

QUESTION 15: Is there a method to detect sessile microorganisms that have resulted in an infection following orthopaedic procedures?

RECOMMENDATION: Yes. Molecular techniques such as polymerase chain reaction (PCR), next-generation sequencing (NGS) and synovial biomarkers such as alpha-defensin or leukocyte esterase have been shown to be powerful tools in detecting prosthetic joint infections (PJI) with negative cultures, although conflicting data exists on PCR. Sonication of explanted prosthetics can enhance both the sensitivity of conventional cultures and PCR.

LEVEL OF EVIDENCE: Strong

DELEGATE VOTE: Agree:85%, Disagree: 9%, Abstain: 6% (Super Majority, Strong Consensus)

RATIONALE

The colonization of prostheses by sessile bacteria is a feared complication of orthopaedic procedures. These microorganisms anchor themselves to the surface of prosthetic implants and form a colony of immobile bacteria cross-linked by an extracellular matrix of polymeric substances, known as biofilm [1]. The presence of biofilm on prosthetic implants, especially that of prosthetic joints, makes both detection and treatment of infections difficult [2]. While there is no gold standard for definitive diagnosis of PJI, a multi-criteria definition created by Musculoskeletal Infection Society (MSIS) is often used to diagnose PJI [3,4]. The MSIS criteria utilizes the obtaining of cultures of joint aspirate or periprosthetic tissue as one of the major criteria to prove the presence of pathogens in the prosthetic joint. Unfortunately, cultures can be unreliable when detecting biofilms [5,6]. Intraoperative cultures alone also can have a high rate of contamination and false positives [7]. Thus, alternative methods of confirming the presence of organisms in PJI have been proposed [8,9]. Some of these diagnostic techniques include PCR, NGS, prosthesis sonication and joint biomarkers.

Polymerase Chain Reaction

The use of PCRs to detect bacterial nucleic acids in prosthesis infections can be an effective way of detecting sessile microorganisms otherwise not picked up in cultures [10,11]. PCR sequencing of bacterial ribosomal nucleic acids has shown to have higher sensitivity in detecting bacteria than culture, as well as identifying polymicrobial infections that may not be picked up by culture [12–15]. Jahoda et al. showed that the use of PCR can detect as few as 590 colony forming units of *S. aureus*, making detection of PJI even in the presence of antibiotics feasible [11]. PCR has also shown benefit in detecting genes responsible for biofilm production and methicillin resistance [11,16].

In spite of the literature describing the merits of PCR, there is data suggesting that the efficacy of PCR is not as high as once thought. Studies have suggested that PCR has similar or less sensitivity for detecting bacteria in PJI as traditional cultures [17–20]. PCR has also been shown to have questionable sensitivity over the last years. A meta-analysis performed by Jun et al. looking at online databases from 2013 to 2017 showed that there has been a decrease in pooled sensitivity compared to a previous meta-analysis performed by Qu et al. in 2013 (0.76, (95% confidence interval (CI) 0.65-0.85) vs.0.86, (95% CI 0.77-0.92) respectively), with no change in specificity [21,22].

Next-Generation Sequencing

Recently, NGS has proven to be efficacious in diagnosis of culture-negative PJIs as well. A prospective study performed by Tarabichi et al. evaluated the accuracy of NGS in identifying PJIs in 78 patients undergoing revision or primary arthroplasties. NGS identified infections in 25 of the 28 cases considered to be PJIs by MSIS criteria (95% CI 71.8% to 97.7%), whereas cultures were only able to identify 17 cases (95% CI 40.6% to 78.5%). In cases where both cultures and NGS were positive, NGS showed a high degree of concordance to traditional cultures as well [23].

NGS has also shown high degrees of detection in synovial fluid samples. Another study conducted by Tarabichi et al. analyzed 86 samples of synovial fluid from the hip or knees of patients undergoing PJI evaluation. They found that NGS had a positive result in 10 samples that were culture-negative. Five of these samples had elevated inflammatory biomarkers, indicating an infectious process, while the other five had negative inflammatory biomarkers. These results suggest that NGS may be a valuable tool for evaluating for PJIs in the preoperative setting, but may also be at risk for false positives [24].

In addition to diagnosing prosthetic infections, NGS may also be useful for identification of causative organisms in culture-negative PJIs [23]. Furthermore, the speed at which NGS can explore an entire genome makes it a superior alternative to PCR [25]. While NGS has exciting potential as a powerful diagnostic tool for culture-negative PJIs, there has been limited data showing

its effectiveness in diagnosing other prosthetic infections. In addition, there has been no direct comparison between the effectiveness PCR and NGS. Finally, it is important to consider that the high sensitivity may predispose NGS to a high false-positive rate and false diagnosis of PJIs [25].

Sonication

The use of sonication to break up biofilm in prosthetic implants has been shown to increase the sensitivity of both cultures and PCR when testing for infection. A prospective study performed by Tani et al. compared the sensitivity and specificity of cultures obtained from sonicated explants to conventional cultures of periprosthetic tissue in 114 patients who underwent hip and knee revisions due to PJI and aseptic loosening. Sonicated cultures had a significantly-increased sensitivity when compared to conventional cultures (77.0% vs. 55.7%). There were no significant differences in specificity of either detection method [26].

There are some studies suggesting that sonication of prosthesis may improve the diagnosing capacity of PCR in the diagnosis of culture-negative PJIs [27–29]. However, their statistical significance remains controversial. A recent meta-analysis of nine studies looking at the efficacy of sonication in PCR was performed by Liu et al. [30] found that PCR for sonication prosthetic fluid was to have clinically acceptable diagnostic values for detecting PJIs, with a pooled sensitivity of 75% (95% CI 0.71 to 0.79) and specificity of 96% (95% CI 0.94 to 0.97) [30].

Joint Biomarkers

Inflammatory biomarkers in the blood such as erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP), as well as synovial fluid leukocyte esterase have been part of the 2011 MSIS criteria and the 2013 consensus group modification criteria in the diagnosis of PJI [3,31]. The updated MSIS criteria put forth by Parvizi et al. in 2018 added the presence of synovial alpha-defensin and synovial CRP as criteria for diagnosis of PJI [4]. Synovial biomarkers such as leukocyte esterase and alpha-defensin have been shown to have high sensitivity and specificity in diagnosis of PJI, and are more specific than serum inflammatory biomarkers [32–34]. The benefit of these biomarkers are that they are faster and less invasive than traditional cultures. Biomarker assays also do not require tissue sampling and may be performed on synovial fluids, which increases the convenience of these tests in diagnosing PJIs in the preoperative setting. The major drawback of joint biomarkers is that they can only indicate the presence of infection and not its specific nature. Therefore, biomarkers are best utilized as a preliminary indicator of the presence or absence of joint infection. They are best followed up by diagnostic assays such as PCR, NGS or cultures to better determine the nature of infection.

Conclusion

There are a number of methods to detect sessile microorganisms in infections following orthopaedic procedures. The use of PCR in the diagnosis of culture-negative PJI has shown to be more sensitive than traditional cultures but there is conflicting data. The use of inflammatory biomarkers in both the blood in synovial fluid is also effective, but cannot characterize the nature of infection or organism involved. NGS is a new test can determine the presence of sessile microorganisms with more precision and speed than traditional cultures. Finally, sonication of explants has shown to improve the sensitivity of both cultures and PCR in diagnosing prosthesis infections.

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