Parallel detection of bacteria with culture methods and 16S rRNA gene analysis: Preliminary results from a prospective cohort study of prosthetic joint infections

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Introduction

Artificial joints have become indispensable treatment for osteoarthritis. Approximately 15,000 primary arthroplasty procedures and 2,000 revisions are performed each year in Denmark (1). The number of patients with prosthetic implants will continue to grow mainly due to an aging population. However, the implants can lead to serious complications, one of which is bacterial infection. The risk is estimated to be 0.5-2% for primary arthroplasties and even higher for revision surgeries. The infections are difficult to diagnose due to vague symptoms and a modest inflammatory response. Routine culture-based methods fail to demonstrate a bacterial agent in a significant proportion of cases. The failure may be due to inappropriate culture conditions, prior antibiotic treatment, fastidious or atypical organisms, and the biofilm mode of growth.

The successful diagnosis and treatment of prosthetic joint infections (PJI) require a close cooperation between surgeons, radiologists, specialists in nuclear medicine, clinical microbiologists and molecular biologists. At Aalborg Hospital we have established a unique diagnostic algorithm within the framework of the PRIS Innovation project. In this presentation the objective was to compare positivity rates of extended cultures for the presence of bacteria with the positivity rates of detection of 16S rRNA genes in parallel samples. We report preliminary results for the first 65 patients recruited consecutively December 2011 through June 2012.

M&M

The study comprised patients seeking medical attention with a painful hip or knee prosthesis. Twenty-five had PA according to the physician’s judgement; the remaining 40 patients had an aseptic loosening (AL). Fifty-four underwent surgical revision, seven had a joint puncture, and four had two procedures (joint puncture + revision or revision + revision).

Results

Surgical revisions (table). In the PJI group (n=18), a total of 74 samples were positive by culture and 58 by 16S rRNA gene analysis. We obtained agreement with 16S rRNA gene analysis in 57 procedures in 56 patients. Using the established sonication procedure (2,3,4) and optimized culture methods we obtained agreement with 16S rRNA gene analysis in 57 procedures in 56 patients. Sonication has also previously been shown to be valuable for culturing of prosthetic implants, supported by a grant from the Danish Agency for Science, Technology and Innovation, Danish Ministry of Science, Innovation and Higher Education.

Parallel detection of bacteria with culture methods and 16S rRNA gene analysis: Preliminary results from a prospective cohort study of prosthetic joint infections

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The prosthetic components were processed by submerging into isotonic buffer (XPBS) for sonication (40 kHz) and aliquots of the buffer were centrifuged to concentrate the bacterial content and used for further processing (2). Bacteriological cultures were cultivated on agar plates and in broth medium under aerobic and anaerobic conditions for 14 days. For full length 16S rRNA gene analysis DNA were extracted and screened by endpoint PCR.

Flow diagram: Joint puncture: 3 samples were obtained, 2 for culture (1 synovial fluid culture and 1 for bacterial culturing) and 1 for 16S rRNA gene analysis. During revision up to 15 samples were obtained: 9 for 16S rRNA analysis, 3 for bacterial culturing and five for other purposes. Prior to this study, bone biopsies, prosthetic components, and biofilm swabs were not available for microbiological diagnosis of PJI.

Table 1: Concordance between 16S rRNA gene analysis (PCR) and optimized culture methods.

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Concordance 16S rRNA (PCR)</th>
<th>Culture positive</th>
<th>PCR positive</th>
<th>PCR negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Joint fluid</td>
<td>16S: 43/65</td>
<td>65/65</td>
<td>65/65</td>
<td>0/65</td>
</tr>
<tr>
<td>Joint biopsy</td>
<td>16S: 43/65</td>
<td>65/65</td>
<td>65/65</td>
<td>0/65</td>
</tr>
<tr>
<td>Synovial fluid</td>
<td>16S: 64/65</td>
<td>65/65</td>
<td>65/65</td>
<td>0/65</td>
</tr>
<tr>
<td>Prosthetic component</td>
<td>16S: 64/65</td>
<td>65/65</td>
<td>65/65</td>
<td>0/65</td>
</tr>
</tbody>
</table>

Discussion

Prosthetic joint replacements are a successful treatment option for patients suffering from osteoarthritis and rheumatoid arthritis. However, bacterial infections can occur, resulting in the need for early detection and appropriate treatment to prevent further damage to the prosthesis. The correct diagnosis of prosthetic joint infection is essential for successful treatment and patient outcomes.

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Acknowledgements

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Reference


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