

## Evidence for Transient *Staphylococcus epidermidis* Bacteremia in Patients and in Healthy Humans

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A new blood lysis-filtration culture technique revealed a high incidence of *Staphylococcus epidermidis* in the blood of patients and of healthy people. Of 2,004 blood cultures from patients, the blood lysis method grew *S. epidermidis* in 233 (11.6%), whereas a conventional two-bottle culture system grew this organism in 48 (2.4%). To determine the incidence deriving from the skin, 100 mock blood cultures by each technique were performed. The antecubital fossa was prepared as for a phlebotomy. The needle was inserted through the skin but not into the vein. Needles were cultured by conventional and lysis-filtration culture. A total of 1 conventional culture of 100 (1%) and 2 lysis-filtration cultures of 100 (2%) grew *S. epidermidis*. Of 100 lysis-filtration and conventional control cultures with broth in place of blood, no cultures were positive. Blood samples from 8 of 117 (6.8%) healthy blood donors were positive for *S. epidermidis* by lysis-filtration, whereas no matching conventional cultures were positive. Phage typing patterns of skin and blood strains from selected individuals were the same. *S. epidermidis* isolates were often concomitant with isolates of bona fide pathogens. I conclude that intermittent, transient, asymptomatic *S. epidermidis* bacteremia occurs frequently in patients and in healthy humans.

Traditionally, *Staphylococcus epidermidis* from blood cultures is interpreted as a contaminant microorganism. Recovery of "excess" *S. epidermidis* isolates by the use of new blood-lysing agents benign to bacteria (16, 17) in lysis-filtration blood culture seemed to deserve further study. Lysis-filtration culture has been shown to be more sensitive than conventional blood culture in rabbit septicemia (18) and in hospital patients (5). Whereas recovery of common pathogens in patients was 36% greater by lysis-filtration, recovery of *S. epidermidis* in rabbits septicemic with *S. epidermidis* was 54% greater by lysis-filtration. Phage typing showed that the injected strain was of the same phage pattern as that isolated from blood samples.

It seemed possible, then, that excess recovery of *S. epidermidis* from many blood cultures from patients represented an intermittent presence of this organism in the bloodstream. This report concerns a number of experiments conducted to test this hypothesis.

### MATERIALS AND METHODS

**Blood cultures from patients.** Blood cultures were drawn by the hospital phlebotomy team, physicians, and nurses. Skin disinfection consisted of rubbing the antecubital fossa with a large alcohol-iodine-saturated swab (Operand; Redi-Products Laboratories, Huntington, W.Va.), allowing the skin to dry, and wiping off

the center of the area with 70% isopropyl alcohol. Blood was taken into a 20-ml syringe via a butterfly venipuncture set equipped with a 21-gauge needle. The syringe was then disconnected from the butterfly set, a new needle was attached, and 5 ml of blood was injected into a 50-ml bottle of blood-lysing solution (16). The lysing solution had 0.7% Tween 20 and 0.5% Rhozyme 41 in 0.01 M Sorenson buffer (pH 8.0). The contents of the bottle were mixed by swirling. A 5-ml portion of blood was also injected into each of two commercial brain heart infusion blood culture bottles (BBL Microbiology Systems, Cockeysville, Md.). A new needle was attached to the syringe to inject each bottle. One of the bottles was later vented, via a sterile needle-filter apparatus for aerobic growth, in a laminar-flow hood in the laboratory. Upon transport to the laboratory, the lysis bottle was again swirled and placed in a water bath at 37°C for 1 h. Vacuum filtration was done in a 0.45- $\mu$ m Sterifil D-GS (Millipore Corp., Bedford, Mass.). After filtration, the lower unit was discarded, and the filter port of the upper unit was capped with the supplied sterile closure. Brain heart infusion broth (50 ml) was added, and the unit was incubated at 35°C. The cultures were observed daily for growth and were subcultured at 7 days before discard.

**Mock blood cultures.** A total of 200 blood donors had skin preparation as described above. Skin puncture was done with a 19-gauge needle attached to a 10-ml syringe. Venipuncture was not completed. The needle was withdrawn and washed by aspiration into 10 ml of Trypticase soy broth (BBL). These cultures were processed by lysis-filtration (100 cultures) and by

conventional culture (100 cultures). This procedure was done to determine the rate of positive cultures from skin puncture, excluding the rate from venipuncture.

**Lysis-filtration and conventional blood culture controls.** In the laminar-flow hood, broth was added to lysis solution, and the mixture was processed through the filtration and culture steps exactly as for blood cultures. The same was done for a one-bottle conventional culture. A total of 100 cultures by each procedure was done. They were intended to provide the rate of *S. epidermidis* contamination to be expected if the technique were conducted with the blood sample itself omitted. Cultures that became turbid were subcultured, and the bacterium was identified.

**Samples from healthy blood donors.** The same conventional and lysis-filtration blood culture techniques were used for blood samples from healthy volunteers to determine the rate of *S. epidermidis* recovery.

## RESULTS

There were 2,004 matched conventional and lysis-filtration blood cultures from patients. Of these, 48 (2.4%) conventional blood cultures and 233 (11.6%) lysis-filtration blood cultures were positive for *S. epidermidis*.

A total of 1 of 100 mock blood cultures (skin puncture cultures) done by the conventional technique and 2 of 100 such cultures done by lysis-filtration were positive for *S. epidermidis*.

Of 100 lysis-filtration cultures and 100 conventional cultures in which 5 ml of broth supplanted the blood sample, there were no recoveries of *S. epidermidis*. This was a control series to test only for the rate of contamination.

Blood samples from 8 of 117 (6.8%) blood donors were positive for *S. epidermidis* by lysis-filtration. The matching conventional blood cultures were all negative.

## DISCUSSION

In a previous study the recovery of *S. epidermidis* from induced septicemia in 189 rabbits was 23.0% by the conventional technique and 49.4% by lysis-filtration (18). Lysis-filtration detected bacteremia of a minimal nature and a benign outcome that was usually not detected by conventional blood culture.

In the present study the two-bottle conventional blood culture system received 10 ml of blood (5 ml per bottle), and the lysis-filtration blood culture system received 5 ml. Presumably this reduced somewhat the number of *S. epidermidis* recoveries by the latter technique.

Mock blood cultures provided evidence that the skin is not a major source of unexpected *S. epidermidis* growth in blood cultures. One theory for the presence of *S. epidermidis* in blood cultures is that a plug of skin may be removed during phlebotomy and that this is then injected into the medium, providing an inoculum of skin-

carried *S. epidermidis*. If this theory is valid, then there should have been at least as many *S. epidermidis* isolates by the mock blood culture technique as by conventional blood culture, and probably more, since the skin organisms during mock blood cultures were not exposed to the phagocytic and killing activity of blood.

Lysis-filtration is an exquisitely sensitive culture technique for demonstrating *S. epidermidis* in human and rabbit blood. Aside from the evidence presented that these isolations represent bacteria in the bloodstream, it seems reasonable that most of the isolations in this study were due to release of these bacteria from phagocytes. The differences between lysis and conventional methods in microbial recovery were insignificant when saline or broth suspensions of *S. epidermidis* were assayed or when skin puncture cultures were done.

The significance of frequent intermittent transient *S. epidermidis* bacteremia remains to be elucidated. It can be stated, however, that the condition appears to be benign and distinct from the pathological presence of *S. epidermidis*. It occurs in both patients and healthy blood donors. The source may be the rich indigenous flora of *S. epidermidis* over all skin and mucous surfaces, with more extraordinary concentrations on the face, scalp, nasal tract, and axillae (6). The use of *S. epidermidis* phage typing was limited in usefulness, as only about 30% of the strains were typable. Nine strains that typed however, showed the same phage type on the skin of an individual as in the blood, indicating that this type of bacteremia is not nosocomially acquired, but is endogenous in origin.

Data on the incidence of proven *S. epidermidis* infection resulting in septicemia were not accurately available in this hospital study. However, the low rate of *S. epidermidis* nosocomial septicemia (2) or of catheter-associated sepsis (12), if applied to this study, would not significantly alter the data or the conclusions.

If contamination were a major factor in *S. epidermidis* isolation under these experimental conditions, then it should also result in the recovery of contaminants of other species. This did not occur. There was no other species, other than proved pathogens, that showed a notable increase with lysis-filtration culture. There were sporadic recoveries of *Propionibacterium acnes* in both series of blood cultures from patients, but for lysis-filtration culture this could only have occurred in culture bottles that for some reason achieved sufficiently microaerophilic conditions to initiate growth of this species. Anaerobic conditions were not intentionally provided. It may be that under anaerobic conditions, isolations of *P. acnes* would be elevated by lysis-filtration culture, as they were for *S.*

*epidermidis*. This is currently under study.

Wilson et al. (15) isolated *S. epidermidis* from one of three culture bottles obtained from each of four patients who had no evidence of infection. The study included 240 patients. The authors concluded that the isolations were probably contaminants.

Reith and Squier (8) studied people with and without focal infections who were otherwise healthy. There were 53 (27%) of 194 people with focal infections who had positive blood cultures, mostly for streptococci. Of 99 people with no focus of infection, 12 (12%) had positive cultures. The authors attributed some of these to seasonal respiratory infections. They designated the staphylococci recovered, as "questionable." It is notable that streptococcal blood isolates fell from 11% (focal infection) to 2% (no infection), whereas staphylococci rose from 9% (focal infection) to 11% (no infection). This basic difference in the incidence of streptococci and staphylococci could have reflected the occurrence of *S. epidermidis* in transient bacteremia of both healthy people and those with infections.

Bacteremia has been noted after esophageal dilatation (4), sigmoidoscopy (7), and genitourinary tract manipulation (13); postoperatively (10); and after the massaging of foci of infection (9). Bacteremia has been induced in humans after relatively atraumatic dental manipulations (1, 3) and after extractions (14). Even simple tooth brushing resulted in bacteremia in 16% of individuals (11) and in 27% of individuals using a water irrigation device (1). It seems reasonable to predict that other everyday acts of living also result in influxes of bacteria into the blood. The act of eating, particularly of hard foods, results in bacteremia (11). None of these are probable causes of *S. epidermidis* bacteremia, as the organisms reported to be in the blood after oral entry have been streptococci, diphtheroids, and, particularly, anaerobic gram-negative microorganisms. It seems likely that the skin is the source of most *S. epidermidis*-caused transient bacteremia. Most individuals have at least a few lesions, perhaps of minimal nature, but even so representing breaks in epidermal integrity. Exercise, postural changes, or body movement creating pressure areas on the skin, particularly in areas highly populated with *S. epidermidis*, might result in its traversing capillary barriers into the bloodstream.

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#### LITERATURE CITED

- Berger, S. A., S. Weitzman, S. C. Edberg, and J. I. Casey. 1974. Bacteremia after the use of an oral irrigation device. *Ann. Intern. Med.* **80**:510-511.
- Christensen, G. D., A. L. Bisno, J. T. Parisi, B. McLaughlin, M. G. Hester, and R. W. Luther. 1982. Nosocomial septicemia due to multiply antibiotic resistant *Staphylococcus epidermidis*. *Ann. Intern. Med.* **96**:1-10.
- de Leo, A. A., F. D. Schoenknecht, M. W. Anderson, and J. C. Peterson. 1974. The incidence of bacteremia following oral prophylaxis on pediatric patients. *Oral Surg. Oral Med. Oral Pathol.* **37**:36-45.
- Golladay, E. S., J. J. Tepas III, L. R. Pickard, J. J. Seibert, R. W. Brown, and J. A. Haller. 1979. Bacteremia after esophageal dilatation: a clinical and experimental study. *Ann. Thorac. Surg.* **30**:19-23.
- Kagan, R. L., W. H. Schuette, C. H. Zierdt, and J. D. MacLowry. 1977. Rapid automated diagnosis of bacteremia by impedance detection. *J. Clin. Microbiol.* **5**:51-57.
- Kloos, W. E., and M. S. Musselwhite. 1975. Distribution and persistence of *Staphylococcus* and *Micrococcus* species and other aerobic bacteria on human skin. *Appl. Microbiol.* **30**:381-395.
- le Frock, J. L., C. A. Ellis, J. B. Turchik, and L. Weinstein. 1973. Transient bacteremia associated with sigmoidoscopy. *N. Engl. J. Med.* **289**:467-469.
- Reith, A. F., and T. L. Squier. 1932. Blood cultures of apparently healthy persons. *J. Infect. Dis.* **51**:336-343.
- Richards, J. H. 1932. Bacteremia following irritation of foci of infection. *J. Am. Med. Assoc.* **99**:1496-1497.
- Rogosa, M., E. G. Hampp, T. A. Nevin, H. N. Wagner, E. J. Driscoll, and P. N. Baker. 1960. Blood sampling and cultural studies in the detection of postoperative bacteremias. *J. Am. Dent. Assoc.* **60**:171-180.
- Sconyers, J. R., J. J. Crawford, and J. D. Moriarty. 1973. Relationship of bacteremia to tooth brushing in patients with periodontitis. *J. Am. Dent. Assoc.* **87**:616-622.
- Sitges-Serra, A., P. Puig, E. Jaurrieta, J. Garau, and A. Alastrue. 1980. Catheter sepsis due to *Staphylococcus epidermidis* during parenteral nutrition. *Surg. Gynecol. Obstet.* **151**:481-483.
- Sullivan, N. M., V. L. Sutter, W. T. Carter, H. R. Attebury, and S. M. Finegold. 1972. Bacteremia after genitourinary tract manipulation: bacteriological aspects and evaluation of various blood culture systems. *Appl. Microbiol.* **23**:1101-1106.
- Sweet, J. B., V. J. Gill, M. J. Chusid, and R. J. Elin. 1978. Nitroblue tetrazolium and Limulus assays for bacteremia after dental extraction: effect of topical antiseptics. *J. Am. Dent. Assoc.* **96**:276-281.
- Wilson, W. R., R. E. van Scoy, and J. A. Washington II. 1975. Incidence of bacteremia in adults without infection. *J. Clin. Microbiol.* **2**:94-95.
- Zierdt, C. H. 1982. Blood-lysing solution nontoxic to pathogenic bacteria. *J. Clin. Microbiol.* **15**:172-174.
- Zierdt, C. H., R. L. Kagan, and J. D. MacLowry. 1977. Development of a lysis-filtration blood culture technique. *J. Clin. Microbiol.* **5**:46-50.
- Zierdt, C. H., D. L. Peterson, J. C. Swan, and J. D. MacLowry. 1982. Lysis-filtration blood culture versus conventional blood culture in a bacteremic rabbit model. *J. Clin. Microbiol.* **15**:74-77.