

## A Study Investigating the *Streptococcus Pneumoniae* Pattern Among Iranian patients

**A.Mehrabi.Tavana, R.A. Ataee, ME Gerami, M.M. Gooya, A Karami, R Ranjbar, M.Rahbar.M. Naderinasab & R Golmohammadi**

Molecular Biology Research Center & Health Management Research center Baqiyatallah (a.s) University of Medical Sciences Iran Ministry of Health the Director of Diseases management Control Center, Milad Hospital, Mashhad University of Medical Sciences, Tehran Iran.

**Citation:** A.Mehrabi.Tavana, R.A. Ataee, ME Gerami, M.M. Gooya, A Karami, R Ranjbar, M.Rahbar.M. Naderinasab & R Golmohammadi. **A Study Investigating the *Streptococcus Pneumoniae* Pattern Among Iranian patients.** Archives of Pharmacy Practice. 2012; 3(2) pp 122-127.

### Abstract

**Objectives:** The aim of this study was to determine the type of *Streptococcus pneumoniae* which is more prevalent in different diseases.

**Method:** 115 Clinical isolates of *Streptococcus pneumoniae* were re – identified and serotyped with Quellung test as a one of the serotyping methods based on SSI protocol - Statens Serum Institute. The data were inserted in to the computer and analyzed by SPSS version 17.0.

**Results:** The results of this study showed that 41% of isolate belongs to Respiratory Tract Infections. It was also known that 22% of isolate belongs to blood infection and 26% to eye infections. Finally 11% of isolates belongs to other infection in the patient too. The findings of this study showed that the types of 6A and 6B are more prevalent in patients totally. In addition, this study indicated that the type of 2 is more prevalent in adult patients.

**Conclusion:** This research revealed that types of 19F, 19A, 19B and 19C are more prevalent in Infant patients. This study also indicated that the most predominant types are in respiratory tract infections (group 7\*), eye infection (group 19\*), blood infection (type 5, 3, 6 and 4) and in CFS (type20) respectively. The diseases could be prevented by using the Pneumococcal vaccine.

### Key words

Patients, Public Health, Iran, *Streptococcus pneumoniae*

### Manuscript History

Article Received on: 15<sup>th</sup> Oct , 2011

Revised on: 23<sup>rd</sup> Jan, 2012

Approved for Publication: 30<sup>th</sup> Jan, 2012

### Corresponding Author

**Dr.A.Mehrabi.Tavana.** Molecular Biology Research Center & Health Management Research center Baqiyatallah (a.s) University of Medical Sciences Iran

Email: [mehrabi@bmsu.ac.ir](mailto:mehrabi@bmsu.ac.ir)

### Introduction

*Streptococcus pneumoniae* as a Gram positive diplococci is a major worldwide causative agent of morbidity and mortality among young children and the aged [1]. The main diseases which are caused by *Streptococcus pneumoniae* acute otitis media, pneumonia and Meningitis [2]. Pneumonia is the main infection of the *Streptococcus pneumoniae* which is an important to cure patients very quickly [3]. It has to be said that Pneumococci account for most cases of community-acquired pneumonia and the incidence of pneumococcal meningitis has been estimated to be close to 1.5 cases per 100,000 individuals per year, with an average mortality rate of 30 to 40% [4, 5]. More than 90 distinct capsular polysaccharides have been identified [6]. The identification of type of Pneumococcal is very important to find out the best vaccine in order to prevent the disease, there many research has been done in different part of the world in order to find out the Epidemiology and public health as well medical aspect of the disease [6]. Therefore, the *Streptococcus pneumoniae* typing has been performed in different countries in the last two decades [7-11]. We had no data regarding the distribution of Pneumococcal stereotyping in patients in Iran before this present study. Therefore; the study was performed to investigate the details of *Streptococcus pneumoniae* typing from 2009-2010 among different Iranian patients. This study was designed to determine any particular typing of Pneumococcal which are isolated from the patients who are admitted in different Hospital across the country.

### Materials And Methods

The present study was conducted on as a prospective study to type the profile of pneumococcal strains from infected Infants and Adults across 20 laboratories in Iran. We studied 115 Positive isolates of Pneumococcal from 2009-2010. These were positive isolates cultured from the nasopharynx, Blood, Sputum CSF and fluid of eye or lung.

#### Patient's selection

115 patients were recruited from among who were

admitted as Pneumococcal infection in 20 hospitals in Iran.

### Laboratory investigation

The patients samples (The nasopharynx swab, Blood, Sputum, CSF and fluid of eye or lung were obtained based on expert Physician treatment procedure and were sent to 20 Laboratories Hospital in order to obtain positive culture. It should be noted that the test organisms were identified as pneumococcal according to Standardized Procedure to culture and analyze ( $\alpha$ -hemolysis, colony morphology, optochin susceptibility, and bile solubility) [12&13].

### Transport Medium

The samples have been sent on the Blood agar Medium as Transport Medium, from each hospital Laboratory to central Laboratory immediately.

### Detection of Pneumococcal

As soon as we have received the positive samples from those Laboratories they were re-sub cultured again on Blood Agar and was incubated for 24 hours in 5%CO<sub>2</sub> at 37 °C. For the next 24 hours its Optochin sensitivity and bile solubility was tested. The Gram Staining was performed too. The positive results were noted.

### Quellung test

A reaction in which anticapsular antibodies bind to the capsule of a bacterium, resulting in the capsule to swell or become more visible, was done and it was seen under the microscope.

The pneumococcal isolates were then typed by immunological techniques according to Company Procedure (SSI, Denmark) and Conventional serotyping was performed by the standard capsular reaction test by using the chessboard system [14] and the test reaction was noted too.

### Statistical analysis

The data was inserted in the computer based on patient's information including (age, sex, site of isolations and type of specimens) and then collected data analyzed by SSPS version 17.0.

## Results

This was a prospective study to see the profile of pneumococcal strains from infected Infants and Adults across 20 laboratories in Iran. A total of 115 Pneumococcal types were included in the study. We have also analyzed the isolate based on demographic characteristics which is shown in Figures 1-2. As it could be showed in Figure 1. The clinical isolate of Pneumococcal can be recovered from different age groups (Infant and Adult) and in both sex too (see Fig.2) but the proportion of each individual type of Pneumococcal might be different. The Frequency of Pneumococcal types, site of isolation and the gender of patients were showed in Tables 1-3. The frequency of each type of Pneumococcal was showed. Types 6, 19, G, 2 are more prevalent (17.4%, 12.2%, 10.4%, 9.6 % respectively) according to this study findings. Based on our study the first significant findings were, Just we had 15 Pneumococcal types in adults whereas in Infants only 6 different types (5, 6, 14, 19 and G) were more prevalent (Figure 1).

Secondly, a few Pneumococcal types was not present in female (7, 8, 10, 20) when it is compare with male types (Figure 2). Thirdly, the distribution of 115 clinical Pneumococcal types is wide and the most type was 6 with 17.4% fourthly, the total distribution of Pneumococcal types more prevalent in Adult (75.7%) than Infant (24.3%). Fifthly, the lung and eyes were the site of isolation which the organism was recovers (57.6%) totally. Finally, the distribution of site isolation of organism may be different in male and female and finally the distribution were vary from Infant and Adults when compare with site of isolation (In Infant 81.5% organism has been isolated from eyes where as in Adults lung was the best site of isolation with 39.6% (Table 4).

## Discussion

In this study, we have serotyped 115 clinical isolates of *Streptococcus pneumoniae* because no work has been done in this area in Iran before this research. This study has indicated that types 6, 19, G and 11 are also more prevalent in the country. In addition, children under 2 years old type 19 is more prevalent, However type 6 is more prevalent in Adult. It should be added that types which were analyzed in this study are present in both sex (male and female) except types 10, 14, 18 and 22 which were present in female only. No published data were found to compare the data in national level but a few studies has been shown similar typing results in different country. In Denmark overall, 92% (93% blood, 87% CSF) of isolates and 94% of all childhood isolates belonged to the 23 vaccine types. In addition, In Denmark, the ten most frequently occurring types from children were (6A + 6B, 18C, 14, 1, 7F, 19F, 9V, 4, and 23F) covered 84% of the cases of bacteremia and meningitis. Pneumococcal strains from adults were types 1, 4, 14, 6A + 6B, 7F, 9V, 3, 12F, and 8 (in order of frequency) [15]. Distribution of Pneumococcal may be different country by country for this reason different vaccine (Heptavalent Pneumococcal Conjugate Vaccine, 7 valant or 13-valent pneumococcal conjugate vaccine – or even 23 valents has been applied in different countries in order to prevent the disease in different age groups [16-18]. Many studies are also mentioned on penicillin resistance Pneumococcal too (19) Further study must be done to find out the details particular for vaccination program in high risk groups.

## Conclusion

This study also indicated that the most predominant types are in respiratory tract infections (group 7\*), eye infection (group 19\*), blood infection (type 3, 5, 6 and 4) and in CFS (type 20) respectively. It seems that Pneumococcal polysaccharides vaccine (PPV), 23-valent could be covered all of types which are involved in patients in order to prevent the disease. Therefore, the vaccine can be recommended for high risk groups in the community in particular cardiac, renal failure, and elderly.

This public health intervention has been successes in different countries because of reduction of meningitis and septicemia related the disease. We hope this first study could be compelled with further studies.

### Acknowledgement

We wish to thank all those who helped us. Without them, we could not have completed this project. We also would like to thank Dr S.M.Zahraee and his co-worker in Iran Ministry of Health for their supports.

### Conflict of Interest

All the authors have no conflict of interest

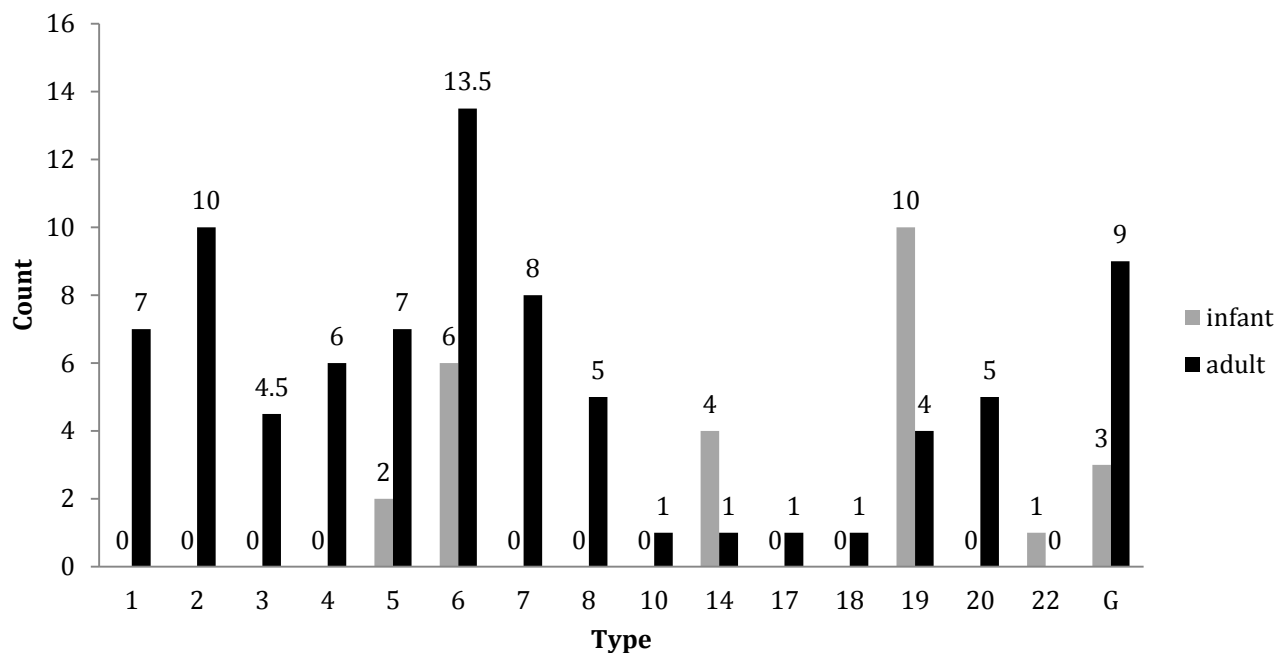
### Funding Disclosure

A part of funding for this project was supported by Iran Ministry of Health and Baqiyatallah (a.s) University of Medical Sciences.

### References

- Centers for Disease Control and Prevention.. Prevention of pneumococcal disease among Infants and young children: recommendations of the Advisory Committee on Immunization Practices (ACIP). *Morb. Mortal. Wkly. Rep*, 2000. 49:1-35.
- Roche PW, et al. Invasive pneumococcal disease in Australia, 2006. *Commun Dis Intell*. 2008; 32(1):18-30.
- Henrichsen J. Six newly recognized types of *Streptococcus pneumoniae*. *J Clin Microbiol* 1995; 33(10):2759-62.
- Brueggemann AB, et al.. Vaccine escape recombinants emerge after pneumococcal vaccination in the United States. 2007. *PLoS Pathog*. 3(11):e168.
- UFFE B. SKOV SORENSEN. Typing of Pneumococci by Using 12 Pooled Antisera. *JOURNAL OF CLINICAL MICROBIOLOGY*, Aug. 1993, p. 2097-2100.
- Moore MR, et al. Population snapshot of emergent *Streptococcus pneumoniae* serotype 19A in the United States. *J Infect Dis*. 2008.197:1016-27.
- Carvalho M dG, et al. Revisiting pneumococcal carriage using broth-enrichment and PCR techniques for enhanced detection of carriage and serotypes. *J Clin Microbiol*. 2010.48:1611-8.
- Feikin *et al*. High rate of pneumococcal bacteremia in a prospective cohort of older children and adults in an area of high HIV prevalence in rural western Kenya. *BMC Infectious Diseases* 2010, **10**:186.
- Bello González T, et al. Pneumococcal carriage in mothers and children of the Panare Amerindians from the State of Bolivar, Venezuela. *Rev Argent Microbiol*. 2010; 42(1):30-4. Spanish
- Lambertsen L, et al. Molecular characterization of invasive penicillin non-susceptible *Streptococcus pneumoniae* from Denmark, 2001 to 2005. *Scand J Infect Dis*. 2010 May; 42(5):333-40.
- Vestheim DF, et al. Caugant DA. Impact of a pneumococcal conjugate vaccination program on carriage among children in Norway. *Clin Vaccine Immunol*. 2010; 17:325-334.
- Murray PR, Baron EJ. Manual of clinical microbiology. 9th ed. / editor-in-chief, Patrick R. Murray; editors, Ellen Jo Baron. [et al.]. ed. Washington, D.C.: ASM; [Oxford: Blackwell, distributor]; 2007.
- Lund, E., and J. Henrichsen. 1978. Laboratory diagnosis, serology and epidemiology of *Streptococcus pneumoniae*, p. 242-262. In T. Bergan and J. R. Norris (ed.), *Methods in microbiology*. Academic Press, London, England. 18
- Sorensen, U. B. 1993. Typing of pneumococci by using 12 pooled antisera. *J. Clin. Microbiol*. 31:2097-2100.
- Sorensen, U. B. 1993. Typing of pneumococcal by using 12 pooled antisera. *J. Clin. Microbiol*. 31:2097-2100
- Nielsen SV, Henrichsen J. Incidence of invasive pneumococcal disease and distribution of capsular types of pneumococcal in Denmark, 1989-94. *Epidemiol Infect*. 1996; 117(3):411-6.
- Melegaro A, et al. Dynamic models of pneumococcal carriage and the impact of the Heptavalent Pneumococcal Conjugate Vaccine on invasive pneumococcal disease. *BMC Infect Dis*. 2010, 8; 10:90.
- Gessner BD, et al. African meningitis belt pneumococcal disease epidemiology indicates a need for an effective serotype 1 containing vaccine, including for older children and adults. *BMC Infect Dis*. 2010, 10; 10:22.
- Centers for Disease Control and Prevention (CDC). Invasive pneumococcal disease in young children before licensure of 13-valent pneumococcal conjugate vaccine - United States, 2007. *MMWR Morb Mortal Wkly Rep*. 2010, 12; 59(9):253-7.
- Chiba, N., et al. Serotype and antibiotic resistance of isolates from patients with invasive Pneumococcal disease in Japan. *Epidemiol infect*, 2010. 138(1).

**Figure1. Demographic characteristics of 115 clinical isolate of Pneumococcal based on age of patients and types of Pneumococcal**



**Figure2. Demographic characteristics of 115 clinical isolate of Pneumococcal based on gender of patients and types of Pneumococcal**

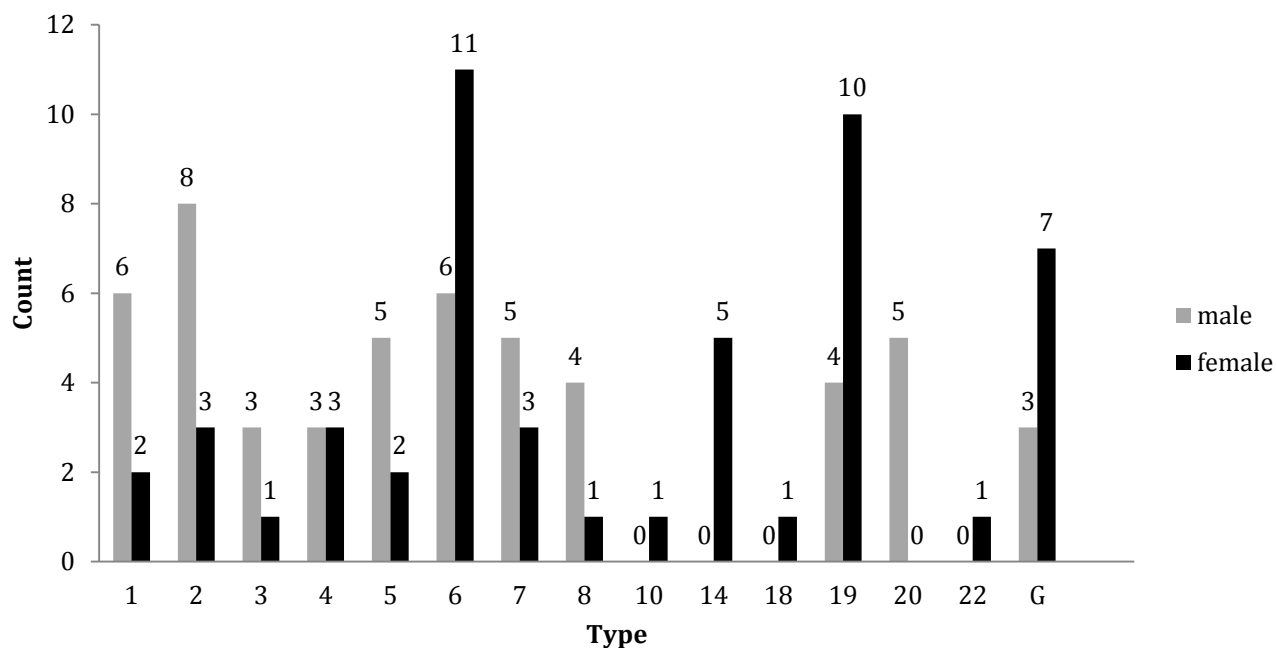


Table 1. The Frequency Of Pneumococcal Types In 115 Infected Patients		
Pneumococcal types	Frequency	%
1	9	7.8
2	11	9.6
3	4	3.5
4	6	5.2
5	9	7.8
6	20	17.4
7	8	7.0
8	5	4.3
10	1	0.9
14	5	4.3
17	1	0.9
18	1	0.9
19	14	12.2
20	5	4.3
22	1	0.9
G	12	10.4
Total	112	97.4
unknown	3	2.6
Total	115	100.0

G= one of the Pneumococcal type

Table 2 The Frequency of Pneumococcal based on site of isolation in 115 infected patients.		
Site of isolation	Frequency	%
eyes	30	26.1
Lung	34	29.6
wound	5	4.3
blood	17	14.8
CSF	2	1.7
other	23	20.0
Total	111	96.5
unknown	4	3.5
Total	115	100.0

**Table 3 The Frequency of Pneumococcal based on site of isolation on Gender**

Site of Isolation	Eyes	Lung	Wound	Blood	CSF	Other	Total
Male	4 (7.8%)	23(45.1%)	1(2.0%)	12(23.5%)	1(2.0%)	10(19.6%)	51
Female	25(47.2%)	9 (17.0%)	2(3.8%)	3(5.7%)	1(1.9%)	13(24.5%)	53

**Table 4The Frequency of Pneumococcal based on site of isolation and their age**

Site of Isolation	Eyes	Lung	Wound	Blood	CSF	Other	Total
Infant	22(81.5%)	1(3.7%)	0	2(7.4%)	0	2(7.4%)	27
Adult	8(30.0%)	33 (39.8%)	5(6.0%)	14 (16.9%)	2(2.4%)	21(25.3%)	83

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.