Classification of staphylococci isolated from ocular tissues

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SUMMARY A total of 145 strains of Gram-positive, catalase-positive, coagulase-positive, and coagulase-negative cocci isolated from the human eye were classified by the system of Baird-Parker (1966). These belonged to subgroups I, II, IV, and V and showed a wide range of antibiotic sensitivities unrelated to subgroup and category of lesion. The role of coagulase-negative staphylococci in producing postoperative ocular infections has been especially emphasised.

Staphylococci are the pathogens or potential pathogens most commonly isolated from ocular tissues. The role of *Staphylococcus aureus* in producing pathology in any organ of the body, including the eye, is not disputed. The acceptance of *Staphylococcus albus* as a pathogen in different sites in the human body is gradually growing (Meers *et al.*, 1975). It has been reported as the second most common pathogen in bacteriologically proven cases of urinary tract infection, and novobiocin-resistant micrococci have been claimed to have a special predilection for the urinary tract of human beings (Gallagher *et al.*, 1965; Meers, 1974; Maskell, 1974). Such cocci have also been recovered from lesions of the eye which regressed when appropriate antimicrobial agents were administered (Khosla *et al.*, 1964; Valenton *et al.*, 1973). An attempt has been made in this study to collect staphylococci from the human eye and to classify them by Baird-Parker's (1966) criteria because, to the best of our knowledge, this has never been done.

Material and methods

Conjunctival swabs were collected either for determining the preoperative conjunctival flora or for isolating various pathogens producing ocular lesions. Preoperative swabs were collected from individuals who came mostly for cataract extraction. Swabs were collected on the day of admission to hospital. Postoperative infections included stitch abscesses, endophthalmitis, panophthalmitis, infection of corneal grafts, and wounds resulting from surgery in hospital. Cadaver eyes were collected

from the mortuary to be used for keratoplasties. In the miscellaneous category were included cases of dry eye syndrome.

Swabs were streaked immediately on sheep blood agar medium. Staphylococci were then subcultured on tryptone yeast extract agar slants and incubated at 37°C for 24 hours. Aerobic and anaerobic acid production from glucose was determined in 6 × ½ in tubes after five days' incubation at 37°C. Further division was then made according to Baird-Parker (1966) with carbohydrate reactions and phosphatase production carried out on solid media in Petri dishes. Acetoin production was tested using Barritt's method after 14 days' incubation at 30°C. The sensitivity of these organisms to penicillin, tetracycline, chloramphenicol, streptomycin, erythromycin, framycetin, and gentamicin was tested by the disc diffusion technique, and the results were interpreted as recommended by Bauer *et al.* (1966). Those found to be resistant were tested for sensitivity to cloxacillin.

Results

A total of 145 strains of staphylococci were classified into various Baird-Parker subgroups. Fifty-seven were positive for tube coagulase test (subgroup I) and the remaining 88 strains were negative. Twenty-seven (47.4%) were lactose-negative, and an additional five did not produce acid from both lactose and maltose. Thus only 44% of strains showed the classical reactions of *Staph. aureus*. The clinical categories of the patients and the corresponding subgroups of the strains are shown in Table 1. In subgroup I, an almost equal number of strains were isolated from hospital inpatients (30) as from

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Table 1 Correlation of clinical categories with biotypes of strains of staphylococci isolated from 145 patients

Clinical category	Baird-Parker			Subgroup
	S I	S II	S IV	S V
Preoperative	13	18	4	10
Postoperative infection	12	12	5	7
Contracted socket	4	1	1	—
Conjunctivitis	15	3	2	—
Corneal ulcer	7	6	3	8
Dacryocystitis	1	1	—	—
Meibomitis	1	2	—	—
Cadaver eye	1	—	—	—
Miscellaneous	3	2	1	2
Total	57 (30)	45 (33)	16 (10)	27 (18)

Numbers in parentheses denote strains isolated from patients admitted to hospital.

outpatients (27), whereas in the remaining subgroups the former (61) outnumbered the latter. In this category, S II strains greatly predominated (45/88), followed in order of frequency by subgroups V (27) and IV (16) in all categories of lesions except corneal ulcers where there were more S V strains.

The staphylococci isolated showed a wide range of antibiotic sensitivities. These are related to the subgroups in Table 2. Only seven strains out of 145 were sensitive to all the antibiotics tested. Most (90.4%) of the strains were resistant to penicillin, followed in order of occurrence by tetracycline, chloramphenicol, streptomycin, and framycetin. Gentamicin-resistant strains (12%) were the least common. Eighty-five (59.0%) strains were resistant to penicillin, tetracycline, and chloramphenicol whereas

only 10.0% were resistant to penicillin, framycetin, and gentamicin. Out of the strains resistant to all the antibiotics, eight were found to be sensitive to cloxacillin only. Antibiotic sensitivities did not appear to be related in any way to particular staphylococcus subgroups.

Discussion

The conjunctival sac harbours bacteria from the time of birth throughout life. Several studies of the flora of healthy eyes from children and adults have been reported. The bacteria commonly found at all ages were staphylococci and diphtheroids; streptococci and pneumococci were rarely present (Locatchar-Khorazo and Seegal, 1972). Staphylococci isolated preoperatively by us belong strictly to the category of normal commensals. Thirteen out of 45 strains belonged to subgroup I staphylococci, which have proven pathological potential. Most non-pathogenic staphylococci isolated belonged to subgroups II (18/32) followed by S V (10/32) and S IV (4/32). These findings are not surprising because the major source of bacterial flora of the eye is the skin of the face and the hands. Noble (1969) reported that S II strains are the predominant flora of the skin. He maintained that S II strains resemble *Staph. aureus* in their great propensity to colonise the nose, which is an important source of ocular staphylococci. Their preponderance in postoperative infections therefore is not surprising. We have classified 36 strains from lesions such as endophthalmitis, panophthalmitis, stitch abscesses, and postoperative infections of corneal grafts and found that 24 of these belonged to subgroups I and

Table 2 Antibiotic resistance patterns of staphylococci isolated from eye

Baird-Parker subgroup	Sensitive to all	P	S	T	C	E	F	G	PTC	PF	PG	PFG
S I <i>Staph. aureus</i> (n = 57)	3	50	25	32	25	33	20	12	26	20	14	13**
S II <i>Staph. albus</i> (n = 45)***	1	43	28	36	33	29	6	4	31	6	4	1*
S IV <i>Staph. albus</i> (n = 17)	2	13	8	14	11	6	0	2	9	0	2	0
S V <i>Staph. albus</i> (n = 26)	1	25	19	23	20	13	4	0	19	4	0	0
Total (n = 145)	7 (5)	131 (90)	80 (55)	105 (72)	99 (68)	81 (56)	30 (21)	18 (12)	85 (59)	30 (21)	20 (14)	14 (10)

***Only seven strains were tested for sensitivity to novobiocin. All were sensitive.

**Only eight were tested for cloxacillin and were sensitive.

*Sensitive to cloxacillin only.

Abbreviations: P = penicillin, S = streptomycin, T = tetracycline, C = chloramphenicol, E = erythromycin, F = framycetin, G = gentamicin. (Figures in parentheses are percentages.)

II. We agree and support the view of Mitchell (1968) and several others that staphylococcus subgroups II and VI can act as relatively feeble secondary pathogens, which require a nidus of damaged tissues before they can become established. Two years ago we conducted an experimental study in which rabbit cornea was traumatised to a depth of 0.2 mm, and a saline suspension of *Staph. albus* was rubbed in. All the animals developed classical hypopyon corneal ulcers, which yielded a pure growth of the infecting organism even after 16 days of infection (Agarwal *et al.*, 1976). We therefore investigate routinely all cases of corneal ulcers for coagulase-negative staphylococci and report the sensitivities of those isolates.

In this series, resistance of strains to penicillin and other antibiotics has been more frequent than in previous studies (Corse and Williams, 1968; Mitchell, 1968). These workers found 53% to 57% of their strains resistant to penicillin compared with over 90% in this series. Simultaneous resistance to several antibiotics was also more frequent in our study. Modification of antibiotic sensitivities may have occurred because of previous chemotherapy but this probability was not seriously studied. Whatever the reason for the emergence of resistant sensitivity patterns, such staphylococci pose an alarming problem to ophthalmologists. It is recommended that repeated isolations of staphylococci (including those that are coagulase-negative) from a single site should be tested for sensitivity to antibiotics, and the infection should be treated accordingly.

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