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Full Length Research Paper

Antibiotic sensitivity of microorganisms isolated from explanted sclera buckles

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The purpose of the present study was to investigate the current microbiological spectrum of culture-positive explanted scleral buckles and their sensitivity to antibiotics. A retrospective, consecutive case series was performed in this study. 38 patients with scleral buckle removal that occurred between January 2003 and December 2010 were retrospectively analyzed. Cultured bacteria were identified using the API system. Susceptibilities of the bacteria were tested using the Kirby-Bauer disk diffusion method. 38 of 50 (76%) explanted scleral buckles were positive by culture. A total of 40 microorganisms were isolated from the 38 buckles. Thirty isolates (75%) were Gram positive and 10 (25%) were Gram negative. Coagulase-negative *Staphylococcus* (CNS) was the most common isolate (62.5%) while 10 cases (40%) of CNS were methicillin-resistant CNS (MR-CNS). 100% of the Gram-positive isolates were sensitive to Vancomycin, 69% to Clindamycin, 59.3% to Gentamicin, 39.3% to Ciprofloxacin and 42.3% to Ofloxacin. MR-CNS was sensitive to Vancomycin and Rifampin, while 80% of the Gram-negative isolates were sensitive to Ciprofloxacin and 75% to Ofloxacin. This study demonstrated a change in bacterial isolates of the explanted scleral buckles and their antibiotic sensitivities, compared with previous reports. No single antibiotic can cover all of the microbes isolated from explanted scleral buckle. A combination of various antibiotics is recommended for the treatment of perioperative infection and initial empiric treatment.

Key words: Microorganism, sclera buckle, antibiotic sensitivity, infection.

INTRODUCTION

Scleral buckle placement is a common procedure for the treatment of rhegmatogenous retinal detachment. Infection, erosion, intrusion, and extrusion are common complications following surgery (Lorenzano et al., 2007). These complications remain as significant problems resulting in recurrent retinal detachment after buckle removal. Buckle infection and extrusion are more common with an acceptable rate of 0.5 to 5.6% (Wirostko et al., 2009). Strategies for reducing the incidence and treatment of scleral buckle infection include the use of preoperative prophylactic antibiotics to the ocular surface and scleral buckle, intraoperative scleral irrigation using prophylactic antibiotics prior to closing of the conjunctiva and initial empiric antibiotic treatment. Pathengay et al. (2004) reported the microbial spectrum and susceptibility of isolates from patients undergoing buckle removal between 1992 and 2002. In this report, they showed that ~50% of the isolates were Gram-positive and *Staphylococcus epidermidis* was the most common organism. 73% of Gram-positive cocci and 86% of Gram-negative bacilli were sensitive to ciprofloxacin, which was recommended for the initial treatment of buckle infection. Frequent use of antibiotics has led to the development of bacterial strains resistant to many commonly used antibiotics. Several recent reports have shown that the growing bacterial resistance to commonly used antibiotics is present in ocular infections (Melo et al., 2011; Brown, 2007; Adebayo et al., 2011). Therefore, the antibiotic choice should be based on regional and periodic microbial spectrum and susceptibility testing. However, recent information is lacking on the bacteriology and antibiotic sensitivities.

This study was designed to describe current bacteriology and antibiotic sensitivities of sclera buckles infection in China and to determine the best suited

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Table 1. The list of isolated organisms from 2003 to 2010.

Isolated microorganisms	2003	2004	2005	2006	2007	2008	2009	2010
Gram-positive organisms								
1. MR-CNS (n=10)		1		2	1	1	2	3
2. MS-CNS (n=15)	2	1	2	1	3	3	2	1
3. α-hemolytic streptococcus(n=3)		1	1		1			
4. Staphylococcus aureus(n=2)						1	1	
Gram-negative organisms								
1. Klebsiella pneumoniae (n=3)		1	2					
2. Pseudomonas aeruginosa (n=3)		1	1	1				
3. Pseudomonas alcaligenes (n=1)					1			
4. Acinetobacter lwoffii (n=1)	1							
5. Serratia marcescens(n=1)						1		
6. Xanthomonas maltophilia(n=1)	1							
Total	4	5	6	4	6	6	5	4

MR-CNS=Methicillin-resistant Coagulase-negative Staphylococcus; MS-CNS= methicillin-sensitive Coagulase-negative Staphylococcus

antibiotics for the treatment of buckle infection.

MATERIALS AND METHODS

We retrospectively examined 38 cases of culture-positive explanted scleral buckles from 50 patients who underwent buckle removal between January 2003 and December 2010 at the Department of Ophthalmology of the Second Xiangya Hospital. Of these patients, 60.5% (23/38) were male and 39.5% (15/38) were female. Age ranged from 4 to 82 years old. The time period from implantation to removal ranged from 1 to 83 months. The patients were subjected to scleral buckle removal followed by scleral irrigation using antibiotics. The explanted sclera buckles were rinsed with thioglycolate broth in sterile tubes and were immediately sent to the Department of Clinical Laboratory within our hospital for micobiologic testing. The tubes were incubated at 37°C for 48 h. Subcultures were performed on Columbia blood agar, Columbia chocolate agar, Sabouraud agar plate and Lowenstein-Jensen media (Oxoid Ltd, UK). The isolates were tested using Gram-stain, acid-fast stain, catalase test and Kligler Iron Agar test (Oxoid Ltd, UK). The isolates were identified at the species level using the API 20E, API 20NE, API 20 STREP and API STAPH system (bioMérieux, France) according to manufacturer's manual.

Susceptibility of the bacteria was tested using the Kirby-Bauer disk diffusion method on Mueller–Hinton (MH) agar or blood MH agar (α-hemolytic streptococcus) plates. The paper discs of the following antibiotics (Oxoid Ltd, UK) were used for the tests: Vancomycin (30 ug/disc), Clindamycin (2 ug/disc), Gentamicin (10 ug/disc), Ciprofloxacin (5ug/disc), Trimeth/sulfa (25 ug/disc), Erythromycin (15 ug/disc), Penicillin (10 units/disc), Azithromycin (15 ug/disc), Ofloxacin (5 ug/disc), Cefazolin (30 ug/disc), Cefepime (30 ug/disc), Tetracycline (30 ug/disc), Rifampin (5 ug/disc), Cefuroxime (30 ug/disc), Amikacin (30 ug/disc), Aztreonam (30 ug/disc), Ceftazidime (30 ug/disc), Cefoxitin (30 ug/disc). The paper discs were gently pressed onto the surface of the agar. After the plates were incubated overnight, the diameter of the inhibition zones surrounding the paper discs was measured. Isolates were characterized as sensitive, intermediate sensitive or resistant based on the diameter of the inhibition zones according to the performance standards of the Clinical and Laboratory Standards Institute (CLSI) document M100. According to CLSI (2010) guidelines (Clinical and Laboratory Standards Institute/NCCLS), methicillin resistant CNS is defined as: resistance to cefoxitin (inhibition zones ≤ 24 mm) using the disk diffusion test.

RESULTS AND DISCUSSION

In this report, 40 microorganisms were isolated from the 38 explanted scleral buckles. The two coexisting species were CNS and α-hemolytic streptococcus in one patient, and CNS and Staphylococcus aureus in anothert patient. Species identification revealed that the 40 micro-organisms belonged to 10 different species, including 4 Gram-positive and 6 Gram-negative organisms (Table 1). Overall, 75% of the isolates were Gram-positive and 25% were Gram-negative. Coagulase-negative *Staphylococcus* (CNS) (62.5%) was the most commonly isolated Gram-positive organism, followed by α-hemolytic *Streptococcus* (7.5%). *Klebsiella pneumonia* (7.5%) and *Pseudomonas aeruginosa* (7.5%) were the most commonly isolated Gram-negative organisms (Table 1). No fungi or mycobacteria were isolated. Our data suggest a high isolation rate of Gram-positive organisms compared to a previous report (75 vs. 54.8%, Pathengay et al., 2004) and CNS was the most commonly isolated. A similar trend has been reported in endophthalmitis (Melo et al., 2011). CNS is one of the most common preoperative conjunctival bacteria (Melo et al., 2011; Kim et al., 2011) and may be the main source of infection at the time of retinal detachment surgery. The isolates presented a low-grade potential for chronic infection by the formation of biofilm (Holland et al., 1991; Asaria et al., 1999).

100% of the Gram-positive isolates were sensitive to Vancomycin, 69% to Clindamycin, 59.3% to Gentamicin,

Table 2. Antibiotic sensitivities of isolated Gram-positive organisms.

Parameter	V (n=30)	CL (n=29)	G (n=27)	CI (n=28)	T (n=27)	E (n=28)	P (n=26)	A (n=27)	O (n=26)	CE (n=26)
Sensitive	30	20	16	11	11	11	3	7	11	11
Resistant	0	7	9	14	15	17	23	20	12	14
Intermediate	0	2	2	3	1	0	0	0	3	1
%Sensitivity	100%	69%	59.3%	39.3%	40.7%	39.3%	11.5%	25.9%	42.3%	42.3%

V=Vancomycin; CL=Clindamycin; G=Gentamicin; CI=Ciprofloxacin; T=Trimeth/sulfa; E=Erythromycin; P=Penicillin; A=Azithromycin; O=Ofloxacin; CE=Cefazolin.

Table 3. Antibiotic Sensitivities of Methicillin-Resistant Coagulase-negative *Staphylococcus*.

Parameter	V (n=10)	CL (n=10)	G (n=10)	CI (n=10)	T (n=9)	E (n=8)	A (n=10)	O (n=10)	CF (n=6)	CE (n=9)	CEF (n=10)	R (n=9)
Sensitive	10	5	5	3	0	1	1	2	0	3	4	9
Resistant	0	5	5	7	8	7	9	6	6	6	5	0
Intermediate	0	0	0	0	1	0	0	2	0	0	1	0
%Sensitivity	100%	50%	50%	30%	0%	12.5%	10%	20%	0%	33.3%	40%	100%

V=Vancomycin; CL=Clindamycin; G=Gentamicin; CI=Ciprofloxacin; T=Trimeth/sulfa; E=Erythromycin; A=Azithromycin; O=Ofloxacin; CF=Cefepime; CE=Cefazolin; CEF=Cefuroxime; R=Rifampin.

Table 4. Antibiotic Sensitivities of isolated Gram-negative organisms.

Parameter	G (n=10)	CI (n=10)	A (n=10)	AZ (n=10)	O (n=8)	CE (n=10)	CEF (n=10)
Sensitive	5	8	7	4	6	7	7
Resistant	4	2	3	6	1	3	1
Intermediate	1	0	0	1	1	0	2
%Sensitivity	50%	80%	70%	40%	75%	70%	70%

G=Gentamicin; CI=Ciprofloxacin; A=Amikacin; AZ=Aztreonam; O=Ofloxacin; CE=Ceftazidime; CEF=Cefepime.

39.3% to Ciprofloxacin and 42.3% to Ofloxacin (Table 2). Compared to a previous report (Pathengay et al., 2004), the sensitivity of the Gram-positive cocci to Vancomycin (100 vs. 93.1%) and Gentamicin (59.3 vs. 56.7%) was similar however the sensitivity to Ciprofloxacin (39.3 vs. 73.3%) and Cefazolin (42.3 vs. 86.7%) was decreased greatly. Resistance to Ofloxacin in Gram-positive isolates was also increased (Ramesh et al., 2010). Ofloxacin, Ciprofloxacin and Cefazolin are commonly used for the treatment of perioperative infection in patients undergoing retinal detachment surgery in China. This increasing trend of Gram-positive isolates resistant to these antibiotics is of additional concern. Additionally, 10/25 (40%) of CNS resistant to cefoxitin (methicillin-resistant CNS) have a low susceptibility rate to the most commonly used antibiotics, but were sensitive to Vancomycin and Rifampin (Table 3). 80% of the Gram- negative isolates were sensitive to Ciprofloxacin and 75% to Ofloxacin (Table 4), suggesting that Ciprofloxacin and Ofloxacin remain good alternatives to Gram-negative isolates.

In summary, this study suggests an increasing trend of Gram-positive isolates in explanted sceral buckle. It also describes an increasing level of Gram-positive isolates resistant to the commonly used antibiotics. No single antibiotic can cover all of the microbes isolated from explanted sceral buckles. Combination therapy is recommended for the treatment of perioperative infection and initial empiric treatment.

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