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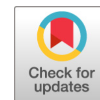
MINIREVIEW



A Real Pain: Diagnostic Quandaries and Septic Arthritis

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ABSTRACT Rapid diagnosis and treatment of an infected joint are paramount in preserving orthopedic function. Here, we present a brief review of the many challenges associated with the diagnosis of both septic arthritis and prosthetic joint infections. We also discuss the many laboratory tests currently available to aid in the accurate diagnosis of joint infection, as well as emerging diagnostics that may have future utility in the diagnosis of these challenging clinical entities.

KEYWORDS culture, diagnostics, prosthetic joint infection, septic arthritis

Septic arthritis (SA), or infection of a joint, requires rapid and aggressive treatment. The majority of SA is due to bacterial infection, with fungal and mycobacterial infections less frequently observed. The acute inflammatory reaction caused by microorganism invasion of synovial tissue can lead to permanent destruction relatively quickly. Consequently, 10 to 32% of patients with SA suffer long-term decreased mobility, with an in-hospital mortality rate of 3 to 15% (1–3). Thus, rapid identification of infected joints is paramount (4, 5). In addition, prosthetic joint infection (PJI) is a complication of total joint arthroplasty with substantial morbidity, yet despite this, can be exceedingly difficult to diagnose. Patients with PJI often require multiple revision surgeries and extended antimicrobial therapy and, in addition to a decreased quality of life, have a 2-fold higher risk of inpatient death with each surgery than that of patients with an aseptic revision (6).

EPIDEMIOLOGY OF SEPTIC ARTHRITIS

SA occurs most commonly among the elderly and in children $<$3 years old (5). Proven or probable SA occurs in Western Europe at an annual rate of 4 to 10/100,000 patient years (7), with SA accounting for 0.01% of all adult emergency room visits from 2009 to 2012 in the United States, and the majority (82 to 84%) of patients require hospitalization (8, 9). Both systemic and local factors influence susceptibility to SA, and these vary according to the patient population and mode of infection, i.e., hematogenous seeding versus direct inoculation (4). Common risk factors and infectious etiologies are shown in Tables 1 and 2, respectively. A detailed description of the etiologic agents of SA is beyond the scope of this review, but they are discussed in detail in references 10 and 11.

Critically, previous damage to the joint architecture caused by rheumatoid arthritis, osteoarthritis, or crystal arthropathies (e.g., gout) increases the risk of SA (5, 7). Risk factors for gonococcal arthritis include female gender and pregnancy, though its incidence in Western countries has decreased over the past few decades (10). Injection drug users are at increased risk of SA and are more likely to have polymicrobial or fungal SA or SA due to less frequently encountered organisms (12). Importantly, direct inoculation of the joint can occur during joint injections and arthroscopic procedures (7). Local risk factors for the development of SA in children include trauma to the joint (e.g., puncture wounds), as well as a history of femoral venipuncture. Similarly, patients with cutaneous ulcers or cellulitis can develop SA either by direct spread to underlying

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TABLE 1 Risk factors for SA by population

Risk factors for:		
Adults	**Children**	**PJI patients**
Older age[a]	Male gender	Obesity
Diabetes	Low socioeconomic status	Diabetes mellitus
Rheumatoid arthritis	Osteomyelitis	Rheumatoid arthritis
Intravenous drug use	Juvenile idiopathic arthritis	Acquired or pharmacologic immunosuppression
Acquired or pharmacologic immunosuppression	Acquired or pharmacologic immunosuppression	Biologic disease-modifying antirheumatic drugs
Skin infection	Skin infection	Arthroplasty revision surgery
Penetrating trauma	Penetrating trauma	Prolonged procedure time
Arthroscopy procedures	Arthroscopy procedures	Smoking
		Bacteremia during previous yr
		Allogeneic blood transfusion

[a]Age of >60 and >80 years.

joints or via vascular channels. Arthrocentesis should therefore be avoided in patients with cellulitis overlying the joint so as to avoid the introduction of bacteria into the joint itself. In children, contiguous spread of bacteria into the joint from primary osteomyelitis can occur prior to ossification of the physeal plates (5, 13).

The absolute number of PJIs continues to increase as the number of arthroplasty (i.e., joint replacement) procedures performed rises, with reported incidences ranging from 0.5 to 3.3% in the United States (11). Prosthetic joints are at a high relative risk of infection, most commonly resulting from direct intraoperative contamination. This can lead to either acute or subacute infections weeks to months following implantation. Early-onset infections typically occur within 12 weeks of surgery and are caused by classically virulent pathogens such as *Staphylococcus aureus* or *Escherichia coli*. In contrast, delayed infections (presenting >3 months after arthroplasty) are caused by more indolent organisms, e.g., coagulase-negative staphylococci and *Cutibacterium* (formerly *Propionibacterium*) species. Pain may be the only symptom present among patients with delayed infection, and the more indolent organisms causing these infections do not necessarily elicit elevation of the biomarkers discussed below. Importantly, the prosthesis itself induces an altered physiology of the joint, leading to an increased risk of hematogenous seeding and subsequent development of infection. Late infections, occurring after 12 months, can be due to indolent organisms or can occur with an acute onset of symptoms after hematogenous seeding of the prosthetic joint with a more virulent organism (11).

TABLE 2 Microbiologic pathogens of SA and PJI

Commonly reported microorganisms[a] in:			
Adult SA	**Pediatric SA**	**PJI**	**Less frequently reported microorganisms**
Staphylococcus aureus	*Staphylococcus aureus*	Coagulase-negative staphylococci	*Neisseria* spp.
Streptococcus spp.[b]	*Kingella kingae*	*Staphylococcus aureus*	*Mycobacterium* spp.[c]
Enterococcus spp.	*S. pneumoniae*	*Streptococcus* spp.	Filamentous fungi
Escherichia coli	*Haemophilus influenzae*	*Enterococcus* spp.	>1 microbe
Salmonella spp.	*Escherichia coli*	*Cutibacterium* (*Propionibacterium*) spp.	*Nocardia asteroides*
Pseudomonas aeruginosa	*Salmonella* spp.	*Pseudomonas aeruginosa*	*Brucella* spp.
Candida spp.		*Candida* spp.	*Pasteurella* spp.
			Proteus spp.
			Serratia spp.
			Anaerobic organisms
			Finegoldia magna
			Streptobacillus moniliformis

[a]This is not intended to be a comprehensive list.
[b]Including *S. pneumoniae*.
[c]*M. tuberculosis* and nontuberculous species.

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CLINICAL MANIFESTATIONS

Symptoms of SA include acute onset of a decreased range of joint motion, localized pain, tenderness, and swelling of the infected joint, commonly (though not always) associated with fever (3). However, neonates (<3 months old) may present with more subtle findings, e.g., irritability with passive joint motion or features of septicemia without fever (14). The absence of fever does not rule out a diagnosis of SA, with only 30 to 60% of cases of culture-confirmed native-joint SA having fever upon initial presentation (15, 16). SA of the knee and hip occurs more commonly in older children and adults, whereas hip and shoulder joints are more frequently affected in neonates (14, 16). Only one joint is affected (i.e., monoarticular) in 80 to 90% of cases (4). For patients with PJI, the most common symptom is pain in the affected joint, followed by fever, effusion, swelling, and drainage from the surgical wound. However, pain may be the only symptom present in patients with delayed or late PJI (11). Pain is also the most common symptom of aseptic failure of the prosthetic joint; therefore, other symptoms and risk factors must be taken into account for diagnosis (11, 17). The severity of symptoms is often impacted by the pyogenic nature of the infecting organism.

The diagnosis of SA is challenging because signs and symptoms overlap those of other joint diseases, e.g., osteoarthritis, gout, rheumatoid arthritis, and juvenile rheumatoid arthritis, diseases that themselves can increase the risk of SA. The possibility of infection should always be considered in patients with a history of chronic joint disease who develop acute symptoms (3). Unfortunately, there is a dearth of diagnostic algorithms to guide clinicians in the diagnosis of native-joint SA, with the exception of the "Kocher criteria" for pediatric hip SA (18). Algorithms for PJI diagnosis and management have been developed by the Infectious Diseases Society of America (IDSA) and the International Consensus Meeting on PJI of the Musculoskeletal Infection Society (MSIS) (11, 17, 19). Despite this, the diagnosis of PJI continues to be challenging.

DIAGNOSTICS

Imaging. Imaging techniques may not reliably distinguish SA from other inflammatory joint conditions, with a reported sensitivity of 30% for radiologic findings suggestive of SA (20). Ultrasonography can be helpful in confirming the presence of a joint effusion and can, in turn, be used to guide arthrocentesis (5). It is the recommended imaging method for pediatric SA, but magnetic resonance imaging (MRI) may be warranted to rule out adjacent osteomyelitis (21). The IDSA recommends a plain radiograph of the affected joint for all cases of suspected PJI, although other imaging modalities such as MRI are not recommended for routine use (17). The utility of additional imaging methods (e.g., scintigraphy) in the diagnosis of PJI are beyond the scope of this article, but for further information, see references 11 and 19.

Laboratory studies. (i) Peripheral blood tests. Blood cultures have been shown to detect the etiologic agent of infection in up to 9 to 11% of SA patients with negative synovial fluid (SF) cultures (15). The IDSA also recommends blood cultures for patients with suspected PJI if the patient presents with fever or acute onset of symptoms or if bloodstream infection is likely (17).

An elevated peripheral white blood cell (WBC) count is one of the four parameters used for clinical prediction of SA in children. WBC counts of $>12,000/mm^3$, combined with four or five other parameters, have a high positive predictive value for SA (93%) (18, 22). However, an elevated WBC by itself was not shown to be an independent predictor of joint infection (22). The sensitivity of elevated serum WBCs in the diagnosis of SA in adults has been analyzed in multiple studies and ranges from 23 to 75% (3).

The serum erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) level are systemic acute-phase reactants that can be elevated in SA. Nevertheless, these can also be elevated in noninfectious inflammatory conditions. Thus, despite a sensitivity of 76 to 97% for an ESR of >30 mm/h, the associated specificity is only 29% for SA in adults. Similarly, CRP values of >100 mg/liter have a reported sensitivity of 82 to 83% for SA but specificities ranging from 27 to 70% (2). CRP levels of >10 mg/liter and ESRs of >30 mm/h have a reported high combined sensitivity up to 96% for acute PJI (11).

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Important caveats in test interpretation are that surgery itself can cause ESR and CRP elevation and that infections with indolent organisms may not cause such elevations (11).

Procalcitonin (PCT) is a serum marker that shows promise in the diagnosis of SA. Serum PCT levels are normally $<$0.05 ng/ml and can rise rapidly following systemic bacterial infections. However, in contrast to CRP and ESR, PCT levels typically remain low for most (though not all) systemic inflammatory disorders. A recent meta-analysis showed a sensitivity of 46% for a serum PCT of $>$0.5 ng/ml for bone and joint infections but a specificity of 90%. An improved sensitivity of 90% was observed when a cutoff of $>$0.2 to 0.3 ng/ml was used (23). Despite an abundance of studies on the utility of serum markers in the diagnosis of SA (excellently reviewed in reference 2), no one marker is by itself predictive of SA. Rather, results should be considered along with the clinical history and physical exam to determine whether arthrocentesis of the suspected joint is warranted, though laboratory test results should not delay joint aspiration.

(ii) SF analyses. The importance of arthrocentesis for the timely and accurate diagnosis of septic joints cannot be overstated (3). Culture of SF continues to be the gold standard for the diagnosis of SA, and ideally, joint aspiration should be performed prior to the initiation of antimicrobials. Patients with inflammatory diseases or immunosuppression and the elderly can have elevated levels of inflammatory markers in both serum and SF, and thus, the sensitivities and specificities described below should be interpreted with caution in these patient populations. Importantly, the presence of crystals in SF does not exclude infection (24).

(iii) WBC count. The most basic test is gross inspection, in which the clarity, color, and viscosity of the SF are examined. While purulent fluid may increase the suspicion of a septic joint, it is not considered to be of high diagnostic value, as turbidity can occur with a variety of pathologies (5). The SF WBC count has long been used to distinguish SA from other inflammatory and noninflammatory conditions. An SF WBC count of $\geq$64,000/μl was recently shown by Borzio et al. to have a sensitivity of 90% for the diagnosis of SA, with a likelihood ratio (LR) of 2.8 (95% confidence interval [CI], 1.2 to 6.7). However, the corresponding specificity was only 40% (25). In an earlier analysis, an SF WBC count of $\geq$100,000/μl had an LR of 28.0 (95% CI, 12.0 to 66.0). Furthermore, the LR for SA was 3.4 (95% CI, 2.8 to 4.2) when $\geq$90% of the total SF WBCs were neutrophils (3). For suspected PJI cases, an SF WBC count of $>$10,000/μl (or $>$3,000/μl in chronic cases) and an elevated synovial neutrophil percentage are considered minor criteria for the diagnosis of PJI by the MSIS criteria (19). The MSIS minor criteria also include a "++" reading when SF is tested with a leukocyte esterase test strip. A recent meta-analysis of this testing modality for PJI showed a high pooled sensitivity of 81% and a specificity of 97%, though a number of individual studies have shown variable performance and these test strips are not cleared for use with SF (26).

(iv) Immunology/chemistry. The utility of cytokine detection directly in SF to rapidly detect joint infection has been investigated, particularly for PJI. CRP and PCT levels in SF appear to have lower sensitivity and specificity for PJI diagnosis than those in serum (27). Several potential biomarkers have been evaluated (e.g., tumor necrosis factor α), all of which can be elevated in response to bacterial infection. The most promising of these appear to be interleukin-6 (IL-6) and α-defensin. In a meta-analysis, SF IL-6 showed a pooled sensitivity and specificity for PJI diagnosis of 91% (95% CI, 82 to 96%) and 90% (95% CI, 84 to 95%), respectively. No significant changes in these results occurred when patients with inflammatory diseases were included, though there was significant heterogeneity within the patient populations examined, and cutoff values still need to be determined for clinical use (28). A point-of-care α-defensin visual immunochromatographic assay is commercially available and showed intraoperative sensitivity and specificity of $>$95% for PJI diagnosis in a recent prospective multicenter study compared to modified MSIS criteria (29). Earlier evaluations demonstrated that median α-defensin levels do not appear to be dissimilar for PJI caused by

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both classically virulent and more indolent organisms (27). It is important to note that point-of-care testing needs per-institution validation by trained laboratory professionals. In addition, this test was neither developed nor validated for the diagnosis of native-joint SA.

While tests that detect cytokines in SF may ultimately prove useful in the diagnosis of SA, they are not yet widely available in most clinical labs and few studies have looked at their utility in native-joint SA diagnosis. However, it is possible that such tests may ultimately be included in future diagnostic algorithms for PJI. Preliminary evidence suggests that SF glucose and lactate may have a role in the rapid detection of native-joint infection. In a recent prospective study, analysis of SF with a glucometer showed promising results as an inexpensive test to rule out SA in an emergent setting, though further studies are needed, particularly in regard to less pyogenic pathogens (30). As with leukocyte esterase test strips, a major concern is the use of this test without appropriate validation.

Microbiologic analyses. (i) Gram stain and culture. Though cheap, rapid, and simple to perform, the sensitivity of an SF Gram stain for the diagnosis of SA is relatively low (29 to 65%) (2, 3). The sensitivity of Gram staining for PJI is believed to be even lower (0 to 27%), and consequently, it is not recommended for patients with suspected delayed PJI (11). Culture of material from the infected joint remains a critical part of the diagnostic workup, as isolation of the microorganism in culture continues to be a prerequisite for antimicrobial susceptibility testing.

Routine SF culture has a sensitivity and specificity of 70 to 90% and 75 to 95%, respectively (4, 11). Prior treatment with antimicrobials, as well as the culture media and the incubation time used, can affect the sensitivity of culture. It is recommended that SF cultures be incubated for at least 4 days, with a mean time to culture positivity of 36.65 ± 27.13 h in high-risk patients (31). Several studies have shown improved sensitivity of culture when SF is inoculated into blood culture bottles. Inoculation of SF into Bactec Peds Plus/F bottles led to the isolation of significantly more pathogens than conventional culture (both agar plates and thioglycolate broth), 62 versus 51 ($P = 0.001$), respectively (32). Of note, not all blood culture system manufacturers have media that are FDA cleared for use with body fluids, and thus, the use of blood culture bottles would constitute an off-label use.

There are important differences in the culture methods used for prosthetic joint specimens and those used for SF. Specifically, the IDSA recommends the collection of a minimum of three and optimally five or six periprosthetic tissue (PPT) samples for both aerobic and anaerobic cultures at the time of revision surgery. Ideally, each specimen should be obtained with a new sterile instrument. Preoperative antibiotics are typically withheld in cases of revision arthroplasty, but despite this practice, the sensitivity of PPT culture has been determined to be 63% when three specimens are positive for the same organism (11). The MSIS recommends that two or more positive cultures can be used as sufficient evidence of PJI. Additionally, several studies have attempted to improve the sensitivity of PPT culture for the diagnosis of PJI. In a recent large prospective cohort study, Peel et al. observed a sensitivity of 92% (95% CI, 85 to 97%) when tissue homogenates were inoculated into aerobic and anaerobic blood culture bottles versus 63% (95% CI, 52 to 73%) for conventional agar and broth cultures (33). Improved sensitivity of SF inoculation into blood culture bottles over conventional SF culture has also been observed in patients following arthroplasty (34). A recent prospective multicenter study by Bémer and colleagues determined that four PPT samples were required when three culture media were used (aerobic blood culture bottle, Schaedler broth [anaerobic], and chocolate agar plate incubated in CO_2) (35). The optimal incubation period for prosthetic joint specimens remains controversial, with some studies showing that a 13-day incubation period for both aerobic and anaerobic cultures is required for optimal recovery of *Cutibacterium* (*Propionibacterium*) species, while other studies have not found this to be the case. However, extended culture incubation appears to be required even when blood culture bottles are used for

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PPT samples (33). The optimal culture incubation and media for recovery of *Cutibacterium* species remain to be defined.

(ii) Explant cultures. It is recommended that the joint explant and associated hardware be sent for culture (17). Because bacteria commonly form biofilms on prosthetic material, sonication has emerged as a method to effectively dislodge the infecting bacteria. Sonication culture has shown improved sensitivity over standard PPT culture for a variety of joints (36). Initial studies used plastic bags for sonication, but this practice was associated with excess levels of contamination and sonication should instead be performed in a sterile container. The optimal cutoff value for sonicate cultures appears to be ≥5 CFU (36). Studies have indicated that the sensitivity of sonication culture may be further increased when sonicate fluid is inoculated into blood culture bottles. The use of BacT/Alert blood culture bottles had a sensitivity of 100% (95% CI, 91 to 100%) in a recent prospective study by Portillo and colleagues. In contrast, the sensitivity of sonicate fluid culture using agar medium alone was only 87% (95% CI, 73 to 96%) while PPT culture had a sensitivity of only 59% (95% CI, 42 to 74%) (37). Despite the reported improved sensitivity of sonication over PPT, the method may be less desirable to laboratories because of its labor-intensive and technically involved processing. Dislodgement of biofilm-associated bacteria by other methods, including vortexing, has also been investigated. Alternative methods reported in the literature include bead mill processing and pretreatment with dithiothreitol. These methods appear promising but require further study.

(iii) Challenges of interpreting culture results for PJI. Isolation of a classically virulent microorganism from a single specimen is a diagnostic criterion for PJI. In contrast, more typically indolent microorganisms should be isolated from more than one PPT sample. However, laboratories have no way to ascertain whether the surgeon used separate instruments to obtain each PPT specimen submitted to the laboratory. Furthermore, standard skin preparation methods, as well as preoperative antibiotics, do not eradicate *Cutibacterium acnes* and this too can complicate the interpretation of culture results.

(iv) Histopathologic analysis. The presence of more than five neutrophils per high-power field (HPF) by histologic analysis of PPT collected during revision surgery constitutes another minor criterion for PJI diagnosis. These tissues are often evaluated intraoperatively by frozen section, and a recent meta-analysis determined that intraoperative frozen section of PPT with five or more neutrophils per HPF showed a positive likelihood of 10.3 (95% CI, 6.3 to 16.6) of diagnosing culture-positive PJI (11, 17, 19, 38). Less virulent organisms remain a challenge, as they may not elicit a robust acute inflammatory response (11).

Molecular diagnostics. Over the past several decades, the increasing availability of molecular assays for microbiologic diagnosis has revolutionized the field of clinical microbiology. Importantly, detection of microbial DNA in a clinical specimen does not distinguish between live and nonviable bacteria. However, it is possible that in the future viable microorganisms will be more easily distinguishable. For example, the addition of propidium monazide prior to the tissue lysis step does not penetrate intact cytoplasmic membranes but only inhibits amplification of DNA from dead organisms (39).

(i) MALDI-TOF MS. Matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) has revolutionized microbial identification by virtue of its rapid turnaround time and high accuracy. However, MALDI-TOF MS lacks sufficient sensitivity (6.3%) for microbial identification directly in SF (40). However, it is capable of identifying microorganisms directly in blood culture bottles inoculated with SF (40).

(ii) Broad-range PCR. A number of studies have looked at the utility of "broad-range" or "universal" PCR, followed by DNA sequencing, for the diagnosis of PJI. Broad-range PCR is, in theory, capable of detecting any bacterial or fungal DNA present in a clinical specimen because it targets conserved ribosomal sequences: 16S rRNA genes in bacteria and 18S and 28S rDNA, internal transcribed spacer 1 (ITS1), and ITS2 in fungi. Despite this, broad-range PCR appears to be less sensitive than culture. In one prospective multicenter study 16S rRNA gene PCR had a sensitivity of 73.3% (95% CI,

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0.66 to 0.79%) and a specificity of 95.5% (95% CI, 84.5 to 99.4%) compared to PPT culture in patients undergoing revision arthroplasty for suspected PJI (41). These findings are consistent with earlier studies, though a meta-analysis by Qu et al. reported a higher pooled sensitivity of 86% (95% CI, 0.77 to 0.92%) (42). Rather, it appears that the main utility of broad-range PCR may be in patients with suspected but culture-negative osteoarticular infections, with 9% of 2,308 cases of culture-negative bone and joint specimens testing positive by broad-range PCR (43).

The diagnosis of joint infection by broad-range PCR coupled with electrospray ionization (ESI)-TOF MS has also been investigated. An earlier version of this technology (Plex-ID PCR-ESI/MS) showed better sensitivity than culture for microorganism detection in sonicate fluid specimens (77.6 versus 69.7%, respectively, $P = 0.01$), though poorer specificity was observed (93.5 versus 99.3%, respectively, $P = 0.0002$) (44). More recently, the IRIDICA platform showed a sensitivity (81 and 86%, respectively [$P = 0.56$]) and specificity (95 and 100%, respectively [$P = 0.045$]) comparable to those of routine culture for patients who met the clinical criteria for PJI in a retrospective study (45). As of April 2017, the manufacturer of the IRIDICA system (Abbott) had ceased producing both test kits and instrumentation; it remains to be seen whether similar technology will reenter the clinical diagnostics market in the future.

There is much interest in the fields of clinical microbiology and infectious disease diagnostics regarding the use of metagenomic sequencing to identify the etiology of infection. Street et al. recently applied metagenomic sequencing for bacterial molecular diagnosis directly to sonicate fluids from patients undergoing revision arthroplasty. These results were compared to those of aerobic and anaerobic cultures of both sonication fluid and PPT samples. The group observed an overall species level sensitivity of 88% (95% CI, 0.77 to 0.94%) and an overall clinically adjusted specificity of 88% (95% CI, 0.79 to 0.93%) when using optimal sequence thresholds. Importantly, contamination with both human host DNA and bacterial DNA was a notable challenge and the authors addressed the need for rigorous laboratory protocols when using this method (46).

(iii) Targeted PCR. Targeted PCR assays have demonstrated clinical utility in the detection of fastidious organisms that are difficult to culture. In particular, *Kingella kingae* has emerged as an important etiologic agent of osteoarticular infections in children under the age of 5 years. Traditional SF culture is known to miss infection with this bacterium. Although the sensitivity of culture for the detection of *K. kingae* can be improved by inoculation into blood culture bottles, the sensitivity remains far below that of targeted PCR. Yagupsky et al. reviewed multiple studies of skeletal system infections in children, including SA and osteomyelitis, that compared culture (conventional and blood culture vial) to various PCR methods and showed *K. kingae* to be positive by culture in 24.8% of the cases compared to 99.5% of the cases by PCR (13). Going forward, it is likely that targets for fastidious organisms will be increasingly incorporated into commercial PCR assays that can be marketed to community-based hospital microbiology labs.

(iv) Multiplex panels. The advantage of multiplex PCR panels is that they provide potential pathogen identification within hours and are relatively technically simple to perform. However, there are currently no FDA-cleared multiplex panels specifically designed for the diagnosis of joint infection in the United States. A number of studies have assessed the potential utility of assays designed for pathogen detection directly in whole blood (Table 3). Additionally, studies have investigated the performance of the Unyvero i60 implant and tissue infection system (Curetis, Holzgerlingen, Germany), a device that is CE marked in Europe for both PJI and SA (47, 48). In a recent multicenter study in France, the system had a concordance rate of 58.1% with culture and 70.1% with rRNA gene PCR testing of intraoperative (PPT, SF, biopsy, and bone) samples from patients suspected of having PJI (48). The sensitivity of the assay was lower among patients with lower numbers of organisms isolated by culture.

Several groups have investigated the off-label use of multiplex PCR panels developed for blood culture identification for rapid assessment of PJI. The BioFire FilmArray Blood Culture ID panel was shown to have a sensitivity of 53% compared to 69% for

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TABLE 3 Molecular assays used for bone and joint infections

Platform	Manufacturer	Technology	Sample type	Microorganisms identified	Approx TAT (h)[a]	FDA cleared for use in BJI	Reference(s)
Unyvero i60	Curetis	Cartridge multiplex PCR assay	Joint aspirate, sonication fluid	114 bacterial and fungal DNA targets and antibiotic resistance markers	5 (30)	No	47, 48
GeneXpert	Cepheid	Modular RT-PCR with fluorescent-probe-based detection in closed system	Skin and soft tissue infection swabs	Skin and soft tissue infection panel for MSSA, MRSA	1 (5)	No	50–52
Septifast	Roche	Multiplex RT-PCR and specific melting point analysis software	FDA cleared for blood	25 most common bacteria and fungi known to cause bloodstream infections	6	No	53
FilmArray	BioFire	Two-stage nested PCR with fluorescent-probe-based detection	Synovial fluid	BJI panel in development	1 (5)	No	54
IRIDICA/Plex-ID	Abbott	PCR with ESI-MS	Sterile fluids and tissues (IRIDICA BAC SFT Assay)	800 bacterial and fungal targets and 4 antimicrobial resistance markers	6	No	44, 45
Metagenomic sequencing	Research laboratory only					No	46

[a]TAT, turnaround time. Values in parentheses are hands-on times in minutes.

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sonicate fluid culture from patients with PJI diagnosed by the IDSA criteria ($P = 0.004$). However, the panel also detected six pathogens that did not grow in culture (17, 49). To date, there have been no published studies evaluating the potential utility of this panel on SFs or other nonsonicate specimens. While the performance of multiplex panels for the diagnosis of SA and PJI requires further study, the potential utility of this approach stems from the rapid diagnosis of the etiology of infection. However, determining the clinical significance of detecting more indolent causes of infection that also are members of the skin microbiota (e.g., coagulase-negative staphylococci) is likely to present a diagnostic challenge. Furthermore, culture will continue to be required for both antimicrobial susceptibility testing and the diagnosis of infections caused by microorganisms not targeted by these multiplex panels.

CONCLUSIONS

The diagnosis of SA and PJI is challenging. Physicians are often faced with a diagnostic conundrum when attempting to diagnose infection from both clinical presentation and laboratory testing standpoints. There is a clear need to rapidly and accurately detect the presence of microorganisms directly in SF, PPT, and sonicate specimens. While emerging cytokine/immunologic and molecular technologies have yet to be incorporated into routine clinical use, the rapid pace of research in this field, particularly with regard to PJI, is encouraging. Microbiologic culture, with methods optimized for SF and PJI specimens, continues to stand as the fundamental diagnostic tool for SA and PJI diagnosis. Going forward, assessing clinical outcomes among patients whose specimens are tested with these newer methodologies will be paramount.

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