THE PENICILLIN SENSITIVITY AND PHAGE TYPES OF STAPHYLOCOCCI ISOLATED FROM HOSPITAL PATIENTS

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Barber & Whitehead (1949) studied the bacteriophage types of penicillin-resistant staphylococci isolated from patients in St Thomas's Hospital in the autumn of 1948. The present investigation is a similar study for comparative purposes carried out during the corresponding months of 1949.

METHODS

All specimens passing through the routine clinical laboratory which on culture gave colonies of staphylococci were investigated. A preliminary slide coagulase test using the method of Cadness-Graves, Williams, Harper & Miles (1943) was performed on all these staphylococcal colonies; if this test gave a doubtful or positive result, the specimen was reserved for further study. Ten to fifteen colonies were picked off from these cultures and streaked across plain agar ditch-plates, the ditch containing 10 units of penicillin per ml., and, if the culture was a pure growth of *Staphylococcus pyogenes*, the remainder of the growth was emulsified in peptone water and spread thinly over another penicillin ditch-plate so that an occasional penicillin-resistant colony could be detected in a predominantly penicillin-sensitive culture. The Oxford staphylococcus was included on all ditch-plates as a control.

After overnight incubation the ditch-plates were examined, and representative colonies were subcultured on to agar slopes and into broth; the broth culture was used for testing for coagulase by the method of Fisk (1940). If this test was negative the culture was discarded.

Coagulase-positive organisms were tested for the production of α-lysin by the method of Gillespie & Simpson (1948) and, if this was negative, by the method of McFarlan (1938). These strains were also phage typed by the method now in use at the Central Public Health Laboratory, Collingdale, using their twenty-five phage filtrates. This method is essentially the same as that described by Wilson & Atkinson (1945).

Streptomycin sensitivity was tested for by using a ditch-plate with 50μg. streptomycin per ml. in the ditch; the Oxford staphylcoccus was used as a control.

RESULTS

Penicillin sensitivity

Strains of *Staph. pyogenes* from 110 patients were examined. The reaction of these strains to penicillin is shown in Table 1. It will be seen that penicillin-resistant strains were isolated from 43 patients. In five of these patients penicillin-sensitive

strains of *Staph. pyogenes* were present in addition to penicillin-resistant strains. Thus, on the basis of their penicillin reaction, 115 strains of *Staph. pyogenes* were isolated from 110 patients.

Table 1. *The comparative incidence of penicillin-resistant strains of staphylococci isolated from in-patients and out-patients*

	No. of patients infected with staphylococci	No. of patients yielding penicillin-resistant strains		
		Penicillin-resistant strains only	Mixed penicillin-resistant and pencillin-sensitive strains	Total
In-patients	55	28	5	33
Out-patients	55	10	—	10
Total	110	38	5	43

All the penicillin-sensitive strains showed a sensitivity similar to that of the Oxford staphylococcus, whereas the penicillin-resistant organisms nearly always grew up to the edge of the ditch and, occasionally, on the ditch. Using the method of Barber (1947), all the resistant strains were shown to produce penicillinase.

Table 2 shows the frequency of isolation of these 115 penicillin-sensitive and penicillin-resistant strains of *Staph. pyogenes* from lesions of in-patients and out-patients. It will be seen that of 55 strains of staphylococci isolated from out-patients 10 were resistant to penicillin, while of 60 strains isolated from in-patients 33 were resistant.

Table 2. *The frequency of isolation of penicillin-sensitive and penicillin-resistant strains of* Staphylococcus pyogenes *from lesions of in-patients and out-patients*

Lesion	Penicillin-sensitive strains isolated from		Penicillin-resistant strains isolated from		Total
	In-patients	Out-patients	In-patients	Out-patients	
Chronic leg ulcers	3	21	—	2	26
Respiratory affections	4	—	9	1	14
Operative and other wounds	4	—	9	1	14
Boils, abscesses and carbuncles	6	9	7	2	24
Burns and dermatitis	2	6	2	—	10
Conjunctivitis	1	2	2	2	7
E.N.T.	5	4	2	1	12
Genito-urinary	2	3	2	1	8
Total	27	45	33	10	115

Table 3 shows the influence of previous penicillin therapy on the proportion of penicillin-resistant strains isolated. Penicillin had been administered to 38 patients within the previous 3 months. Of these 38 patients, 29 yielded penicillin-resistant strains. Of the 72 patients who had not recently had penicillin, only 14 yielded penicillin-resistant organisms. This is a reduction from approximately 76% to approximately 19%.

Table 3. *The influence of previous penicillin therapy on the proportion of penicillin-resistant strains of* Staphylococcus pyogenes

	Patients having had penicillin		Patients not having had penicillin	
	Yielding penicillin-resistant strains	Not yielding penicillin-resistant strains	Yielding penicillin-resistant strains	Not yielding penicillin-resistant strains
In-patients	26	4	7	18
Out-patients	3	5	7	40
Total	29	9	14	58

Phage types

All strains of staphylococci were tested first with phages in critical dilution; all strains not showing a reaction, trace reactions being ignored, were tested with undiluted phages. In the table of results (Table 4), related phages have been grouped together. All the staphylococcal strains allocated to any particular phage group do not necessarily give identical reactions. Thus, group 6/7/47 includes all strains of staphylococci reacting with the phages 6, 7, 47B, 47C, 2311 and 3298; group 3 includes strains reacting with phages 3A, 3B, 3C, and 51; group 29/31/52 includes strains reacting with phages 29, 29A, 31, 31A, and 52; while those placed in group 52A gave strong reactions with phage 52A only and no reaction, or at most trace reactions, with other phages.

Table 4. *The distribution of penicillin-sensitive and penicillin-resistant strains of* Staphylococcus pyogenes *among the various phage groups*

Phage group	Total number of penicillin-sensitive strains	Total number of penicillin-resistant strains	Total
6/7/47	15	18	33
52A	19	13	32
29/31/52	9	1	10
3	6	—	6
42	4	—	4
Non-typable	19	11	30
Total	72	43	115

Of the 115 strains of *Staph. pyogenes* isolated, 85 gave clear-cut reactions with the phages used; 41 with diluted phages and 44 only with undiluted phages.

Table 4 shows the distribution among the five phage groups. It will be seen that of the 32 penicillin-resistant strains of *Staph. pyogenes* which could be phage typed all but one fall into the two groups 6/7/47 and 52A.

The phage types of the staphylococci isolated from the five patients yielding both penicillin-resistant and penicillin-sensitive strains are shown in Table 5. From two patients the sensitive and resistant strains gave different phage reactions, from two other patients both strains could not be phage typed, and from one patient the sensitive and resistant strains were of the same phage type.

Table 5. *The phage type of penicillin-sensitive and penicillin-resistant strains of* Staphylococcus pyogenes, *each pair having been isolated from the same patient*

Patient	Phage type of sensitive strain	Phage type of resistant strain
76	52	47
79	N.T.*	N.T.
81	42B/47	47/3298
82	52A	52A
83	N.T.	N.T.

* N.T. = non-typable.

Streptomycin sensitivity

Of the 115 strains of *Staph. pyogenes* isolated, 104 strains were as sensitive to the action of streptomycin as the Oxford staphylococcus and 11 strains were more resistant. Of the 11 resistant organisms, seven were isolated from patients who had had streptomycin therapy.

α-Lysin production

Of the 115 strains of *Staph. pyogenes* isolated 110 strains were found to produce α-lysin; 95 were found to be positive using the method of Gillespie & Simpson (1948), but 15 strains which gave negative results by this method were found to be α-lysinogenic by the method of McFarlan (1938). Neither method was able to demonstrate the production of α-lysin by five of the strains.

DISCUSSION

During the year since Barber & Whitehead (1949) investigated penicillin-resistant strains of *Staph. pyogenes* at St Thomas's Hospital, there has been no significant increase in the percentage of penicillin-resistant strains isolated, the actual percentages being 31·5% in 1948 and 37·4% in 1949.

In the present series, 11 strains of *Staph. pyogenes* were found to be resistant to streptomycin, whereas a year previously none were streptomycin resistant.

The most striking change in the present series, compared with that of 1948, is the increase in the number of penicillin-resistant staphylococci of phage type 52A. In 1948 only four out of 35 of the resistant strains were of this type, whereas in the present series 13 out of 32 are of phage type 52A.

This increase in the frequency of isolation of staphylococci of phage type 52A may be due to the introduction of a nasal carrier of these organisms. For the past year a nearby maternity hospital, where many of the staff are known to be carriers of penicillin-resistant staphylococci of phage type 52A (Barber, Hayhoe & Whitehead, 1949), has become more closely associated with St Thomas's Hospital. During the period of the present investigation, 17 strains of *Staph. pyogenes*, all penicillin resistant, were isolated from patients at this maternity hospital, and of these 12 were of phage type 52A.

SUMMARY

1. A series of 110 patients with staphylococcal infections has been investigated. From 43 patients a penicillin-resistant staphylococcus was isolated: in cultures from 38 patients all staphylococcal colonies tested were penicillin resistant and in five both sensitive and resistant strains were isolated. Seven patients yielded a streptomycin-resistant staphylococcus.

2. Of the 84 strains of staphylococci which could be phage typed, 33 belonged to the group of phages 6/7/47 and 32 were of type 52A. Of the 32 typable penicillin-resistant strains 18 belonged to the group of phages 6/7/47, and 13 were type 52A.

3. Of the 115 strains isolated 110 were α-lysinogenic.

4. These results are compared with those found a year previously and the significant changes are discussed.

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