

Frequency of Isolation of *Staphylococcus intermedius* from Humans

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We collected 3,397 consecutive isolates of coagulase-positive staphylococci from various specimens of hospitalized patients. All were retrospectively classified as *Staphylococcus aureus*, except two which were identified as *S. intermedius*: one isolated from the nasal flora of a healthy carrier and the other isolated from pleural fluid, probably as a sample contaminant.

Staphylococcus intermedius is a coagulase-positive staphylococcal species. It is recognized essentially as a common component of the skin, oral, or nasal flora of healthy dogs, where it may be also an invasive pathogen (1, 2, 8, 18, 22). In human beings, *S. intermedius* is rarely isolated, even among individuals with frequent exposure to animals (23). It is, however, essentially responsible for some canine-inflicted human wound infections, thus representing a zoonotic pathogen (7, 12, 22). It appears also to be a rare agent of non-canine-inflicted wound infections (12) or an opportunistic pathogen responsible for invasive infections in immunocompromised patients. One case of infective endocarditis in a human immunodeficiency virus-seropositive patient (13) and one case of catheter-related bacteremia (25) have been reported. *S. intermedius* was also considered the etiologic agent in an outbreak of food poisoning (10). Of the coagulase-positive staphylococci, only *S. aureus* subsp. *aureus*, which is widespread in nature, including humans, mammals, and birds, is a frequent human opportunistic infective agent (11). The other coagulase-positive species include *S. aureus* subsp. *anaerobius*, the coagulase-positive strains of *S. hyicus*, *S. delphini*, and *S. schleiferi* subsp. *coagulans*. *S. aureus* subsp. *anaerobius* was isolated from sheep (4); the coagulase-positive strains of *S. hyicus* were isolated from pigs, cattle, and goats (5); *S. delphini* was isolated from dolphins (28); and *S. schleiferi* subsp. *coagulans* was isolated from dogs (9) and rarely from humans (26).

Here we report the rate of occurrence of *S. intermedius* among coagulase-positive staphylococcal isolates obtained from specimens of hospitalized patients. In the context of a study initially done to detect *S. aureus* genomic variants of gamma toxin, DNAs of coagulase-positive isolates were tested retrospectively for the presence of a specific DNA sequence belonging to the *hlgC* gene for gamma toxin, which has been described only in *S. aureus* (3, 15–17, 21). The screening method, dot blot DNA hybridization, was chosen because it allowed simultaneous testing of a few hundred isolates. The probe used (1806 5'-TTGTTTTATCTTCTGTCCCTT-3' 1787 in *hlgC* from strain ATCC 49775; EMBL-GenBank accession no. X81586) had a nucleotide sequence strictly conserved within the genes encoding the three known variants of gamma toxin. It was 5' ³²P labelled by standard methods (20). For the dot blot assay, performed as described by Rifai et al. (19), staphylococci were grown in 200 µl of CCY (Casamino Acids-yeast extract) modified broth medium (6).

After hybridization, a negligible background was obtained. The probe used did not hybridize with the DNA of *S. epidermidis*, *S. haemolyticus*, *S. saprophyticus*, *S. lugdunensis*, *S. schleiferi* subsp. *schleiferi*, *S. schleiferi* subsp. *coagulans*, *S. simulans*, *S. cohnii*, *S. warneri*, *S. hyicus*, *S. sciuri*, or *S. chromogenes*. This probe also did not hybridize with the DNAs of 30 of the 51 strains of *S. intermedius* mentioned by Prévost et al. (14). The 21 remaining strains were not tested for the presence of the *hlg* gene (15a). However, all of the 51 isolates of this species tested so far produce another toxin, the nucleotide sequence of which has 61% identity with that of the gamma toxin.

During 7 months of 1994, we obtained 3,397 consecutive isolates of coagulase-positive staphylococci from various samples of hospitalized patients. The isolates whose DNAs hybridized weakly or did not hybridize with the probe were identified by the ID 32 STAPH system (bioMérieux, Marcy-L'Étoile, France) after control for the production of free coagulase. Of the 3,397 isolates, 3,395 were identified as *S. aureus* and 2 were identified as *S. intermedius*. One *S. intermedius* isolate was obtained from a nasal swab in pure culture in the context of a protocol intended to detect healthy nasal carriers of *S. aureus*. The carrier was an 82-year-old woman with chronic renal insufficiency undergoing continuous ambulatory peritoneal dialysis. She was not known to have had recent contact with animals. The numeral profile obtained with the ID 32 STAPH system was 3671 5660 1. The other *S. intermedius* isolate was obtained from pleural fluid in a 63-year-old man who had received a cardiac transplant 3 months before. He was hospitalized 2 months after surgery because of right ventricular failure with a hemodynamic pleural effusion which had been twice evacuated. The first puncture was sterile, whereas the second was positive for *S. intermedius*. However, the absence of clinical and biological signs of infection, the late culture, the small number of colonies obtained from blood agar, and the nature of the transudate effusion led to the conclusion of sample contamination from an undetermined origin. The numeral profile obtained with the ID 32 STAPH system was 3673 5660 1. These two profiles are typical for *S. intermedius* and differ markedly from those of *S. aureus*, *S. hyicus*, and *S. schleiferi* in the database of the manufacturer. Additionally, the biochemical reactions for both isolated strains differ from those of the type strain of *S. delphini* by the trehalose acidification and heat-stable nuclease activity tests.

In a routine medical laboratory, detection of free coagulase is frequently used as the major criterion to distinguish *S. aureus* from the other staphylococci usually encountered in human samples. This simple identification procedure may lead to misidentifications for at least two reasons: there are some rare but

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true coagulase-negative *S. aureus* strains (24, 27), and other coagulase-positive staphylococcal species, such as *S. intermedius* or *S. schleiferi* subsp. *coagulans*, have been found occasionally in human beings. To our knowledge, our work is the only one giving the prevalence of *S. intermedius* isolates among coagulase-positive staphylococci obtained in a routine analysis laboratory in a clinical setting. This frequency in hospitalized patients, about $6 \cdot 10^{-4}$, is very low; these patients had no increased risk of acquiring an animal-linked *S. intermedius* infection. Previous isolations of *S. intermedius* from humans were described as either case reports (12, 13, 25) or in the context of studies on populations with an increased risk of acquiring *S. intermedius* infections (veterinarians and patients bitten by dogs) (7, 12, 22, 23). In the latter population, the prevalence of *S. intermedius* isolation is, as expected, higher than in our study. No coagulase-positive strain of *S. hyicus* or *S. schleiferi* subsp. *coagulans* was identified. No case of a true human infection caused by *S. intermedius* was detected in this series. The very low frequency of coagulase-positive staphylococci other than *S. aureus* in human specimens does not justify efficient but expensive identification of all coagulase-positive staphylococci by using commercially available galleries. A cheaper alternative for identification of *S. intermedius* may be the use of biochemical tests for β -galactosidase or pyrrolidonyl arylamidase activity, acetoin production, or polymyxin B resistance (11).

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