC13 in preventing serotype 3 infection.

Conclusions

We observed a decreasing trend in the incidence rate of serotype 3 infection in Taiwan during the COVID-19 pandemic. Risk status of patients was found associated

with severe outcomes and mortality. Being older than 65 years old was another factor in increased risk of mortality. Although most of the isolates were susceptible to antimicrobial agents, we found that susceptibility of ST13 and ST9625 were relatively low against penicillin and ceftriaxone. Developing a more effective vaccine against serotype 3 and monitoring the antimicrobial-resistant sequence types are necessary.

Declaration of competing interest

All authors declare no competing interests.

Acknowledgements

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References

1. Dao TH, Rosch JW. JMM Profile: *Streptococcus pneumoniae*: sugar-coated captain of the men of death. *J Med Microbiol* 2021;**70**:001446.
2. Luck JN, Tettelin H, Orihuela CJ. Sugar-coated killer: serotype 3 pneumococcal disease. *Front Cell Infect Microbiol* 2020;**10**: 613287.
3. Groves N, Sheppard CL, Litt D, Rose S, Silva A, Njoku N, et al. Evolution of *Streptococcus pneumoniae* serotype 3 in England and Wales: a major vaccine evader. *Genes (Basel)* 2019;**10**:845.
4. Chien YC, Lee YL, Liu PY, Lu MC, Shao PL, Lu PL, et al. National surveillance of antimicrobial susceptibilities to dalbavancin, telavancin, tedizolid, eravacycline, omadacycline and other comparator antibiotics and serotype distribution of invasive *Streptococcus pneumoniae* isolates in adults: results from the Surveillance of Multicenter Antimicrobial Resistance in Taiwan (SMART) programme in 2017—2020. *J Glob Antimicrob Resist* 2021;**26**:308. 16.
5. van Hoek AJ, Andrews N, Waight PA, George R, Miller E. Effect of serotype on focus and mortality of invasive pneumococcal disease: coverage of different vaccines and insight into non-vaccine serotypes. *PLoS One* 2012;**7**:e39150.
6. Lewnard J, Bruxvoort KJ, Fischer H, Hong VX, Grant LR, Jódar L, et al. Effectiveness of 13-valent pneumococcal conjugate vaccine against medically-attended lower respiratory tract infection and pneumonia among older adults. *Clin Infect Dis* 2021:ciab1051. Available at: https://doi.org/10.1093/cid/ciab1051. [Accessed 10 August 2022].
7. Clinical and Laboratory Standards Institute (CLSI). *Performance standards for antimicrobial susceptibility testing*. 32nd ed. USA: Clinical and Laboratory Standards Institute; 2022, ISBN 978-1-68440-135-2. CLSI supplement M100.
8. Yun KW, Cho EY, Hong KB, Choi EH, Lee HJ. *Streptococcus pneumoniae* type determination by multiplex polymerase chain reaction. *J Korean Med Sci* 2011;**26**:971—8.
9. Janapatla RP, Chen CL, Dudek A, Li HC, Yang HP, Su LH, et al. Serotype transmission dynamics and reduced incidence of invasive pneumococcal disease caused by different serotypes after implementation of non-pharmaceutical interventions during COVID-19 pandemic. *Eur Respir J* 2021; **58**:2100978.
10. LeBlanc JJ, ElSherif M, Ye L, MacKinnon-Cameron D, Ambrose A, Hatchette TF, et al. *Streptococcus pneumoniae* serotype 3 is masking PCV13-mediated herd immunity in Canadian adults hospitalized with community acquired pneumonia: a study from the Serious Outcomes Surveillance (SOS) Network of the Canadian immunization research Network (CIRN). *Vaccine* 2019;**37**:5466—73.
11. Silva-Costa C, Gomes-Silva J, Prados L, Ramirez M, Melo-Cristino J. Portuguese Group for the Study of Streptococcal Infections, et al. Pediatric invasive pneumococcal disease three years after PCV13 introduction in the national immunization plan—the continued importance of serotype 3. *Microorganisms* 2021;**9**:1428.
12. Slotved HC, Dalby T, Harboe ZB, Valentiner-Branth P, de Casadevante VF, Espenhain L, et al. The incidence of invasive pneumococcal serotype 3 disease in the Danish population is not reduced by PCV-13 vaccination. *Heliyon* 2016;**2**:e00198.
13. Ahl J, Littorin N, Forsgren A, Odenholt I, Resman F, Riesbeck K. High incidence of septic shock caused by *Streptococcus pneumoniae* serotype 3A retrospective epidemiological study. *BMC Infect Dis* 2013;**13**:492.
14. Inverarity D, Lamb K, Diggle M, Robertson C, Greenhalgh D, Mitchell TJ, et al. Death or survival from invasive pneumococcal disease in Scotland: associations with serogroups and multilocus sequence types. *J Med Microbiol* 2011;**60**:793—802.
15. Goettler D, Streng A, Kemmling D, Schoen C, von Kries R, Rose MA, et al. Increase in *Streptococcus pneumoniae* serotype 3 associated parapneumonic pleural effusion/empyema after the introduction of PCV13 in Germany. *Vaccine* 2020;**38**:570. 7.
16. Africano HF, Serrano-Mayorga CC, Ramirez-Valbuena PC, Bustos IG, Bastidas A, Vargas HA, et al. Major adverse cardiovascular events during invasive pneumococcal disease are serotype dependent. *Clin Infect Dis* 2021;**72**:e711. 9.
17. Hespanhol V, Bárbara C. Pneumonia mortality, comorbidities matter? *Pulmonology* 2020;**26**:123–9.
18. Kalin M, Örtqvist Å, Almela M, Aufwerber E, Dwyer R, Henriques B, et al. Prospective study of prognostic factors in community-acquired bacteremic pneumococcal disease in 5 countries. *J Infect Dis* 2000;**182**:840–7.
19. Azarian T, Mitchell PK, Georgieva M, Thompson CM, Ghouila A, Pollard AJ, et al. Global emergence and population dynamics of divergent serotype 3 CC180 pneumococci. *PLoS Pathog* 2018; **14**:e1007438.
20. Yang Y, Hua CZ, Fang C, Xie YP, Li W, Fu Y, et al. Properties of mucoid serotype 3 *Streptococcus pneumoniae* from children in China. *Front Cell Infect Microbiol* 2021;**11**:648040.
21. Porat N, Soley C, Marengolciene MM, Greenberg D, Givon-Lavi N, Trefler R, et al. An international serotype 3 clone causing pediatric noninvasive infections in Israel, Costa Rica, and Lithuania. *Pediatr Infect Dis J* 2008;**27**:709. 12.
22. Sugimoto N, Yamagishi Y, Hirai J, Sakanashi D, Suematsu H, Nishiyama N, et al. Invasive pneumococcal disease caused by mucoid serotype 3 *Streptococcus pneumoniae*: a case report and literature review. *BMC Res Notes* 2017;**10**:1—6.
23. Chiba N, Murayama SY, Morozumi M, Iwata S, Ubukata K. Genome evolution to penicillin resistance in serotype 3 *Streptococcus pneumoniae* by capsular switching. *Antimicrob Agents Chemother* 2017;**61**:e00478. 17.
24. Scherer EM, Beall B, Metcalf B. Serotype-switch variant of multidrug-resistant *Streptococcus pneumoniae* sequence type 271. *Emerg Infect Dis* 2021;**27**:1689.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jmii.2023.01.013.