Catalase-negative, methicillin-resistant *Staphylococcus aureus* as a cause of septicemia

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Staphylococcus aureus *catalase-negativo resistente a meticilina como causa de septicemia*

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key words

Catalase-negative

Methicillin-resistant

abstract

A catalase-negative methicillin-resistant *Staphylococcus aureus* (MRSA) was isolated from blood, venous catheter spike and bone marrow collected from an HIV-positive man with lobar pneumonia and sepsis after ten days of hospitalization. The isolate was resistant to oxacillin (positive for penicillin-binding protein 2'), ceftriaxone, clindamycin and clarithromycin, and susceptible to vancomycin. This is the first case of septicemia due to a catalase-negative *S. aureus* reported in Brazil, and, to our knowledge, it is the first case of catalase-negative MRSA reported in the literature. We believe that the patient acquired the *S. aureus* infection within the hospital environment since it was isolated ten days after hospitalization, it was isolated in a venous catheter spike, and the antibiotic resistance profile is similar to other *S. aureus* isolates recovered from infections in our hospital.

unitermos

Catalase-negativo

Resistente a meticilina

resumo

Em um paciente HIV-positivo, com pneumonia lobar e septicemia, foi isolada, após dez dias de internação, uma cepa de Staphylococcus aureus *catalase-negativa, resistente a meticilina/oxacilina (MRSA), de culturas de sangue, cateter venoso central e medula óssea. A cepa era resistente a oxacilina (PBP 2' positivo), ceftriaxona, clindamicina e claritromicina, e sensível a vancomicina. Este é o primeiro caso, reportado no Brasil, de uma septicemia por* S. aureus *catalase-negativo e, em nosso conhecimento, o primeiro caso de um* S. aureus *catalase-negativo resistente a meticilina. Nós acreditamos que o paciente tenha adquirido a infecção no ambiente hospitalar, uma vez que esta cepa foi isolada aos dez dias de internação, foi isolada em cateter venoso central e o perfil de sensibilidade aos antimicrobianos é semelhante ao dos* S. aureus *de infecções nosocomiais que ocorrem em nosso hospital.*

Introduction

The occurrence of catalase-negative *S. aureus* as an infective agent is rare, with no reports in Brazil and no cases of a MRSA catalase-negative *S. aureus* reported in the literature.

In 2000, we isolated a catalase-negative *Staphylococcus aureus*, methicillin-resistant, from blood, venous catheter spike and bone marrow, collected from an HIV-positive man.

Considering that the detection of catalase-negative staphylococci is an uncommon event, clinical laboratories should be encouraged to include catalase testing in their *S. aureus* identification protocols in order to collect more information about incidence and potential virulence of these unusual isolates.

Case report

In February 2000, a 30-year-old HIV-positive man who was using lamivudine, zidovudine, saquinavir, and sulfatrimethoprim for *Pneumocystis* prophylaxis presented

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to the emergency care service of the Emílio Ribas Infectology Institute (IIER). He complained of fever, general myalgia, chest pain, and dyspnea. Lobar pneumonia was diagnosed and, because the patient also presented clinical signs of septicemia, he was hospitalized and treated with ceftriaxone, clarithromycin, and clindamycin. After a convulsive episode and loss of consciousness due to concomitant neurotoxoplasmosis, pyrimethamine and hydrocortisone therapies were also started. During his 11 days of hospitalization the neurological and septic conditions of the patient did not improve, and he eventually died.

Bacteriology

A blood culture drawn on the fourth day of hospitalization was negative, but cultures from blood, the venous catheter spike, and bone marrow, collected ten days after admission all grew gram-positive cocci. Opaque, yellow-pigmented, beta-haemolytic colonies of 1-2mm in diameter were isolated on 5% sheep blood agar, after aerobic incubation at 35°C. The cellular morphology was typical of *S. aureus*, but the catalase was negative. The strain has not grown in anaerobic atmosphere.

The isolate was initially identified as *S. aureus* at the Emílio Ribas Infectology Institute (IIER) lab, São Paulo, Brazil. This identification was obtained using the Vitek GPI card (biolab Merieux), but only when the catalase reaction was entered as positive rather than negative. Subsequently, it was referred to the Bacteriology Dept. of Adolfo Lutz Institute (IAL), São Paulo, Brazil, for confirmation by standardized biochemical testing (4) and for antibiotic susceptibility testing. The minimum inhibitory concentration (MIC) of the vancomycin was determined by E-test (AB-Biodisk, Solna, Sweden) according to the manufacturer's instructions and it was found to be susceptible to vancomycin (MIC 2μg/ml). The strain was resistant to oxacillin, ceftriaxone, clindamycin and clarithromycin by both disk diffusion (9) and the Vitek (ATB-GP card).

The strain was also referred to the National Centre for *Streptococcus* (NCS), Alberta, Canada, where the identification of catalase-negative *S. aureus* was confirmed by conventional biochemical testing (6) and by cellular fatty acid analysis (Microbial ID, Inc, Newark, DE). The strain was also confirmed as methicillin-resistant by the detection of penicillin-binding protein 2' (PBP 2') using the MRSA screen™ test (Denka Seiken Co., Ltd., Tokyo, Japan). The low-affinity PBP in MRSA, termed PBP 2' or PBP 2a, is encoded by the chromossomal gene mecA (5) and is thought to function as a b-lactam-resistant transpeptidase.

The **Table** shows the phenotypic tests performed by the three labs which identified the strain.

Discussion

Members of the genus *Staphylococcus* are gram-positive cocci that are arranged in groups, or clusters, are non-motile and do not produce spores. Staphylococci have a cell wall that is typical of all gram-positive bacteria and a G + C content range of 30-40mol% (4). Most species are catalase-positive, except *Staphylococcus saccharolyticus* and *Staphylococcus aureus* subsp. *anaerobius*, which are catalase-negative and grow faster under anaerobic conditions (6). *Staphylococcus aureus* is routinely identified by its ability to produce coagulase, and a heat-stable nuclease but these characteristics are not limited to this species since *S. schleiferi*, *S. hyicus* and *S. intermedius* may also be positive in both of these tests (4).

To this date, ten cases of infection due to catalase-negative *Staphylococcus aureus* have been published, eight of them in humans (1, 2, 3, 7, 8, 10, 11, 12). This is the first case of septicemia due to catalase-negative *S. aureus* reported in Brazil, and the fourth case that has been reported in the literature (3, 11, 12). Furthermore, to our knowledge, it is the first published case of catalase-negative MRSA infection.

It is difficult to estimate the true incidence of catalase-negative *Staphylococcus aureus* strains that are recovered from clinical specimens because most bacteriology laboratories do not routinely perform the catalase test on coagulase-positive colonies with typical *Staphylococcus aureus* morphology. When these atypical strains are encountered, the porphyrin test is a useful additional identification tool (13). All members of *Staphylococcus* genus, including catalase-negative strains, will be porphyrin-positive.

We believe that the fatal outcome of this case was due to the patient's severe underlying clinical condition, immunodeficiency, and the continuation of inappropriate antibiotic therapy, which had been initiated to treat the original diagnosis of lobar pneumonia that was assumed to be caused by pneumococci or possibly by *Mycoplasma*.

It could be that the patient acquired the MRSA infection within the hospital environment through the central venous

Table **Phenotypic characteristics of the catalase-negative, methicillin-resistant *Staphylococcus aureus* isolate described in this study**

Test	IIER*	IAL**	NCS***
Gram stain	Gram-positive *cocci* in clamps	Gram-positive *cocci* in clamps	Gram-positive *cocci* in clamps
Catalase	Negative	Negative	Negative
ALA	NP	Positive	Positive
Slide coagulase	NP	NP	Negative
Tube coagulase	Positive	Positive	Positive
Heat-stable nuclease	NP	NP	Positive
Bacitracin	Susceptible	NP	Resistant
Novobiocin sensitivity	Susceptible	NP	Susceptible
Voges Proskauer	NP	NP	Positive: weak
Pyrrolydonylamidase	Negative	Negative	Negative
Nitrate reduction	NP	Positive	Positive
Arginine	Negative	Positive	Positive
Ornithine	NP	Negative	Negative
Urease	NP	Positive	Positive
40% bile growth	Negative	Negative	NP
10% bile growth	Positive	NP	NP
Esculin hydrolysis	Negative	NP	Negative
Glucose	Acid	Acid	NP
Arabinose	Negative	Negative	NP
Cellobiose	Negative	NP	Negative
Inulin	Negative	NP	NP
Lactose	Negative	Acid	Acid
Maltose	NP	Acid	Acid
Mannitol	Acid	Acid	Acid
Mannose	Acid	Acid	Acid
Melezitose	Negative	NP	NP
Melobiose	Negative	NP	NP
Raffinose	Negative	Negative	Negative
Ribose	Negative	NP	NP
Salicin	Negative	NP	NP
Sorbitol	Negative	Negative	NP
Sorbose	NP	Negative	NP
Sucrose	Acid	Acid	NP
Trehalose	Acid	Acid	Acid
Xylose	Negative	Negative	Negative
Pyruvate	NP	Negative	NP
Indol production	NP	Negative	NP
DNAse	NP	Positive	NP
NaCl 6.5% growth	Positive	Positive	NP
Peptone, glucose	Acid	NP	NP
Hemicellulase	Negative	NP	NP
Urea	Negative	NP	NP
Tetrazolium chloride	Negative	NP	NP
Pullulan	Negative	NP	NP

*IIER, Instituto de Infectologia Emílio Ribas; **IAL, Instituto Adolfo Lutz; ***NCS, National Centre for Streptococcus; NP, not performed.

catheter, since the organism was isolated ten days after hospitalization, the previous blood culture, collected four days after he was admitted, was negative and it was also isolated in the venous catheter spike.

The antibiotic resistance profile shown for this *S. aureus* strain is similar to other *S. aureus* isolates recovered from infections in our hospital (data from the local hospital infection control service), suggesting that this was a nosocomially acquired infection. Further molecular analysis will be necessary to establish the clonal relatedness of this unusual MRSA isolate to others that have been recovered from this environment.

As noted by others (1, 12), the occurrence of catalase-negative *S. aureus* as an infective agent should be recognized, and clinical laboratories should be encouraged to include catalase testing in their *S. aureus* identification protocols in order to collect more information about incidence and potential virulence of these unusual isolates.

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