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Detection and Quantitation by Lysis-Filtration of Bacteremia after Different Oral Surgical Procedures

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Patients with bacteremia after dental extraction, third-molar surgery, dental scaling, endodontic treatment, and bilateral tonsillectomy were studied by means of lysis-filtration of blood samples with subsequent aerobic and anaerobic incubation. Samples were obtained before, during, and 10 min after treatment. Bacteremia was observed in 100% of patients after dental extraction, 55% of patients after third-molar surgery, 70% of patients after dental scaling, 20% of patients after endodontic treatment, and 55% of patients after bilateral tonsillectomy. Anaerobic microorganisms were isolated more frequently than aerobic microorganisms were, and viridans group streptococci were the most commonly isolated bacteria. Ten minutes after treatment, the frequency as well as the magnitude of bacteremia showed pronounced reduction.

Bacteremia is induced by a wide variety of clinical procedures and manipulations, particularly those involving heavily colonized mucous membranes or infected sites. It has been known for a long time that invasive procedures of oral tissues result in the translocation and release of microorganisms from the oral cavity into the bloodstream. Generally, the microorganisms are eliminated by the reticuloendothelial system within a few minutes. However, bacteremia is a potential hazard to patients with abnormal heart valves or other cardiac abnormalities predisposing them for infective endocarditis. Consequently, numerous studies have been performed to estimate the type, frequency, and magnitude of postsurgical bacteremia (for a review, see reference 1). The incidence of dental bacteremia reported in different studies has ranged from 17 (10) to 93.4% (2) after multiple extractions, and the incidence of bacteremia after tonsillectomy has ranged from 28 (4) to 38% (11). The interpretation of the results from available studies concerning different oral procedures is complicated since patient selection procedures, inclusion criteria, and especially, the microbiological techniques used are not comparable. In this study we used a lysis-filtration technique to investigate the frequency, type, and magnitude of bacteremia in patients after five different standard oral surgical procedures.

MATERIALS AND METHODS

Patients. A total of 100 patients (56 males, 44 females; ages, 15 to 75 years; mean age, 38 years) who were referred to Huddinge University Hospital, Karolinska Institute, for dental treatment or tonsillectomy participated in the study. None of the patients were receiving any medication, and none of the patients had been treated with any antimicrobial agent for at least 6 weeks. The study was approved by the Ethics Committee of Huddinge University Hospital.

Dental extraction. A single tooth was extracted because of dental caries or chronic periradicular osteitis in 20 patients (14 males, 6 females; ages, 28 to 75 years; mean age, 52 years). Analgesia was given by local injection of prilocaine (30 g/liter) containing felypressin (0.00054 g/liter) as a vasoconstrictor. Blood samples were taken before injection of local anesthetic. Another sample was obtained during dental extraction, and a third sample was obtained 10 min after the extraction was terminated.

Surgical removal of a third lower molar. One impacted (no communication with the oral cavity) or partially impacted (communication with the oral cavity) lower third molar was surgically removed in 20 patients (9 males, 11 females; ages, 15 to 43 years; mean age, 26 years). All operations were performed by using a standardized operation technique. Analgesia was given by local injection of prilocaine (30 g/liter) containing felypressin (0.00054 g/liter) as a vasoconstrictor. One blood sample was obtained before injection of local anesthetic. Another sample was taken during surgery, and a third sample was taken 10 min after the operation was terminated.

Dental scaling. Subgingival calculus was removed from the lingual part of the lower front teeth by means of periodontal hand instruments (a procedure that constantly caused bleeding from the gingival crevice) in 20 patients (12 males, 8 females; ages, 22 to 65 years; mean age, 50 years) with periodontitis. One blood sample was taken before the initiation of dental scaling. Another sample was obtained during dental scaling, and a third sample was obtained 10 min after the scaling was terminated.

Endodontic treatment. After application of the rubber dam, the necrotic pulp of a single tooth was removed by a conventional endodontic technique in 20 patients (14 males, 6 females; ages, 23 to 63 years; mean age, 36 years). Care was taken not to force any instruments beyond the apical foramen. One blood sample was obtained before initiation of treatment. Another sample was taken during endodontic treatment, and a third sample was taken 10 min after the treatment was terminated and the rubber dam was removed.

Tonsillectomy. Bilateral tonsillectomy was performed under general anesthesia by dissection and snaring in 20 patients (7 males, 13 females; ages, 15 to 41 years; mean age, 26 years) with chronic tonsillitis. One blood sample was taken before initiation of anesthesia. Another sample was obtained during tonsillectomy, and a third sample was ob-

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tained 10 min after the operation was terminated and extubation of the endotracheal tube had been performed.

Blood sampling procedure. Prior to sampling, the skin of the cubital fossa was carefully washed with chlorhexidine in 70% ethyl alcohol followed by 8.5% povidone iodine solution. An indwelling catheter was placed in the cubital vein of the arm under strict aseptic conditions. Blood samples (8.3 ml) were drawn by means of the indwelling catheter into VACUTAINER tubes containing sodium polyanaetholsulfonate (1.7 ml of a 0.35% solution; Becton Dickinson & Co., Rutherford, N.J.). The samples were immediately processed in the microbiological laboratory.

Blood culture technique. The blood samples were injected into bottles with 0.193 liter of a lysing solution (pH 10) containing 0.08% Na_2CO_3 and 0.005% Triton X-100 (Rohm and Haas, Darmstadt, Federal Republic of Germany) plus 0.003 liter of a commercial streptokinase-streptodornase compound (Varidase; Lederle Laboratories, Madrid, Spain) to avoid clogging of the filters. Vacuum filtration was performed in a 0.45-μm-pore-size filter system (Millipore AB, Solna, Sweden) under a continuous flow of nitrogen. The lysis-filtration system has been evaluated for aerobic microorganisms by Sullivan et al. (14) and for anaerobic bacteria by Heimdahl et al. (6). After filtration, the filters were divided with scissors and placed onto two brainheart infusion agar (Difco Laboratories, Detroit, Mich.) plates supplemented with 5% horse blood for aerobic and anaerobic incubation, respectively, at 37°C for 10 days.

Identification of microorganisms. Aerobic and anaerobic bacteria were identified by the methods described in the *Manual of Clinical Microbiology* (9). Quantitative counts were estimated from the numbers of colonies visible on the filters.

Statistics. Differences in incidences were analyzed by the Fisher exact test.

RESULTS

Incidence and magnitude of bacteremia. No microorganisms were found in any of the 100 pretreatment blood samples.

In total, 201 different isolates were obtained from the samples collected during and 10 min after the different surgical procedures. *Streptococcus intermedius* and *Actinomyces* spp. were the most common isolates (Table 1). Anaerobic strains were more frequently isolated than aerobic strains were (117 versus 84 isolates, respectively). Many strains of viridans group streptococci were isolated only after anaerobic incubation, and no bacterial strain was isolated after aerobic incubation that was not also found after anaerobic incubation.

The incidence of bacteremia (Table 2) ranged from 100% (dental extraction) to 20% (endodontic treatment). Extraction produced a significantly higher frequency of bacteremia than the other procedures did ($P < 0.05$). The incidences of bacteremia in samples taken during the different surgical procedures (Table 3) were higher (range, 95 to 15%) than they were in samples taken 10 min afterward (range, 40 to 0%).

The magnitude of bacteremia ranged from 2.19 CFU/ml during third-molar surgery to 0.2 CFU/ml during endodontic treatment (Table 3). There was no relationship between the type of microorganism isolated and its colony count.

Viridans group streptococci were isolated from blood samples from all five groups of patients (Tables 1 to 3) and were isolated most frequently after dental extraction (85%) and dental scaling (55%).

Ten minutes after the different procedures, viridans group streptococci were isolated from only two patients in the dental extraction group, two patients in the third-molar surgery group, and one patient in the endodontic treatment group (Table 3). The magnitude of bacteremia with viridans group streptococci in positive blood samples ranged from 1.0 CFU/ml (during dental extraction) to 0.12 CFU/ml (10 min after third-molar surgery).

Anaerobic bacteremia (Table 3) was observed in blood samples obtained during dental extraction (70%), third-molar surgery (45%), dental scaling (60%), and bilateral tonsillectomy (40%).

Anaerobes were still isolated from blood samples after 10 min in the four groups of patients (range, 5 to 35% of the patients), and the magnitude of anaerobic bacteremia in positive blood samples ranged from 1.7 (during third-molar surgery) to 0.14 (10 min after dental scaling) CFU/ml.

DISCUSSION

The results of studies of postoperative transient bacteremia vary from one study to another, depending on the type of surgical treatment and the method used for isolation of microorganisms from blood. Fastidious microorganisms requiring special conditions or nutrients may not survive or grow in many common blood culture systems. Because of the rapid growth of some microorganisms, they may inhibit or outnumber other organisms. Phagocytic cells and antimicrobial substances in blood may prevent the growth of microorganisms in blood cultures. Antimicrobial agents in blood from patients on antimicrobial treatment may inhibit the growth of susceptible microorganisms. Different methods have been used to overcome these problems. Increased numbers of microorganisms isolated from patients with transient postoperative bacteremia have been reported after introduction of prereduced anaerobic media (3), increased volume of blood samples (D. C. Shanson, Abstr. 4th Int. Symp. Rapid Methods Automation Microbiol. Immunol., abstr. no. P11, 1984), addition of a papain digest of ox liver to brain heart infusion cysteine broth (12), or application of a blood culture system based on lysis-filtration (6).

In the present study of bacteremia in patients after different standardized oral surgical procedures, the lysis-filtration technique was used with subsequent aerobic and anaerobic incubation. The observed incidences as well as magnitudes of bacteremia were higher than those described previously, confirming the high sensitivity of the combination of lysis-filtration and brain heart infusion agar (6). Asymptomatic bacteremia (7, 16) and a high frequency of contamination have been reported in connection with blood culture systems based on lysis of blood (5, 6), and therefore, preoperative blood samples must also be obtained in order to estimate the true postoperative bacteremia. In the present study, we decreased the frequency of contamination from 7%, as reported previously (6), to 0% in 300 blood samples by changing the laboratory procedures from daily microscopic examination to one single examination after 10 days. This indicates that although meticulous care is taken to avoid contamination during sampling, laboratory contamination is still frequent unless special care is taken. Using these principles, we were not able to demonstrate bacteremia in any patient before the surgical manipulations were initiated.

During the oral surgical procedures used in this study, we observed that bacteremia differed considerably in quality and quantity between different treatment groups. Bacteremia was not related to the extent of surgery, since a single

TABLE 1. Microorganisms obtained from blood samples collected during and 10 min after different oral surgical procedures and the numbers of patients from whom each species was isolated

Microorganism isolated	No. of patients from whom species were isolated after:				
	Dental extraction	Third-molar surgery	Dental scaling	Endodontic treatment	Bilateral tonsillectomy
Aerobes					
Staphylococcus epidermidis		2			
Micrococcus spp.				1	
Streptococcus spp.				1	
Enterococcus faecalis		1			
Corynebacterium hofmanii		2		1	
Corynebacterium murium		1			
Haemophilus spp.			2		
Neisseria spp.				1	
Viridans group streptococci					
Streptococcus mitior	7	2	6	2	3
Streptococcus sanguis	4	1	4		
Streptococcus mutans	4	1			1
Streptococcus salivarius				1	1
Streptococcus intermedius	15	6	7	1	6
Anaerobes					
Streptococcus spp.	2		6	1	6
Peptostreptococcus micros		2			
Peptostreptococcus asaccharolyticus		2			
Veillonella parvula	5	1	1		2
Clostridium indolis	1				
Clostridium malenominatum			1		
Clostridium ramosum	1				
Actinomyces israelii	2	1	4		
Actinomyces meyeri			3		
Actinomyces naeslundii	5	2	4		
Actinomyces odontolyticus	7		1		1
Actinomyces viscosus	1		2		
Actinomyces spp.	1		2		
Bifidobacterium adolescentis		2			
Bifidobacterium spp.		2			
Eubacterium spp.	2	5			2
Propionibacterium acnes			1		1
Lactobacillus acidophilus	1	1			
Lactobacillus leichmanii	1				
Lactobacillus plantarum	1		1		
Bacteroides gingivalis					1
Bacteroides ureolyticus	1	1			
Bacteroides spp.	2	3	2	1	1
Fusobacterium nucleatum			1		1
Other gram-positive cocci	2		6	1	6
Other gram-negative cocci					1
Total no. of aerobes/total no. of anaerobes	30/35	16/22	19/35	8/3	11/22

dental extraction produced a higher incidence of bacteremia than third-molar surgery and bilateral tonsillectomy did. It appeared that dental extraction was associated with both aerobic and anaerobic bacteremia significantly more often than the other procedures were. The explanation for this is obscure, but heavy colonization of the tooth surfaces with aerobic and anaerobic microorganisms in combination with the pumping movements used in dental extraction may be of importance.

Viridans group streptococci, which are considered to be

TABLE 2. Incidence and magnitude of bacteremia during and 10 min after different oral surgical procedures

Surgical procedure	% of patients with bacteremia	Mean CFU (range) of bacteremia/ml[a]	% Viridans group streptococci	Mean CFU (range) of viridans group streptococci/ml[a]	% Anaerobes	Mean CFU (range) of anaerobes/ml[a]
Dental extraction	100	1.12 (0.12–6.26)	85	0.93 (0.12–3.73)	75	0.60 (0.12–2.53)
Third-molar surgery	55	1.34 (0.12–9.88)	40	0.77 (0.12–4.21)	45	1.25 (0.12–5.66)
Dental scaling	70	0.66 (0.12–3.25)	55	0.36 (0.12–4.21)	65	0.48 (0.12–2.77)
Endodontic treatment	20	0.54 (0.12–1.57)	15	0.40 (0.12–0.96)	5	0.48 (0.48)
Bilateral tonsillectomy	55	1.40 (0.12–10.5)	40	0.93 (0.12–5.3)	40	0.99 (0.12–5.18)

[a] Mean CFU per milliliter in positive samples.

TABLE 3. Incidence and magnitude of bacteremia during and after different oral surgical procedures in relation to sampling time and numbers of viridans group streptococci and anaerobic bacteria

Sample	Dental extraction	Third-molar surgery	Dental scaling	Endodontic treatment	Bilateral tonsillectomy
Sample obtained before surgery					
No. of bacteremic patients	0	0	0	0	0
Mean CFU/ml	0	0	0	0	0
Sample obtained during surgery					
No. of bacteremic patients	19/20[a]	11/20	13/20	3/20	11/20
Mean CFU/ml	1.45	2.19	0.90	0.20	1.40
Sample obtained 10 min after surgery					
No. of bacteremic patients	8/20	8/20	6/20	1/20	0/20
Mean CFU/ml	0.35	0.17	0.14	1.57	0
Sample obtained during surgery					
No. of bacteremic patients with viridans group streptococci	17/20	8/20	11/20	2/20	8/20
Mean CFU/ml	1.0	0.93	0.36	0.12	0.93
Sample obtained 10 min after surgery					
No. of bacteremic patients with viridans group streptococci	2/20	2/20	0/20	1/20	0/20
Mean CFU/ml	0.3	0.12	0	0.96	0
Sample obtained during surgery					
No. of bacteremic patients with anaerobic bacteria	14/20	9/20	12/20	0/20	8/20
Mean CFU/ml	0.74	1.7	0.64	0	0.99
Sample obtained 10 min after surgery					
No. of bacteremic patients with anaerobic bacteria	7/20	4/20	6/20	1/20	0/20
Mean CFU/ml	0.30	0.18	0.14	0.48	0

[a] Values are number of bacteremic patients/total number of patients in that group tested.

potential infective endocarditis pathogens, were isolated from all five groups of patients, indicating the need for antimicrobial prophylaxis in connection with these types of oral procedures in patients predisposed to infective endocarditis. Other potential endocarditis pathogens, such as staphylococci and enterococci, were observed only as single isolates. *Candida albicans*, which is endogenous in the oral activity, and *Actinobacillus actinomycetemcomitans*, which has been described as being associated with progressive periodontitis (13) and rare cases of endocarditis (8), were not isolated from any of the 300 blood samples obtained in the present investigation. This was not due to the type of isolation technique or medium used, because simulated in vitro studies showed excellent recovery of these microorganisms.

Aerobic incubation did not increase the number of different isolates compared with incubation under anaerobic conditions and can probably be omitted in this type of study. Furthermore, all aerobic microorganisms were also isolated from filters after anaerobic incubation.

Elimination of microorganisms from blood was rapid, and after 10 min, the viridans group streptococci were eliminated from 42 of 46 patients, which is consistent with the results from similar studies (15). The anaerobic microorganisms were more often observed after 10 min, probably because of greater magnitude in blood observed during surgery.

From the results of the present study, it appears that after different oral surgical procedures, aerobic and anaerobic bacteremias are more frequent and appear in higher magnitudes than have been reported previously. The lysis-filtration technique in combination with a standard oral surgical procedure, such as the extraction of a single tooth, which produces a high frequency of bacteremia, should provide a suitable model for studies of different measures for the prevention of bacteremia. Such studies are now in progress.

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