Mediastinitis after Cardiac Surgery: Improvement of Bacteriological Diagnosis by Use of Multiple Tissue Samples and Strain Typing

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The diagnosis of postsurgical mediastinitis (PSM) among patients with sternal wound complication (SWC) after cardiac surgery is sometimes difficult, as fever, elevated C-reactive protein levels, and chest pain can be caused by a general inflammatory reaction to the operative trauma and/or sternal dehiscence without infection. The definitions of PSM usually used emphasize clinical signs and symptoms easily observed by the surgeon. The aim of the study was to investigate whether the use of standardized multiple tissue sampling, optimal culturing methods, and strain typing, together with a microbiological criterion for infection, could identify more infected patients than clinical assessment alone. Patients reexplored due to SWC after cardiac artery bypass grafting (CABG) or heart valve replacement (HVR) with or without CABG performed at the Department for Cardio-Thoracic Surgery at the Uppsala University Hospital between 10 March 1998 and 9 September 2000 were investigated prospectively. Tissue samples were taken from the sternum or adjacent mediastinal tissue, preferably before the administration of antibiotics. Culturing was performed both directly (on agar plates) and using enrichment broth. Species identification was performed by standard methods, and strain typing was performed by pulsed-field gel electrophoresis. A total of 41 cases with at least five tissue samples each were included in the study group. Of these patients, 32 were infected according to the microbiological criterion (i.e., the same strain was found in ≥50% of the samples). *Staphylococcus epidermidis* was the primary pathogen in 38% of the cases (12/32), *S. aureus* was the primary pathogen in 31% (10/32), *P. acnes* was the primary pathogen in 25% (8/32), and *S. simulans* and *S. haemolyticus* were the primary pathogens in 3% (1/32) each. All cases of *S. aureus* infection and 86% (12/14) of coagulase-negative staphylococcus (CoNS) infections were identified from primary cultures. All cases fulfilling the microbiological criterion for *S. aureus* infection were clinically diagnosed as cases of infection, but among the 14 cases fulfilling the criterion for microbiological diagnosis of CoNS infection, only 10 appeared to qualify clinically as cases of infection. Among the patients with sternal dehiscence in whom a microbiological diagnosis was established, 67% (12/18) had a CoNS infection, compared to 14% (2/14) of those without sternal dehiscence. The difference was statistically significant. PSM caused by *S. aureus* is readily identified by the surgeon, whereas 30% of cases with CoNS infections may be misinterpreted as noninfected. Multiple sampling before administration of antibiotics, primary culturing on agar plates, species identification, strain typing, and susceptibility testing should be used to ensure a fast and microbiologically correct diagnosis which identifies the primary pathogen and infected patients among those with minor infective symptoms. The role of *P. acnes* as a possible cause of PSM needs further investigation. PSM caused by CoNS is significantly related to sternal dehiscence.

Postoperative deep sternal wound infection occurs in 0.6 to 16% of patients who have undergone cardiac surgical procedures performed using median sternotomy (13, 19). The variance in reported incidence levels may depend on differences in definitions of infection.

The diagnosis of postsurgical mediastinitis (PSM) among patients with sternal wound complication after cardiac surgery is sometimes difficult, since fever, elevated C-reactive protein levels, and chest pain can be caused by a general inflammatory reaction to the operative trauma and/or sternal dehiscence without infection (1).

According to the definition suggested by The Hospital Infection Control Practices Advisory Committee, Centers for Disease Control and Prevention (CDC), Atlanta, Georgia, and the adjustments made to that definition by R. M. El Oakley et al. for cases involving cardiothoracic surgery, PSM is a deep infection involving retrosternal tissue and/or the sternal bone (3, 5). The list of criteria for organ or space surgical-site infection (SSI) used by the National Nosocomial Infection Surveillance system of the CDC (5) includes signs and symptoms that can be directly observed by the surgeon. The National Nosocomial Infection Surveillance system also offers the possibility of using a microbiological criterion for detection of infection without visible signs of infection ("Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ or space"). The criterion does not specify the requirements for culturing, species identification, strain typing, and interpretation of bacteriological findings.

The criteria used by the individual researcher for defining PSM are often modifications of the suggestions from The Hospital Infection Control Practices Advisory Committee and usually emphasize the signs and symptoms easily observed by the surgeon. Those often include purulent discharge from the mediastinal area (11, 12, 16), in some cases together with fever (≥38°C) and sternal dehiscence (4, 10, 18). L'Ecuyer et al.,

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who did not include purulent discharge or sternal dehiscence in their criteria for chest infection, found that those signs occurred in only 67% and 26% of the patients, respectively (8).

In many studies on PSM, there is a requirement for a positive bacteriological culture in addition to observable signs and symptoms (4, 9, 11, 12, 18, 20). This requirement might serve to limit diagnostic accuracy, as even in cases with undoubted and visible signs of infection, bacteriological cultures can return negative results due to antibiotics given to the patient pre- or intraoperatively.

Coagulase-negative staphylococci (CoNS) are recognized as important pathogens in PSM but are also known not to produce the same fulminant clinical signs and symptoms of infection as, for instance, *S. aureus*. The finding of staphylococci in a sample from mediastinal tissue or fluid is difficult to evaluate, as CoNS, and sometimes also *S. aureus*, are inhabitants of human skin and thus can easily contaminate a sample.

A majority of the CoNS causing PSM have been reported as methicillin resistant (MR) during the last decade (9, 13). As this resistance limits the therapeutic options, it is important not to overestimate the proportion of MR strains or the number of infected patients.

To be able to choose an adequate long-term treatment and avoid unnecessary use of broad-spectrum antibiotics, a definition of PSM should be established which is based on both clinical and bacteriological criteria. This is especially important concerning infections caused by CoNS, as those infections seem to increase, thus creating great diagnostic difficulties.

This study was undertaken to investigate whether it was possible to use a microbiological criterion for PSM to identify infected patients among those with sparse clinical signs and symptoms while avoiding inclusion of those patients for whom the microbiological findings represent contamination. A similar approach has been used in orthopedic surgery to differentiate between infectious and noninfectious loosening after total hip arthroplasty (6).

Aim. The aim of the study was to investigate whether the use of standardized multiple tissue sampling, optimal culturing methods, and strain typing, together with a microbiological criterion for infection, could identify a greater number of infected patients than clinical assessment alone.

MATERIALS AND METHODS

Design. The study was a prospective cohort study of reexplorative surgical procedures after primary elective operations for cardiac artery bypass grafting or heart valve replacement with or without cardiac artery bypass grafting, performed at the Department for Cardio-Thoracic Surgery at the Uppsala University Hospital from 10 March 1998 to 9 September 2000. According to the study protocol, five tissue samples for bacteriological culturing were to be taken from the sternum or adjacent mediastinal tissue of each patient. Sampling was made irrespective of clinical assessment of the reason for reexploration. The clinical diagnosis was not known to the bacteriologist who processed the samples.

Microbiological diagnosis of infection. The microbiological diagnostic criterion for infection was, in cases in which the infecting bacterial species was CoNS or *S. aureus*, the finding of the same strain in at least half of the samples, and in cases in which the infecting bacterial species was other than CoNS or *S. aureus*, the finding of the same species in at least half of the samples. Bacterial isolates whose pulsed-field gel electrophoresis (PFGE) patterns were identical were defined as belonging to the same strain (17).

Clinical diagnosis of infection. Six reasons for reexploration were identified: bleeding, arrhythmia or restenosis, reconstructive surgery, sternal dehiscence without signs or symptoms of SSI, sternal dehiscence with signs or symptoms of SSI, and SSI without signs or symptoms of sternal dehiscence.

The clinical diagnosis of infection or noninfection was made by the surgeon performing the reexploration. The diagnosis was based on preoperative signs and symptoms, such as fever, chest pain, purulent or serous secretion from the wound, sternal dehiscence, and bleeding, and intraoperative findings, such as pus, necrotized tissue, bleeding vessels, and a gap between the sternal halves. The assessment of infection or noninfection was noted in the patient's record. After termination of the study period, the patients' records (with the clinical diagnoses) were read and compared with the microbiological results.

Tissue sampling. Pieces of tissue were taken from the sternum or adjacent mediastinal tissue. Each specimen was put into a sterile glass tube which contained Transport medium for anaerobes (in-house modification of Cary and Blair pre-reduced anaerobic sterilized Transport medium) and had been gassed out with carbon dioxide.

Samples were taken before administration of peroperative antibiotics if possible.

Bacteriological methods. All samples were processed by either of two microbiological technical assistants and interpreted by the investigator. Culturing of specimens took place in a laminar airflow bench. Each specimen was handled with a sterile forceps and cultured on plates containing blood agar (Columbia blood agar base [Acumedia], supplemented with horse blood), chocolate agar (Columbia blood agar base [Acumedia], supplemented with heated horse blood) and anaerobic agar (Fastidious anaerobe agar [Lab M], supplemented with horse blood). The specimen was then put into a sterile tube containing enrichment broth for anaerobic bacteria (brain heart infusion broth [BBL], supplemented with yeast extract [Acumedia], hemin, vitamin K1, cysteine hydrochloride, and resazurin). Subculturing from broth to agar plates (same plates as described above) was performed whenever growth was visible and on days 4 and 14, irrespective of visible growth.

Plates were incubated according to standard recommendations and inspected after 24 h, 48 h, and 7 days. Bacteria belonging to the genera *Staphylococcus*, *Enterobacteriaceae, Streptococcus*, and *Propionibacterium* were identified according to standard methods. Staphylococci were tested for DNase activity (DNase agar; Acumedia). DNase-negative isolates not fermenting trehalose or mannitol, resistant to bacitracin, and sensitive to novobiocin were identified as *S. epidermidis*. DNase-negative isolates not fulfilling the criteria for *S. epidermidis* were identified to the species level using Staph-API (Biomérieux SA, Marcy l'Etoile, France). DNase-positive isolates were identified as *S. aureus*. All staphylococcal isolates were subjected to PFGE.

Chromosomal analysis of staphylococci by PFGE. Isolates were grown overnight in 3 ml of Todd Hewitt broth at 35°C. Absorbance was read at wavelength 420 nm (Novaspec 11; Pharmacia, Uppsala, Sweden) and adjusted to an optical density of 1.0. The bacteria were harvested by centrifugation for 15 min at 3,500 rpm. They were resuspended in 3 ml of Tris-HCl buffer, pH 7.6. A total of 150 μl of the suspension was mixed with 150 μl of 2% InCert agarose (FMC, Rockland, Maine) in Tris-HCl buffer at 55°C. The mixture was poured into the slots of a plastic mold (Bio-Rad, Richmond, Calif.) and cooled for 1 h at 4°C. The plugs were incubated at 35°C overnight in 4 ml of lysis buffer 1 (6 mM Tris-HCl [pH 7.6], 1 M NaCl, 100 mM EDTA [pH 7.5], 0.5% Brij 58, 0.2% deoxycholate, 0.5% sodium lauryl sarcosine [Sarcosyl], 20 μg of RNase [Boehringer Mannheim, Mannheim, Germany]/ml, 1 mg of lysozyme [Boehringer Mannheim]/ml, 1 mg of lysostaphin [Sigma, St. Louis, Mo.]/ml). The lysis buffer 1 medium was replaced by 4 ml of lysis buffer 2 (1% sodium lauryl sarcosine [Sarcosyl], 0.5 M EDTA [pH 9.5], 50 μg of proteinase K [Boehringer Mannheim]/ml), and the plugs were incubated overnight at 55°C. The plugs were washed three times in 4 ml of Tris-EDTA buffer at 35°C. Digestion by restriction enzyme was performed overnight at 25°C by placing a 2-mm-thick slice of each plug in 225 μl of restriction buffer containing 20 IU of *Sma*I (Boehringer Mannheim). The plugs were placed into the slots of a 1% agarose (Ultra Pure Agarose; GIBCO BRL, Paisley, United Kingdom) gel in 0.5× Tris-EDTA buffer and sealed with 1% agarose. Electrophoresis was performed in a contour-clamped homogenous electric field apparatus (Gene Path; Bio-Rad) at 6.0 V/cm for 20 h at 14°C, with initial pulse times of 5.3 s and final pulse times of 34.9 s.

Test for methicillin susceptibility. The testing was performed by disk diffusion according to the recommendations by the Swedish Reference Group for Antibiotics (www.srga.org). A dense inoculum with abundant growth was prepared on an Iso-Sense agar plate (Oxoid Ltd., Basingstoke, Hampshire, England), supplemented with horse blood. A disk containing 1 μg of oxacillin was placed on the agar surface. The plate was incubated at 30°C for 20 to 24 h. A zone of ≥12 mm in diameter was interpreted as indicating susceptibility, and a zone of ≤9 mm in diameter was interpreted as indicating resistance.

Statistical methods. Risk ratios (RR) were calculated by chi-square tests and Fisher's exact test, using the program EPI-INFO (CDC and the World Health

TABLE 1. Number of species isolates found in 166 culture-positive
tissue samples

Species	No. of isolates
S. epidermidis	89
S. simulans	5
S. haemolyticus	5
S. lugdunensis	1
CoNS spp.	3
CoNS spp. total	103
P. acnes	55
S. aureus	46
α-Hemolytic streptococci	5
Citrobacter spp.	2
C. parapsilosis	2
Total	213

TABLE 2. Number of samples per operation and primary pathogen
in the 41 cases with ≥5 tissue samples

No. of samples per case	No. of cases	Number of cases with:			
		Same species or strain:		All samples negative	Same species in less than half of the samples
		In all samples[a]	In >50% of samples[b]		
7	1		1		
6	3	2		1	
5	37	18	11	4	4
Total	41	20	12	5	4

[a] Of the cases from which six samples each were used, the two species detected in all samples were *S. epidermidis* (one case) and *S. aureus* (one case); of the cases from which five samples each were used, the five species detected in all samples were *S. epidermidis* (six cases), *S. aureus* (six cases), *P. acnes* (four cases), *S. simulans* (one case), and *S. haemolyticus* (one case).

[b] Of the cases from which seven samples each were used, the species detected in more than 50% of samples was *P. acnes* (one case); of the cases from which five samples each were used, the three species detected in more than 50% of samples were *S. epidermidis* (five cases), *S. aureus* (three cases), and *P. acnes* (three cases).

Organization). An RR of >1.0 with a 95% confidence interval (CI) not including 1.0 was regarded as significant, as was a *P* value of <0.01.

RESULTS

Study group. A total of 112 reexplorative surgical procedures were performed with 74 patients during the study period.

When investigating the microbiological findings and comparing microbiological and clinical findings, each operation was regarded a new case. Sampling according to protocol (≥five tissue samples) was performed in 41 operations.

Samples. A total of 210 tissue samples were obtained from the 41 study group operations. Of these samples, 166 were culture positive and 44 were culture negative.

Bacterial isolates. In the 166 positive samples, 213 bacterial isolates were found. Among these were 103 CoNS isolates, 55 *P. acnes* isolates, and 46 *S. aureus* isolates. The numbers of isolates of different species are presented in Table 1.

MR among staphylococcal isolates. Among the 89 *S. epidermidis* isolates, susceptibility to methicillin was tested in 88. Of the tested isolates, 56.8% (*n* = 50) were MR and 43.2% (*n* = 38) were methicillin susceptible (MS). All *S. simulans* (5) and *S. haemolyticus* (5) isolates were MR, and the single isolate of *S. lugdunensis* was MS. Among the CoNS species, one was MS and the other two were not tested. In total, 60% (60/100) of the tested CoNS isolates were MR and 40% (40/100) were MS.

All *S. aureus* isolates were susceptible to methicillin.

PFGE. All 89 *S. epidermidis* isolates were analyzed by PFGE. A total of 24 different patterns (i.e., strains) were recognized. Of the strains, 50% (12/24) were MR and 50% (12/24) were MS.

Of 46 *S. aureus* isolates, 40 were analyzed by PFGE. Eight different patterns were found.

Microbiological diagnosis of infection. Using the microbiological diagnostic criteria, 32 of the 41 cases were identified as infected. *S. epidermidis* was the primary pathogen in 38% (12/32), *S. aureus* was the primary pathogen in 31% (10/32), *P. acnes* was the primary pathogen in 25% (8/32), and *S. simulans* and *S. haemolyticus* were the primary pathogens in 3% (1/32) each.

Nine cases were categorized as noninfected. Among those, all samples were culture negative for five patients. For the

remaining four patients, the same species or strain was found in less than half of the samples.

The findings in the individual cases are summarized in Table 2.

Duration from culturing to positive culture. All 10 cases of *S. aureus* infection were identified from primary cultures on day 2 or 3, as were 11 out of 12 cases of *S. epidermidis* infections and the *S. simulans* infection. Only one case of *S. epidermidis* infection and the *S. haemolyticus* infection were identified in subcultures from the day 4 enrichment broth. All eight cases of *P. acnes* infection were identified in subcultures from the day 4 enrichment broth.

For the four cases with same species or strain in less than half of the samples, there was no evidence of growth in the primary cultures.

Clinical diagnosis of infection. Among the 41 cases included in the study group, 6 were reexplored due to bleeding, 1 was reexplored due to arrhythmia or restenosis, 8 were reexplored due to sternal dehiscence without signs or symptoms of SSI, 16 were reexplored due to sternal dehiscence with signs or symptoms of SSI, and 10 were reexplored due to SSI without signs or symptoms of sternal dehiscence. According to our definition, 15 were clinically noninfected and 26 were infected.

Comparison of microbiological and clinical diagnosis of infection. Among the 32 cases fulfilling the criterion for microbiological infection, 21 were clinically assessed as infected and 11 were clinically assessed as noninfected. The corresponding figures for the 9 cases not fulfilling the criterion for microbiological infection were 5 clinically infected and 4 noninfected. The figures are presented in Table 3.

The overall probability for agreement between the results which were positive by clinical assessment and those fulfilling the microbiological criterion for diagnosis of infection was not significant (RR, 1.2; CI [95%], 0.63 to 2.23; *P* = 0.7).

All 10 cases fulfilling the microbiological criterion for *S. aureus* infection were clinically assessed as infected. Among the 14 cases fulfilling the microbiological criterion for CoNS

TABLE 3. Comparison of clinical and microbiological diagnoses of infection

Assessment by microbiological criterion	Clinical assessment (no. of cases)		
	Infected	Noninfected	Total
Infected	21	11	32
Noninfected	5	4	9
Total	26	15	41

infection, 10 were clinically assessed as infected and 4 were clinically assessed as uninfected.

The overall probability for agreement between the results which were positive by clinical assessment and those fulfilling microbiological criterion for infection was significant when the pathogen was *S. aureus* (RR, 2.0; CI [95%], 1.3 to 3.0; $P = 0.005$) but was not significant for the other pathogens.

The figures for the different pathogens are presented in Table 4.

Bacterial findings and sternal dehiscence. According to clinical findings, the cases could be divided into two groups, based on the presence or absence of sternal dehiscence. Among the 41 cases in the study group, 24 had sternal dehiscence and 17 did not. Of the cases with and without sternal dehiscence, 75% (18/24) and 82% (14/17), respectively, fulfilled the microbiological criterion for infection. According to the microbiological definition, infection caused by CoNS was significantly more prevalent among cases with sternal dehiscence (RR, 2.6; CI [95%], 1.3 to 5.1; $P = 0.009$).

The bacteriological findings for the two groups are presented in Table 5.

DISCUSSION

Possibility of identification of staphylococcal strains. In this study, species identification and MS or MR were insufficient markers for determination of whether staphylococcal isolates belonged to the same strain or not. Eighty-six percent (89/103) of CoNS isolates were *S. epidermidis*, and among those, MS and MR strains were approximately equally common. All *S. aureus* isolates were susceptible to methicillin. Using PFGE, we identified 24 different *S. epidermidis* strains and 8 different *S. aureus* strains.

TABLE 4. Cases fulfilling the microbiological criterion for infection grouped according to infective pathogen found in tissue samples and clinical assessment of infection/non-infection

Infecting species as assessed by microbiological criterion	Clinical assessment (no. of cases)		RR (95% CI)[a]	P value[a]
	Infected	Noninfected		
S. aureus	10	0	2.0 (1.3–3.0)	0.005
CoNS	10[b]	4[c]	ns	ns
P. acnes	1	7	ns	ns
Total	21	11		

[a] ns, not significant. RR, relative risk ratio for clinical assessment of infection in relation to pathogen.
[b] *S. epidermidis*, eight cases; *S. simulans*, one case; *S. haemolyticus*, one case.
[c] *S. epidermidis*, four cases.

TABLE 5. Primary pathogen according to microbiological criterion of infection in cases with and without sternal dehiscence

Infecting species as assessed by microbiological criterion	Clinical diagnosis (no. of cases) of:		RR (95% CI)[a]	P value[a]
	Dehiscence	No dehiscence		
S. aureus	3	7	ns	ns
CoNS	12[b]	2[c]	2.6 (1.3–5.1)	0.009
P. acnes	3	5	ns	ns
Total	18	14		

[a] ns, not significant. RR, relative risk ratio for dehiscence in relation to pathogen.
[b] *S. epidermidis*, 10 cases; *S. simulans*, 1 case; *S. haemolyticus*, 1 case.
[c] *S. epidermidis*, one case.

Number of samples and culturing method. Most studies do not define the number of samples in which a pathogen must be found for that pathogen to be considered the causative agent (9, 11, 12). It can be technically difficult to obtain a sample from fluid or tissue aseptically, especially during an ongoing surgical procedure in infected tissue. When dealing with organisms that belong to the normal skin flora, the question of contamination always arises. For one of our previous studies (14, 15), six samples were taken at the end of the operation from the subcutaneous wound wall of each of a number of patients undergoing elective cardiothoracic surgery. Contamination of the wound with CoNS and/or *S. aureus* occurred in 71% (44/62) of the patients. However, the same strain was found in ≥50% of the samples for only three patients. A finding of the same species or strain in at least half of the samples, provided that five or more tissue samples were obtained, thus seemed to rule out a significant level of contamination.

In 53% (17/32) of the cases fulfilling the microbiological criterion for infection, one or more species or strains other than the primary pathogen were isolated from the samples. If only one sample had been taken from those patients, that other species or strain could have been misinterpreted as being the infective agent.

It is obvious that in most cases, there are sufficient numbers of bacteria in the infected tissue to make it possible to detect them in primary cultures. We detected all cases of *S. aureus* infection and 86% (12/14) of the microbiologically defined CoNS infections from primary cultures. This made it possible to report culturing results (i.e., microbe and antibiogram) to the clinician faster than with the traditional way, i.e., that of subculturing only from enrichment broth.

In two cases, subculturing from enrichment broth on day 4 was necessary to microbiologically identify a CoNS infection. All eight cases of *P. acnes* infection were identified in subcultures from the day 4 enrichment broth. It is thus obvious that to be recognized as an infective agent, this slowly growing anaerobic organism needs culturing conditions that differ from those of the staphylococci.

The subculturing on day 14 did not add any information and can thus be omitted in future clinical use.

Microbiological infection and species in relation to clinical infection. In the study group of 41 patients, 26 (63%) were clinically considered to have an infection and 15 (37%) were

considered noninfected. Adding a microbiological criterion for infection could help identify infected patients among those lacking obvious visible signs of infection. In the group of 15 clinically uninfected patients, the use of a microbiological criterion identified 11 more infected patients, four with CoNS and seven with *P. acnes*.

The probability for agreement between diagnosis of infection based on clinical signs and symptoms and diagnosis of infection based on our microbiological criterion was significant only for cases caused by *S. aureus*, as no case of microbiologically detected *S. aureus* infection was defined as noninfected according to clinical determination. The conclusion is that infection caused by *S. aureus* was easily identified by the surgeon, based on local and/or general signs and symptoms of infection. Microbiologically defined infections caused by CoNS and *P. acnes* apparently did not always cause clinical infective signs and symptoms to the same extent as *S. aureus*.

Among the 14 cases with microbiological CoNS infection, 4 were not assessed by the surgeon as examples of infection. The reasons for the conflicting diagnoses could have been sparse clinical signs and symptoms of infection or symptoms and intraoperative findings of sternal dehiscence and/or sternal gap masking the infection. All four were microbiologically diagnosed from primary cultures on day 2, which supports the infective diagnosis. In cases of this kind, the microbiological diagnosis might help the surgeon to avoid making the mistake of misclassifying the patient as noninfected and the consequential mistake of not prescribing adequate long-term treatment with antibiotics.

The role of *P. acnes* in SSI has been under debate. There are undoubted cases of *P. acnes* osteitis reported (7). The findings by Kamme and Lindberg (6) of a substantial number of *P. acnes* isolates in cases of orthopedic surgery, both from control patients (i.e., without loosening of the hip prosthesis) and from patients with loosened prostheses and nonsignificant bacterial growth levels, convince us that the findings for *P. acnes* should be evaluated carefully. In our study eight cases were identified as infected with *P. acnes*. If, however, the microbiological diagnostic criterion for *P. acnes* infection had included the same requirement as for CoNS and *S. aureus*, i.e., the finding of the same strain in at least half of the samples, some or all of those patients might not have been identified as infected. No method for strain typing of the *P. acnes* isolates was easily available, unfortunately. The role of *P. acnes* as a cause of PSM needs further investigation. The aspect of species identification versus strain typing should be included in such studies.

Five patients were clinically assessed as infected but microbiologically assessed as uninfected. The reason for the differences in assessment was that the results included negative samples (2 patients), multiple samples with less than half of the samples testing as positive (2 patients), and samples which tested positive for a mixture of species or strains (1 patient). These results could be due to antibiotic treatment of the patient, nonrepresentative samples, or prolonged transportation of samples.

The accuracy of clinical diagnosis of infection is hard to question, and one advantage of an improved diagnostic tool for postoperative SSI is the potential to avoid underdiagnosis of CoNS infections, which were missed in 29% (4/14) of the examined cases when not using the microbiological criterion.

The answer to the question of which patient is to be regarded as having a true infection depends, however, on the diagnostic criteria used for infection.

Species in relation to sternal dehiscence. Among the patients with sternal dehiscence for whom a microbiological diagnosis was established, 67% (12/18) had a CoNS infection, compared to 14% (2/14) among those without sternal dehiscence. The difference was statistically significant.

Among the 24 patients with sternal dehiscence, 8 were clinically diagnosed as noninfected. Six of these fulfilled the criteria for microbiological diagnosis of infection, four for CoNS and two for *P. acnes*. Our conclusion is that infection, especially infection caused by CoNS, should be highly suspected for patients with sternal dehiscence, irrespective of clinical signs and symptoms of infection. Based on our data, we suggest that in patients with sternal dehiscence, there should be extensive effort made to improve the accuracy of microbiological diagnosis by multiple sampling before the administration of antibiotics and by the use of good diagnostic methods in the laboratory, including species and strain identification.

MR among CoNS isolates. A total of 60% of the CoNS isolates and 57% of the *S. epidermidis* strains were MR. Among the 14 cases microbiologically infected with CoNS, 64% (9/14) were infected with MR strains. This is a lower proportion than that reported by other authors, who have found MR CoNS in 73% (16) and 92% (9) of studied cases. It is also a slightly lower proportion than that reported for a retrospective study covering the years 1984 to 1995 in the same setting, for which we found that 69% of CoNS isolates were MR (13). To correctly diagnose a postoperative mediastinitis caused by staphylococci, the susceptibility to methicillin in the infective strain is essential, as the MS strains are possible to treat with isoxazolylpenicillins and cephalosporins. An overestimation of MR might lead to unnecessary use of vancomycin. This should be avoided, in consideration of both the risk for selection of genes coding for vancomycin resistance (2) and the interest of the patient, who would otherwise have to undergo a long-term course of intravenously administered antibiotic treatments.

Summary and conclusions. Clinical diagnosis of PSM has a good accuracy for cases that, as determined by findings in tissue samples, are caused by *S. aureus*. When adding a microbiological criterion for infection, requiring multiple tissue samples and finding of the same species or strain in ≥50% of samples, another 30% of cases with CoNS infections, in addition to those clinically assessed as infected, were identified. Our bacteriological method made it possible to obtain a fast diagnosis, as the tissue pieces were cultured directly on to the agar plate before they were put in enrichment broth. This method identified all *S. aureus* infections and a majority of CoNS infections. To ensure a microbiologically correct diagnosis, both with respect to the identification of infective cases among patients lacking clinical symptoms of infection and with respect to correct identification of the primary pathogen, multiple samples before administration of antibiotics, species identification, strain typing, and susceptibility testing should be used. *P. acnes* needs further investigation as a possible cause of PSM. PSM caused by CoNS is significantly related to sternal dehiscence, which is not the case for PSM caused by *S. aureus*. Among our patients was one with PSM caused by *S. simulans*,

which to our knowledge is a finding that has not been reported before.

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