A comparison of the distribution of pneumococcal types in systemic disease and the upper respiratory tract in adults and children

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SUMMARY

The serotype distribution of 874 strains of *Streptococcus pneumoniae* was determined in relation to patients' age and to frequency of isolation from systemic disease. Types 14 and 18, in pre-school children, and types 1, 4, 7, 8 and 12 in patients over 5 years of age were significantly associated with systemic disease whereas type 23 in pre-school children, and type 6 in older patients was associated with upper respiratory tract carriage. No significant difference was found in the incidence of other types in systemic disease compared to upper respiratory tract carriage.

Fifteen diagnostic pneumococcal antisera (to types 1, 3, 4, 6, 7, 8, 9, 11, 12, 14, 17, 18, 19, 22 and 23) sufficed for typing 87 % of strains.

INTRODUCTION

Serotyping is an accurate and precise method for the identification of *Streptococcus pneumoniae* strains. It allows identification 24–48 h earlier than is possible by current bacteriological procedures for presumptive identification (e.g. optochin susceptibility), and also provides epidemiological information. Serotyping is rarely performed in diagnostic laboratories probably because of the need for a wide range of pneumococcal antisera and because it is usually carried out by the capsular reaction test. Coagglutination, which was used in this study, has been successfully adapted for serotyping *S. pneumoniae* strains (Kronvall, 1973; Trollfors *et al.* 1980; Smart, 1986). It is considerably easier technically, for routine laboratories, than the capsular reaction test.

Serotypes of *S. pneumoniae* from clinical material vary with the type of specimen and the age of the patient (Colman & Hallas, 1983; Broome *et al.* 1980). Here we compare the distribution of pneumococcal types in systemic pneumococcal infection to those isolated from the upper respiratory tract of patients of different ages to determine if particular types are associated with systemic disease.

MATERIALS AND METHODS

Bacterial strains

Streptococcus pneumoniae strains isolated in 15 bacteriology laboratories throughout Scotland (between January 1982 and December 1985) were sent to the Bacteriology Department, Stobhill Hospital, Glasgow for serotyping. Patients' identification, the source of the isolate and the clinical diagnosis accompanied each strain. Strains which were isolated from blood, cerebrospinal fluid, pleural fluid, post mortem lung tissue and other normally sterile body cavities were considered to be responsible for systemic disease. Strains isolated from the nasopharynx were regarded as upper respiratory, as were isolates from non-purulent sputa with obvious oro-pharyngeal contamination. The latter strains conformed to each of the following criteria.

(1) Strains isolated with mixed respiratory tract flora in numbers below 10^5 c.f.u./ml of homogenized sputum (Dixon & Miller, 1965).

(2) *S. pneumoniae* isolates from sputum in which no pneumococcal antigen was detected (Miller *et al.* 1978).

(3) *S. pneumoniae* strains isolated in absence of signs and symptoms of clinical infection.

S. pneumoniae isolates from ears and eyes were excluded.

Serotyping

Strains were serotyped by coagglutination as previously described (Smart, 1986) with reagents prepared from diagnostic pneumococcal antisera (Statens Seruminstitut, Copenhagen).

A heavy suspension in phosphate buffered saline (PBS, pH 7·2) was first tested with three polyvalent reagents prepared from antiserum pools A to I (Olcén, 1978) and then with the appropriate pools. The monotype or serotype was determined by testing with the single monotypic or serogroup reagent appropriate to the pool which gave the positive reaction.

For the purpose of this study the term serotype refers to both monotypes and serogroups. Details of serotyping within serogroups is the subject of a further report.

Statistical analysis

Serotype frequencies were compared using the χ^2 test in two by two contingency tables and incorporating Yates Correction for Continuity (Siegal, 1956).

RESULTS

The sources of 874 *S. pneumoniae* strains together with the age distribution of the patients are shown in Table 1. Three hundred and sixty were from cases of unequivocal pneumococcal disease and 514 were isolated from the upper respiratory tract. Blood culture was the most common source of strains from systemic disease (235); 67% of blood culture isolates were from patients over 50 years of age. Of the upper respiratory isolates, 73% were obtained from the nasopharynx of children under 5 years old whereas only 6% of the strains examined were from

Table 1. *Sources and age distribution of* Streptococcus pneumoniae *strains*

Source	Numbers of isolates and age group			
	< 5 years	5–50 years	> 50 years	Total
Blood	34	43	158	235
Cerebrospinal fluid	17	20	29	66
Pleural fluid and post-mortem lung	6	6	27	39
Others*	1	11	8	20
Total systemic disease	58	80	222	360
Nasopharynx	289	80	25	394
Commensal in sputum†	0	38	82	120
Total upper respiratory isolates	289	118	107	514

*Isolates from normally sterile sites, e.g. joint fluid and peritoneal fluid.

† Sputa with no pus in gram film; no infection noted in case sheet; no pneumococcal antigen detected in sputum; and mixed growth of *S. pneumoniae* and upper respiratory organisms isolated.
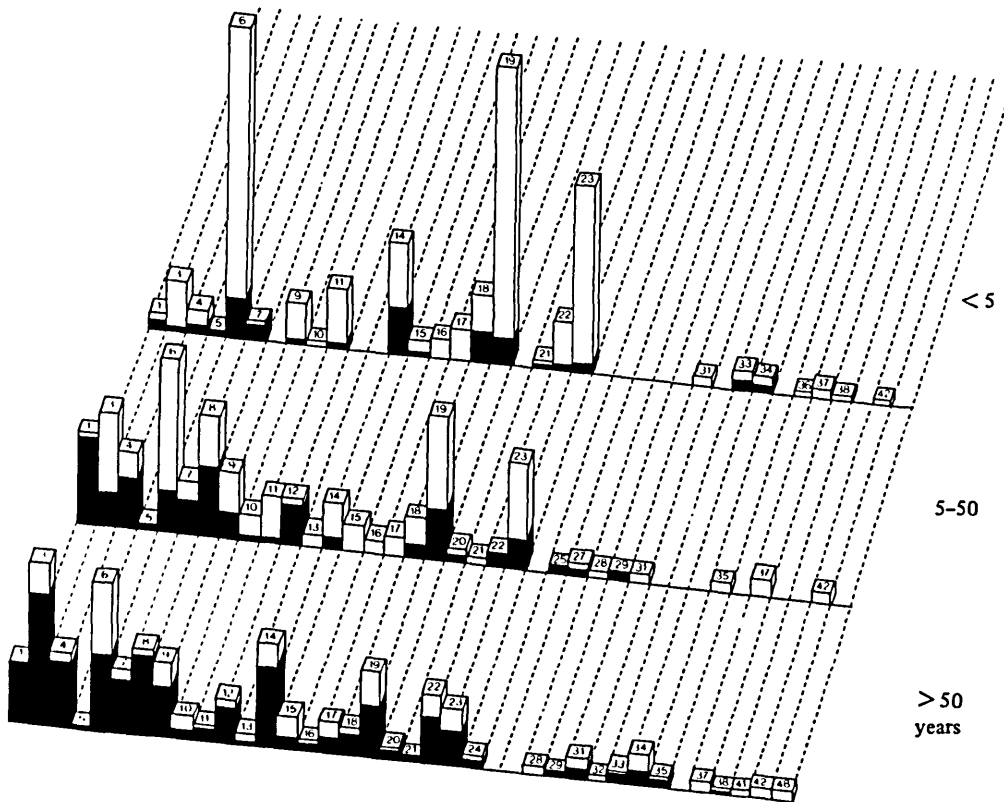

Fig. 1. Comparison of the distribution of pneumococci isolated from systemic infections (shaded area) and the upper respiratory tract (unshaded area) of patients in three age groups. Pneumococcal types are shown at the top of each column.

Table 2. *Frequency of the 15 most common types isolated from systemic disease and the upper respiratory tract*

	Systemic disease		Upper respiratory tract	
Type	Number of isolates	%	Number of isolates	%
1	30	8·3	3	< 1·0
3	38	10·5	32	6·2
4	25	6·9	11	2·1
6	35	9·7	105	20·0
7	22	6·1	6	1·2
8	29	8·0	10	1·9
9	17	4·7	21	4·1
11	5	1·4	21	4·1
12	16	4·4	3	< 1·0
14	34	9·4	27	5·3
17	4	1·1	15	2·9
18	15	4·2	15	2·9
19	26	7·2	89	17·3
22	14	3·9	17	3·3
23	14	3·9	63	12·3
Others	36	10·0	76	14·8
Total	360	100	514	100

Table 3. *The association of particular serotypes of* Streptococcus pneumoniae *with systemic disease and upper respiratory carriage*

	Upper respiratory carriage	No significant difference	Systemic disease	
P value*	< 0·05		< 0·05	< 0·001
< 5 years	23	3, 6, 9, 17, 19, 22		14, 18
> 5 years	6	3, 9, 11, 14, 17, 18, 19, 22, 23	4, 7, 8, 12	1

* Determined using χ^2.

patients over 50 years of age. In contrast, 68 % of sputum isolates were from patients over 50.

The distribution of types in relation to age (Figure 1) shows that the strains belonged to 38 types (20 monotypes and 18 serogroups). Isolates that belonged to type 24 and the higher types formed a small proportion of isolates in all age groups. Types 8 and 12 were not found in pre-school children but were found in both the upper respiratory tract and implicated in systemic pneumococcal infection in the other age-groups.

The frequency of the 15 most common types in systemic disease and the upper respiratory tract is shown in Table 2. We found types 6 and 19 were predominant amongst strains isolated from the upper respiratory tract and these were also amongst the most common types associated with systemic disease in all age groups. There was considerable variation in the distribution of types in each category. Types 1, 4, 7, 8 and 12 were isolated between 3 and 8 times more often from systemic disease than from the upper respiratory tract, whereas types 6, 11, 17,

Table 4. *Occurrence of the 15 most common types in descending order of frequency*

All strains	6, 19, 23, 3, 14, 8, 9, 4, 1, 22, 18, 7, 11, 12, 17	= 87%
< 5 years		
Upper respiratory tract	6, 19, 23, 14, 11, 3, 22, 9, 18, 17, 4, 1, 7	= 92%
Systemic disease	14, 6, 18, 19, 7, 23, 4, 1, 9, 11, 22, 3	= 88%
5–50 years		
Upper respiratory tract	6, 19, 3, 23, 8, 9, 11, 14, 4, 18, 7, 17, 1, 12	= 84%
Systemic disease	1, 8, 19, 4, 6, 12, 3, 7, 9, 23, 14, 18, 22, 11	= 93%
> 50 years		
Upper respiratory tract	6, 19, 3, 9, 22, 23, 14, 17, 4, 7, 8, 12, 18, 11	= 68%
Systemic disease	3, 14, 6, 8, 4, 1, 7, 9, 19, 22, 12, 23, 18, 17, 11	= 90%

19 and 23 were isolated from 2 to 3 times more often from the upper respiratory tract than from systemic disease.

Table 3 summarizes the statistical analysis of types associated with systemic disease and upper respiratory carriage respectively. Because too few naso-pharyngeal strains were available from older people, the serotype distribution was assessed in only two age groups. Namely pre-school children and older children and adults. In pre-school children serotypes 14 and 18 were isolated significantly more often from systemic disease than from the upper respiratory tract ($P < 0.001$). In older patients, however, the serotypes significantly associated with systemic disease were types 1, 4, 7, 8 and 12 ($P < 0.05$). Type 23 in pre-school children and type 6 in older patients was significantly associated with upper respiratory tract carriage ($P < 0.05$). No significant difference was found in the isolation frequency of other types from systemic disease compared to upper respiratory carriage.

DISCUSSION

The distribution of pneumococcal types isolated from cases of systemic disease in Scotland was similar to that reported from England (Colman & Hallas, 1983) and the USA (Broome *et al.* 1980). However, there were some differences. Type 12 was an important cause of systemic disease in Scottish adults but was an uncommon cause of systemic disease in England. The low incidence of systemic disease due to types 8 and 12 that we observed in children was also found by other workers (Colman & Hallas, 1983; Austrian, 1981) and was paralleled by a low rate of nasopharyngeal carriage in Scottish children.

The types most often carried in the upper respiratory tract in our survey were similar in all age groups and were the same as the commensal types reported in the USA (Kaiser & Schaffner, 1974; Gray, Converse & Dillon, 1979).

Statistical analysis of the distribution of types (summarized in Table 3) indicates three patterns of isolation. Types 1, 4, 7, 8 and 12 in children and adults and types 14 and 18 in children under 5 years of age are clearly associated with systemic disease and, as suggested by Austrian (1981) this may be related to the chemical composition of their capsular polysaccharide. Conversely, other serotypes notably type 23 in pre-school children and type 6 in older children and adults, were associated with nasopharyngeal carriage rather than systemic disease. The remaining types showed no statistically significant difference between frequency of

isolation from the nasopharynx and from systemic disease. These included type 3, well recognized as a virulent type and to be associated with high mortality (Austrian, 1979). Type 3 was the most common type isolated here from systemic disease although it ranked only third in order of frequency amongst isolates from blood and cerebrospinal fluid. It was isolated twice as often from systemic disease in patients over 50 years of age than from patients under 50 years. In contrast type 1, uncommon as a commensal, was isolated three times as often from systemic disease in patients less than 50 years old than from patients over 50 years.

Bacteraemia is seen in only a quarter of patients with pneumococcal pneumonia. Moreover, bacteraemia can be detected in patients who have no focus of infection and in whom the organism has presumably spread to the blood from the healthy nasopharynx (Austrian, 1981). Thus the definition of a strain as 'virulent' on the basis of isolation from blood may not be justifiable. Bacteraemia may more truly reflect the potential for exposure of an individual to that strain or immunological incapacity of the host.

Hodges & McLeod (1946) studied pneumococcal pneumonia which had a high attack rate amongst students in a US Army Air force residential technical school. They found that an increase in the incidence of pneumonia was preceded by a high carrier rate of the types causing infection among the students. Our observations suggest that in the community at large as well as residential institutions, seeding of the population with a particular type can result in its frequent appearance as a cause of systemic disease. Type 6 in pre-school children and, in older children and adults type 19 also, showed a relatively high incidence in both systemic disease and in nasopharyngeal carriage.

This study demonstrates that, for a few serotypes of *S. pneumoniae*, their frequency of isolation from cases of systemic disease is not directly related to their frequency of occurrence in the community. We also found it to be age-dependent. Nevertheless for the majority of types the incidence of systemic disease is proportional to their frequency of isolation from the nasopharynx. Further studies are clearly required to determine the mechanisms underlying these relationships. Since almost 90 % of *S. pneumoniae* strains in this study were serotyped by coagglutination using only 15 antisera of the 46 currently available commercially (Statens Seruminstitut, Copenhagen) (Table 4), we suggest that routine serotyping is no longer beyond the scope of diagnostic bacteriology laboratories and would contribute to our understanding of the epidemiology of pneumococcal infection – which remains a major cause of morbidity and death.

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