Relapse of *Enterococcus hirae* prosthesis valve endocarditis

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Abstract (41 words)

*Enterococcus hirae*, a Gram-positive bacterium, is a rare isolate in clinical specimens. We report an unusual case of a relapse of prosthetic valve endocarditis due to *E. hirae* six months after the initial episode. Clonal relationship was proven by genomic analysis.

Case report

In April 2008, a 78 year-old woman presented with a 5-months history of fever, generalized weakness, and a 7 kilos weight loss. She had a history of diabetes mellitus, hypertension, and had undergone in 2001 an aortic valve replacement with a bioprosthetic valve. On admission, her temperature was 38°C and cardiac examination revealed a 3/6 systolic murmur. Transthoracic and transesophageal echocardiographies showed no evidence of endocarditis. Five blood cultures yielded colonies of Gram-positive cocci with a typical morphology of *Streptococcus*. On blood agar culture, colonies were circular, smooth, reaching 0.2 to 0.5 mm after 24 to 48 h of incubation in the presence of 5 % CO₂. The strain was identified by phenotypic method and genetic analysis. The phenotypic identification was based upon colony and Gram stain morphologies and a negative catalase reaction. The cocci reacted with both Lancefield groups F and D antisera (Streptex®, Diamondial, Sees, France). The biochemical identification remained difficult even by using semi-automated system. The accurate identification to species level was not possible with the IDGP N052 card (bioMérieux, Marcy-l’Etoile, France) due to low discrimination (50.52 % for *Enterococcus durans* and 49.48 % for *Enterococcus faecium*). The rapid ID 32 STREP identification (bioMérieux) failed with an unacceptable profile to *Enterococcus gallinarum*, with the current database.

The strain was correctly identified to the species level as *Enterococcus hirae* by genetic methods using 16SrRNA and sodA_int genes sequencing as previously described (5,12). Sequence analysis of this strain yielded 99.81 % and 99.76 % identities with the sequences of the type strain of *E. hirae* (16S rRNA gene, GenBank accession number no. AB362598) and *E. hirae* (sodA gene, GenBank accession number no. AJ387916), respectively. As the growth was deficient, antibiotic susceptibility testing was performed with an agar diffusion technique using a Mueller-Hinton agar supplemented with 5 % horse blood (bioMérieux). The strain was susceptible to amoxicillin (MIC, 0.032 µg/mL, measured by E-test technique), moxifloxacin, vancomycin, teicoplanin, erythromycin, rifampin but resistant to clindamycin,
tetracycline, fosfomycin and exhibited low-level resistance to streptomycin, kanamycin and gentamicin.

Despite a normal echocardiography, the diagnosis of possible infective endocarditis was established (Table 1), and intravenous amoxicillin (200 mg per kg a day) and gentamicin (3 mg per kg a day) were initiated. After 2 weeks, gentamicin was discontinued, and rifampin (20 mg per kg a day) was initiated. Antimicrobial treatment was administered for 6 weeks. The patient improved clinically, became afebrile 48 hours after antibiotic initiation, and blood cultures became negative. Colonoscopy investigation revealed multiple colonic polyps, which were endoscopically removed. Histological examination showed no evidence of cancer.

Four months after discontinuation of antimicrobial therapy, the patient was readmitted for fever. Echocardiography showed a vegetation involving the aortic prosthetic valve, and antimicrobial therapy with amoxicillin and gentamicin was initiated. Two days later, the two blood culture obtained before initiation of antimicrobial therapy yielded a Gram-positive coccus with the same cultural characteristics and the same antibiotic susceptibility pattern as described above, which was identified as *Enterococcus hirae*. A diagnosis of definite endocarditis was made according to the modified Duke criteria (7) (Table 1), and the patient received the same antimicrobial therapy than for the initial episode, i.e. intravenous amoxicillin for 6 weeks, intravenous gentamicin for 2 weeks, and oral rifampin for 4 weeks. She was contra-indicated for surgery because of a poor general condition.

The evolution was quickly favorable, the patient became afebrile, and blood cultures remained negative. Echocardiography performed one month later did not reveal any vegetation or dysfunction of the aortic bioprosthesis. Control of colonoscopy showed a 20 mm colonic polyp which was removed; histological examination revealed an adenoma without evidence of neoplasia.

Both *E. hirae* were then genetically compared in order to differentiate relapse from reinfection of the endocarditis. Analysis of the genomic patterns of both isolates after pulsed field gel electrophoresis after restriction by *SmaI* showed that they were clonally-related according to Tenover criteria (Figure 1-16).

The patient remained clinically well one year after completion of therapy.

*Enterococci* are frequently identified as important causes of infections in humans, such as bacteremia, endocarditis, and urinary tract infections. Most of enterococcal strains isolated from clinical samples belong to two species, *Enterococcus faecalis* and *Enterococcus faecium*. *Enterococcus hirae* mainly causes infections in various animal species (2, 3), and
only three cases of human infection have been reported to date (1, 4, 11). In these three reports, *E. hirae* was responsible for septicemia, spondylodiscitis and native valve endocarditis.

We describe a case of *E. hirae* prosthetic valve endocarditis, which relapsed despite adequate antibiotic therapy according to the European guidelines (14). The patient initially presented with *E. hirae* bacteremia and a diagnosis of possible endocarditis was established as she met 1 major and 2 minor of the modified Duke criteria for the diagnosis of endocarditis (7). When bacteremia relapsed, a diagnosis of definite endocarditis was made, as the patient met 2 major criteria. To our best knowledge, our case constitutes the first description of prosthetic valve endocarditis due to *E. hirae*, and the fourth report of human infection caused by this bacterium. The source of infection in our patient was probably the digestive tract, as colonoscopy showed multiple colonic polyps. Currently, there is no demonstrated relationship between *E. hirae* infection and colonic pathology, but very few cases of infections due to this particular bacterium have been reported; however enterococci are commensal species of the human intestinal tract, and *E. hirae* has been involved in colonic pathology in animals (6,9). Therefore, we decided to perform a colonoscopy in order to seek for intestinal disease, as it is recommended for other bacteria colonizing the gastrointestinal tract.

The only other reported case of *E. hirae* endocarditis, by Poyart *et al.* in 2002, was a 72-year old man with a native valve endocarditis, who was treated with ampicillin, gentamicin and rifampin (11). Despite an in vitro susceptibility of the strain, this regimen was not able to sterilize the vegetation and endocarditis relapsed 3 months after discontinuation of antimicrobial treatment. The same phenomenon occurred in our patient, although adequate antimicrobial therapy was administered for 6 weeks, suggesting that *E. hirae* would be difficult-to-treat bacteria, causing relapsing infections (8). Moreover, we hypothesize that the particularly low virulence of this strain living likely in a dormant state may explain the delayed relapse. The treatment of infections due to this particular bacterium may necessitate prolonged antimicrobial therapy, and a closer surveillance of clinical parameters (echocardiography and blood cultures after discontinuation of antibiotics) in order to detect a relapse. For our patient, we chose to add rifampin to the amoxicillin/gentamicin combination which is recommended for treatment of enterococcal prosthetic valve endocarditis (14). Rifampin has shown good activity in *Staphylococcus aureus* biofilm (13), and is recommended for treatment of staphylococcal prosthetic valve endocarditis (14). However, it has not been extensively studied in *Enterococcus* spp. infections, although some promising results have been reported (10,18). As optimal therapy for the treatment of *E. hirae* infection
remains unknown and valve replacement was contra-indicated for our patient, we hypothesized that addition of rifampin could optimize antibiotic therapy and reduce the risk of relapse.

At last, identification of enterococci, other than *Enterococcus faecalis* and *Enterococcus faecium*, in the absence of additional tests (11) can still remain difficult, with potentially serious implications for clinical management. As reported previously, Vitek2® automated system may experience difficulties in properly identifying *E. hirae* and *Lactococcus garvieae*, another Gram positive cocci involved in endocarditis. Accurate identification of enterococci can be achieved with molecular techniques (17).

Thus, misidentification of unusual *Enterococcus* species by the different semi-automated commercial identification methods might occur and accurate molecular identification is required as this species remains rare (8,15).

Acknowledgments:

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References


**Table 1.** List of modified Duke criteria for each episode of endocarditis

<table>
<thead>
<tr>
<th></th>
<th>First episode</th>
<th>Second episode</th>
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<tr>
<td><strong>Major criteria</strong></td>
<td>5 separate blood cultures yielded <em>Enterococcus hirae</em></td>
<td>Echocardiography positive for infective endocarditis with demonstration of aortic vegetation</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2 separate blood cultures yielded <em>E. hirae</em></td>
</tr>
<tr>
<td><strong>Minor criteria</strong></td>
<td>Bioprosthetic aortic valve</td>
<td>Bioprosthetic aortic valve</td>
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<td></td>
<td>Fever</td>
<td>Fever</td>
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NB: sensitivity of the modified Duke criteria is diminished when infection affects a prosthetic valve.
FIG. 1. : Pulsed-Field Gel Electrophoresis banding patterns after *SmaI* digestion of both clinical isolates of *E. hirae*. Percentages of similarity are shown above the dendrogram. Isolate dates are on the right.