A Review Paper

Systematic Review of Novel Synovial Fluid Markers and Polymerase Chain Reaction in the Diagnosis of Prosthetic Joint Infection

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Abstract

Prosthetic joint infection (PJI) may be underreported because of difficulty in making a diagnosis, especially in infections with low-virulence organisms. Reports of PJI cases misdiagnosed as aseptic loosening suggest that current screening and diagnostic tools are not sensitive enough to detect all infections and that PJI likely is underdiagnosed.

We reviewed the literature on recently developed novel synovial biomarkers and polymerase chain reaction (PCR) technologies, of which several have proved promising as highly sensitive and specific tools for detecting PJI. We followed the recommendations of PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses).

Of 90 papers screened by title or abstract and then by full text, 15 met our inclusion criteria. Sensitivities reported in the included studies ranged from 63% to 100% for α-defensin, from 46.8% to 90.9% for interleukin 6, from 28.6% to 100% for leukocyte esterase, and from 67.10% to 95.7% for PCR. Specificities ranged from 95% to 100% for α-defensin, from 85.7% to 97.6% for interleukin 6, from 63.6% to 96.5% for leukocyte esterase, and from 12.3% to 97.8% for PCR.

α-Defensin is a highly sensitive and specific screening tool that may help improve the accuracy of PJI detection, particularly in low-grade infections.

Take-Home Points

- Novel synovial markers and PCR have the potential to improve the detection of PJIs.
- Difficult-to-detect infections of prosthetic joints pose a diagnostic problem to surgeons and can lead to suboptimal outcomes.
- AD is a highly sensitive and specific synovial fluid marker for detecting PJIs.
- AD has shown promising results in detecting low virulence organisms.
- Studies are needed to determine how to best incorporate novel synovial markers and PCR to current diagnostic criteria in order to improve diagnostic accuracy.

Approximately 7 million Americans are living with a hip or knee replacement.[1] According to projections, primary hip arthroplasties will increase by 174% and knee arthroplasties by 673% by 2030. Revision arthroplasties are projected to increase by 137% for hips and 601% for knees during the same time period.[2] Infection and aseptic loosening are the most common causes of implant failure.[3] The literature shows that infection is the most common cause of failure within 2 years after surgery and that aseptic loosening is the most common cause for late revision.[3]

Recent studies suggest that prosthetic joint infection (PJI) may be underreported because of difficulty making a diagnosis and that cases of aseptic loosening may in fact be attributable to infections with low-virulence organisms.[2,3] These findings have led to new efforts to develop uniform criteria for diagnosing PJIs. In 2011, the Musculoskeletal Infection Society (MSIS) offered a new definition for PJI diagnosis, based on clinical and laboratory criteria, to

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increase the accuracy of PJI diagnosis.[4] The MSIS committee acknowledged that PJI may be present even if these criteria are not met, particularly in the case of low-virulence organisms, as patients may not present with clinical signs of infection and may have normal inflammatory markers and joint aspirates. Reports of PJI cases misdiagnosed as aseptic loosening suggest that current screening and diagnostic tools are not sensitive enough to detect all infections and that PJI is likely underdiagnosed.

According to MSIS criteria, the diagnosis of PJI can be made when there is a sinus tract communicating with the prosthesis, when a pathogen is isolated by culture from 2 or more separate tissue or fluid samples obtained from the affected prosthetic joint, or when 4 of 6 criteria are met. The 6 criteria are (1) elevated serum erythrocyte sedimentation rate (ESR) (>30 mm/hour) and elevated C-reactive protein (CRP) level (>10 mg/L); (2) elevated synovial white blood cell (WBC) count (1100-4000 cells/μL); (3) elevated synovial polymorphonuclear leukocytes (>64%); (4) purulence in affected joint; (5) isolation of a microorganism in a culture of periprosthetic tissue or fluid; and (6) more than 5 neutrophils per high-power field in 5 high-power fields observed.

In this review article, we discuss recently developed novel synovial biomarkers and polymerase chain reaction (PCR) technologies that may help increase the sensitivity and specificity of diagnostic guidelines for PJI.

Methods

Using PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses), we performed a systematic review of specific synovial fluid markers and PCR used in PJI diagnosis. In May 2016, we searched the PubMed database for these criteria: ((((((PCR[Text Word]) OR IL-6[Text Word]) OR leukocyte esterase[Text Word]) OR alpha defensin[Text Word]) AND (("infection/diagnosis"[MeSH Terms] OR "infection/surgery"[MeSH Terms])))) AND (prosthetic joint infection[MeSH Terms] OR periprosthetic joint infection[MeSH Terms]).

We included patients who had undergone total hip, knee, or shoulder arthroplasty (THA, TKA, TSA). Index tests were PCR and the synovial fluid markers α-defensin (AD), interleukin 6 (IL-6), and leukocyte esterase (LE). Reference tests included joint fluid/serum analysis or tissue analysis (ESR/CRP level, cell count, culture, frozen section), which defined the MSIS criteria for PJI. Primary outcomes of interest were sensitivity and specificity, and secondary outcomes of interest included positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (+LR), and negative likelihood ratio (–LR). Randomized controlled trials and controlled cohort studies in humans published within the past 10 years were included.

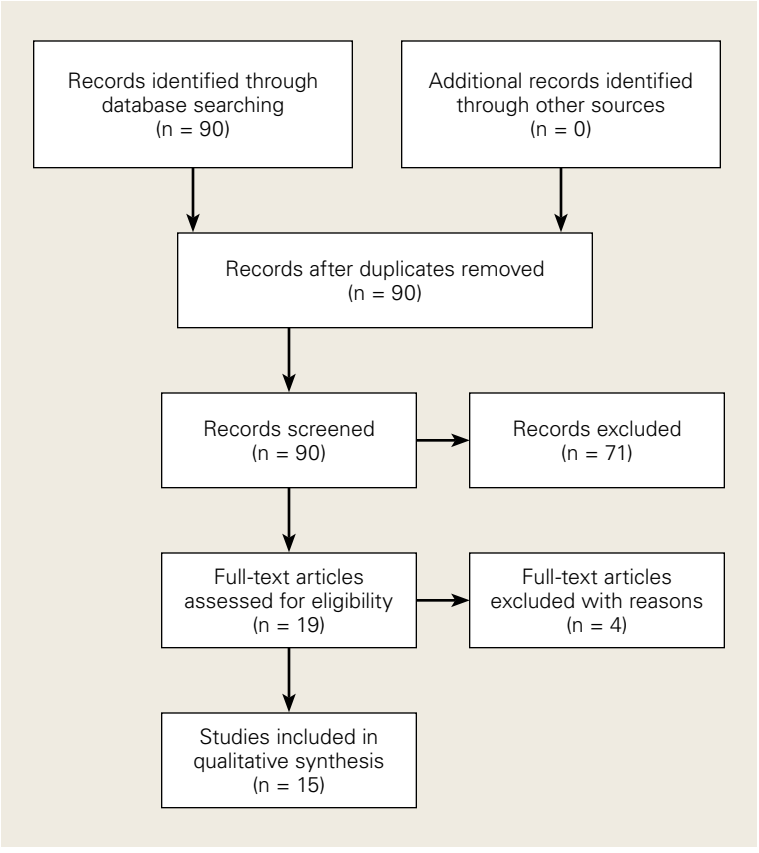

Figure 1. PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow diagram for literature selection.

Results

Our full-text review yielded 15 papers that met our study inclusion criteria (**Figure 1**).

α-Defensin

One of the novel synovial biomarkers that has shown significant promise in diagnosing PJIs, even with difficult-to-detect organisms, is AD. Frangiamore and colleagues[5] conducted a prospective study comparing patients with painful TSAs that required revision (n = 33). Patients were grouped based on objective clinical, laboratory, and histologic criteria of infection, which included preoperative clinical signs (swelling, sinus track, redness, drain-

age), elevated serum ESR or CRP, intraoperative gross findings (purulence, necrosis) and positive intraoperative frozen section. Synovial fluid aspiration was obtained preoperatively or intraoperatively. Of the 33 patients, 11 patients met the authors criteria for suspected PJI prior to final intraoperative culture results; 22 patients did not. Of the samples taken intraoperatively, *Propionibacterium acnes* was the most commonly isolated organism (9 cases), followed by coagulase-negative *Staphylococcus* (4 cases). AD demonstrated a sensitivity of 63%, specificity of 95%, +LR ratio of 12.1, and –LR ratio of 0.38. AD showed a strong association with growth of *P acnes* in the infected group (median signal-to-cutoff ratio, 4.45) compared with the noninfected group (median signal-to-cutoff ratio, 1.33) as well as strong associations with frozen section histology. Frangiamore and colleagues[5] concluded that the use of AD in diagnosing PJIs with difficult-to-detect organisms was promising.

AD has shown even more impressive results as a biomarker for PJI in the hip and knee, where infection with low virulence organism is less common. In 2014, Deirmengian and colleagues[6] conducted a prospective clinical study of 149 patients who underwent revision THA or TKA for aseptic loosening (n = 112) or PJI (n = 37) as defined by MSIS criteria. Aseptic loosening was diagnosed when there was no identifiable reason for pain, and MSIS criteria were not met. Synovial fluid aspirates were collected before or during surgery. AD correctly identified 143 of the 149 patients with confirmed infection with sensitivity of 97.3% (95% confidence interval [CI], 85.8%-99.6%) and specificity of 95.5% (95% CI, 89.9%-98.5%). Similarly, Bingham and colleagues[7] conducted a retrospective clinical study of 61 assays done on 57 patients who underwent revision arthroplasty for PJI as defined by MSIS criteria. Synovial fluid aspirates were collected before or during surgery. AD correctly identified all 19 PJIs with sensitivity of 100% (95% CI, 79%-100%) and specificity of 95% (95% CI, 83%-99%). Sensitivity and specificity of the AD assay more accurately predicted infection than synovial cell count or serum ESR/CRP level did.

These results are supported by another prospective study by Deirmengian and colleagues[8] differentiating aseptic failures and PJIs in THA or TKA. The sensitivity and specificity of AD in diagnosing PJI were 100% (95% CI, 85.05%-100%). Synovial fluid was collected from 46 patients before and during surgery: 23 with PJI and 23 with aseptic failure as defined by MSIS criteria. All patients were tested for AD or LE. Of the 23 PJI cases, 18 were associated with a positive culture, with the most common organism being *Staphylococcus epidermidis* (n = 6). AD correctly diagnosed 100% of PJIs, whereas LE correctly diagnosed only 78%; the difference was statistically significant ($P < 0.001$).

In a prospective study of 102 patients who underwent revision THA or TKA secondary to aseptic loosening or PJI, Frangiamore and colleagues[9] also demonstrated the value of AD as a diagnostic for PJI in primary and revision hip and knee arthroplasty. Based on MSIS criteria, 54 cases were classified as non-infected first-stage revision, 24 as infected first-stage revision, 35 as non-infected second-stage revision, and 3 as infected second-stage revision. For patients with first-stage revision THA or TKA, AD had sensitivity of 100% (95% CI, 86%-100%), specificity of 98% (95% CI, 90%-100%), PPV of 96% (95% CI, 80%-99%), and NPV of 100% (95% CI, 93%-100%). +LR was 54 (95% CI, 8-376), and –LR was 0. When combining all patients, AD outperformed serum ESR and CRP and synovial cell count as a biomarker for predicting PJI.

Table 1 and **Figure 2** provide a concise review of the findings of each study.

Interleukin 6

Another synovial fluid biomarker that has shown promise in PJI diagnosis is IL-6. In 2015, Frangiamore and colleagues[10] conducted a prospective clinical study of 32 patients who underwent revision TSA. Synovial fluid aspiration was obtained before or during surgery. MSIS criteria were used to establish the diagnosis of PJI. IL-6 had sensitivity of 87% and specificity of 90%, with +LR of 8.45 and –LR of 0.15 in predicting PJI. Synovial fluid IL-6 had strong associations with frozen section histology and growth of *P acnes*. Frangiamore and colleagues[10] recommended an ideal IL-6 cutoff of 359.1 pg/mL and reported that, though not as accurate as AD, synovial fluid IL-6 levels can help predict positive cultures in patients who undergo revision TSA.

Lenski and Scherer[11] conducted another retrospective clinical study of the diagnostic value of IL-6 in PJI. Revision total joint arthroplasty (TJA) was performed for aseptic loosening (38 patients) or PJI (31 patients) based on criteria modeled after MSIS criteria. All joints were aspirated for synovial fluid IL-6, synovial fluid lactate dehydrogenase, synovial fluid glucose, synovial fluid lactate, synovial

Table 1. **Summary of α-Defensin Articles That Met Our Inclusion Criteria**

Authors	Year	Country	Journal	Study Type	Patients, N	Sensitivity, %	Specificity, %
Frangiamore et al[5]	2015	US	*JSES*	Prospective	33	63	95
Deirmengian et al[6]	2014	US	*JBJSA*	Prospective	149	97.3	95.5
Bingham et al[7]	2014	US	*CORR*	Retrospective	57	100	95
Deirmengian et al[8]	2015	US	*CORR*	Prospective	46	100	100
Frangiamore et al[9]	2016	US	*JOA*	Prospective	102	100	98

Abbreviations: *CORR, Clinical Orthopaedics and Related Research*; *JBJSA, Journal of Bone and Joint Surgery, American volume*; *JOA, Journal of Arthroplasty*; *JSES, Journal of Shoulder and Elbow Surgery.*

fluid WBCs, and serum CRP. IL-6 had sensitivity of 90.9%, specificity of 94.7%, +LR of 17.27, and –LR of 0.10. An optimal IL-6 cutoff value of 30,750 pg/mL was determined.

Randau and colleagues[12] conducted a prospective clinical study of 120 patients who presented with painful THA or TKA and underwent revision for PJI, aseptic failure, or aseptic revision without signs of infection or loosening. Synovial fluid aspirate was collected before or during surgery. PJI was diagnosed with the modified MSIS criteria. IL-6 sensitivity and specificity depended on the cutoff value. A cutoff of >2100 pg/mL yielded sensitivity of 62.5% (95% CI, 43.69%-78.9%) and specificity of 85.71% (95% CI, 71.46%-94.57%), and a cutoff of >9000 pg/mL yielded sensitivity of 46.9% (95% CI, 29.09%-65.26%) and specificity of 97.62% (95% CI, 87.43%-99.94%). The authors concluded that synovial IL-6 is a more accurate marker than synovial WBC count.

Table 2 and **Figure 3** provide a concise review of the findings of each study.

Leukocyte Esterase

LE strips are an inexpensive screening tool for PJI, according to some studies. In a prospective clinical study of 364 endoprosthetic joint (hip, knee, shoulder) interventions, Guenther and colleagues[13] collected synovial fluid before surgery. Samples were tested with graded LE strips using PJI criteria set by the authors. Results were correlated with preoperative synovial fluid aspirations, serum CRP level, serum WBC count, and intraoperative histopathologic and microbiological findings. Whereas 293 (93.31%) of the 314 aseptic cases had negative test strip readings, 100% of the 50 infected cases were positive. LE had sensitivity of 100%, specificity of 96.5%, PPV of 82%, and NPV of 100%.

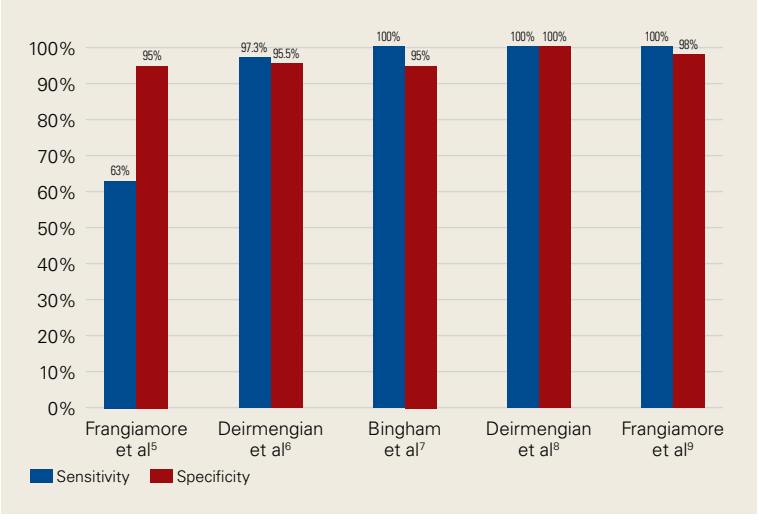

Figure 2. Summary of α-defensin primary outcomes of interest.

Wetters et al[14] performed a prospective clinical study on 223 patients who underwent TKAs and THAs for suspected PJI based on having criteria defined by the authors of the study. Synovial fluid samples were collected either preoperatively or intraoperatively. Using a synovial fluid WBC >3k WBC per microliter, the sensitivity, specificity, PPV, and NPV were 92.9%, 88.8%, 75%, and 97.2%, respectively. Using positive cultures or the presence of a draining sinus tract, the sensitivity, specificity, PPV, and NPV were 93.3%, 77%, 37.8%, and 98.7%, respectively. Of note, the most common organism found at the time of revision for infection was coagulase-negative *Staphylococcus* (6 out of 39).

Other authors have reported different findings that LE is an unreliable marker in PJI diagnosis. In one prospective clinical study of 85 patients who underwent primary or revision TSA, synovial fluid was collected during surgery.[15] According to

Table 2. **Summary of Interleukin 6 Articles That Met Our Inclusion Criteria**

Authors	Year	Country	Journal	Study Type	Patients, N	Sensitivity, %	Specificity, %
Frangiamore et al[10]	2015	US	*JBJSA*	Prospective	32	87,[a] 86[b]	90,[a] 95[b]
Lenski & Scherer[11]	2014	US	*JOA*	Retrospective	69	90.9	94.7
Randau et al[12]	2014	Germany	*PLoS One*	Prospective	120	62.5,[c] 46.9[d]	85.7,[c] 97.6[d]

[a]These are the results of interleukin 6 (IL-6) compared to the authors' definition of prosthetic joint infection (PJI). [b]These are the results of IL-6 compared to the definition of PJI by the Musculoskeletal Infection Society. [c]Cutoff, >2100 pg/mL. [d]Cutoff, >9000 pg/mL.
Abbreviations: *JBJSA*, *Journal of Bone and Joint Surgery, American volume*; *JOA*, *Journal of Arthroplasty*; *PLoS*, *Public Library of Science.*

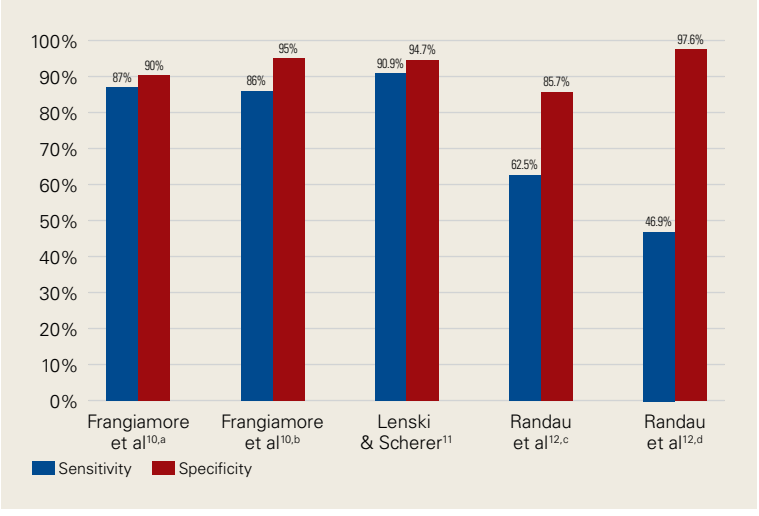

Figure 3. Summary of interleukin 6 (IL-6) primary outcomes of interest.
[a]These are the results of IL-6 compared to the authors' definition of prosthetic joint infection (PJI). [b]These are the results of IL-6 compared to the definition of PJI by the Musculoskeletal Infection Society. [c]Cutoff, >2100 pg/mL. [d]Cutoff, >9000 pg/mL.

MSIS criteria, only 5 positive LE results predicted PJI among 21 primary and revision patients with positive cultures. Of the 7 revision patients who met the MSIS criteria for PJI, only 2 had a positive LE test. LE had sensitivity of 28.6%, specificity of 63.6%, PPV of 28.6%, and NPV of 87.5%. Six of the 7 revision patients grew *P acnes*. These results showed that LE was unreliable in detecting shoulder PJI.[15]

In another prospective clinical study, Tischler and colleagues[16] enrolled 189 patients who underwent revision TKA or THA for aseptic failure or PJI as defined by the MSIS criteria. Synovial fluid was collected intraoperatively. Fifteen of the 52 patients with a MSIS defined PJI had positive cultures with the most common organism being coagulase-negative *Staphylococcus* (7). Two thresholds were used to consider a positive LE test. When using the first threshold that had a lower acceptance level for positivity, the sensitivity, specificity, PPV, and NPV were 79.2% (95% CI, 65.9%-89.2%), 80.8 (95% CI, 73.3%-87.1%), 61.8% (95% CI, 49.2%-73.3%), and 90.1% (95% CI, 84.3%-95.4%), respectively. When using the higher threshold, the sensitivity, specificity, PPV, and NPV were 66% (95% CI, 51.7%-78.5%), 97.1% (95% CI, 92.6%-99.2%), 89.7% (95% CI, 75.8%-97.1%), and 88% (95% CI, 81.7%-92.7%), respectively. Once again, these results were in line with LE not being a reliable marker in diagnosing PJI.

Table 3 and **Figure 4** provide a concise review of the findings of each study.

Polymerase Chain Reaction

Studies have found that PCR analysis of synovial fluid is effective in detecting bacteria on the surface of implants removed during revision arthroplasties. Comparison of the 16S rRNA gene sequences of bacterial genomes showed a diverse range of bacterial species within biofilms on the surface of clinical and subclinical infections.[17] These findings, along with those of other studies, suggest that PCR analysis of synovial fluid is useful in diagnosing PJI and identifying organisms and their sensitivities to antibiotics.

Gallo and colleagues[18] performed a prospective clinical study on 115 patients who underwent revision TKAs or THAs. Synovial fluid was collected intraoperatively. PCR assays targeting the 16S rDNA were carried out on 101 patients. PJIs were classified based on criteria of the authors of this study, of which there were 42. The sensitivity, specificity, PPV, NPV, +LR, and -LR for PCR were 71.4% (95% CI, 61.5%-75.5%), 97% (95% CI, 91.7%-99.1%), 92.6% (95% CI, 79.8%-97.9%), 86.5% (95% CI, 81.8%-88.4%), 23.6 (95% CI, 5.9%-93.8%), and 0.29 (95% CI, 0.17%-0.49%), respectively. Of note the most common organism detected in 42 PJIs was coagulase-negative *Staphylococcus*.

Marin and colleagues[19] conducted a prospective study of 122 patients who underwent arthroplasty for suspected infection or aseptic loosening as defined by the authors' clinicohistopathologic

Table 3. **Summary of Leukocyte Esterase Articles That Met Our Inclusion Criteria**

Authors	Year	Country	Journal	Study Type	Patients,[a] N	Sensitivity, %	Specificity, %
Guenther et al[13]	2014	Germany	*IO*	Prospective	364	100	96.5
Wetters et al[14]	2012	US	*JOA*	Prospective	223	92.9,[b] 93.3[c]	88.8,[b] 77[c]
Nelson et al[15]	2015	US	*JSES*	Prospective	85	28.6	63.6
Tischler et al[16]	2014	US	*JBJSA*	Prospective	189	79.2	80.8

[a]There were no controls in these studies. [b]Compared with white blood cell count. [c]Compared with positive cultures or present sinus tract.
Abbreviations: *IO, International Orthopedics*; *JBJSA, Journal of Bone and Joint Surgery, American volume*; *JOA, Journal of Arthroplasty*; *JSES, Journal of Shoulder and Elbow Surgery.*

criteria. Synovial fluid and biopsy specimens were collected during surgery, and 40 patients met the infection criteria. The authors concluded that 16S PCR is more specific and has better PPV than culture does as one positive 16S PCR resulted in a specificity and PPV of PJI of 96.3% and 91.7%, respectively. However, they noted that culture was more sensitive in diagnosing PJI.

Jacovides and colleagues[20] conducted a prospective study on 82 patients undergoing primary TKA, revision TKA, and revision THA. The synovial fluid aspirate was collected intraoperatively. PJI was diagnosed based on study specific criteria, which was a combination of clinical suspicion and standard laboratory tests (ESR, CRP, cell count and tissue culture). Using the study's criteria, PJI was diagnosed in 23 samples, and 57 samples were diagnosed as uninfected. When 1 or more species were present, the PCR-Electrospray Ionization Mass Spectrometry (PCR-ESI/MS) yielded a sensitivity, specificity, PPV, and NPV value of 95.7%, 12.3%, 30.6%, and 87.5%, respectively.

The low PCR sensitivities reported in the literature were explained in a review by Hartley and Harris.[21] They wrote that BR 16S rDNA and sequencing of PJI samples inherently have low sensitivity because of the contamination that can occur from the PCR reagents themselves or from sample mishandling. Techniques that address contaminant (extraneous DNA) removal, such as ultraviolet irradiation and DNase treatment, reduce Taq DNA polymerase activity, which reduces PCR sensitivity. The simplest way to avoid the effects of "low-level contaminants" is to decrease the number of PCR cycles, which also reduces sensitivity. However, loss of contaminants has resulted in increased specificities in studies that have used BR 16S rDNA PCR. The authors also stated that, when PCR incorporates cloning and sequencing, mass spectroscopic detection, or species-specific PCR, sensitivity is higher with increased contamination.

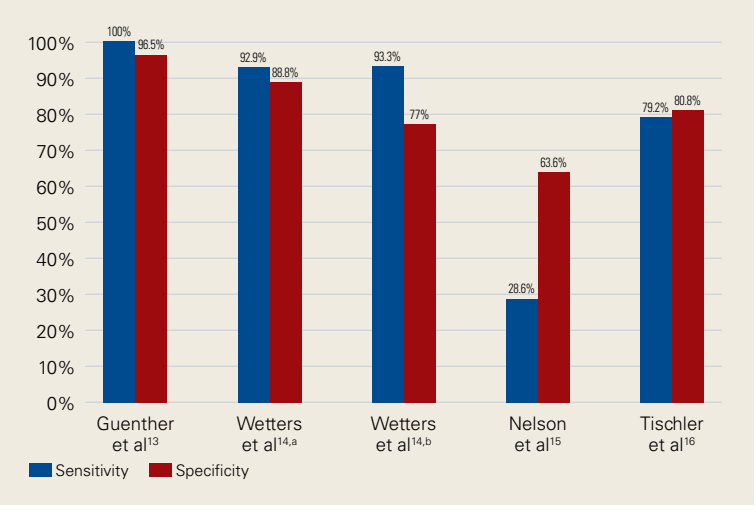

Figure 4. Summary of leukocyte esterase (LE) primary outcomes of interest.
[a]These are the results of LE compared to over 3,000 white blood cells per microliter as a marker of infection. [b]These are the results of interleukin 6 compared to positive cultures or the presence of a draining sinus tract.

Table 4 and **Figure 5** provide a concise review of the findings of each study.

Discussion

Although there is no gold standard for the diagnosis of PJIs, several clinical and laboratory criteria guidelines are currently used to help clinicians diagnose infections of prosthetic joints. However, despite standardization of diagnostic criteria, PJI continue to be a diagnostic challenge. Diagnosing PJI has been difficult for several reasons, including lack of highly sensitive and specific clinical findings and laboratory tests, as well as difficulty in culturing organisms, particularly fastidious organisms. More effective diagnostic tools are needed to avoid failing to accurately detect infections which lead to poor outcomes in patients who undergo TJA. Moreover, PJIs with low-virulence organisms

Table 4. **Summary of Polymerase Chain Reaction Articles That Met Our Inclusion Criteria**

Authors	Year	Country	Journal	Study Type	Patients,[a] N	Sensitivity, %	Specificity, %
Gallo et al[18]	2008	Czech R	*NM*	Prospective	115	71.4	97
Marin et al[19]	2012	Spain	*JCM*	Prospective	122	67.1	97.8
Jacovides et al[20]	2012	US	*JBJSA*	Prospective	82	95.7	12.3

[a]There were no controls in these studies.
Abbreviations: Czech R, Czech Republic; *JBJSA*, *Journal of Bone and Joint Surgery, American volume*; *JCM*, *Journal of Clinical Microbiology*; *NM*, *New Microbiology*.

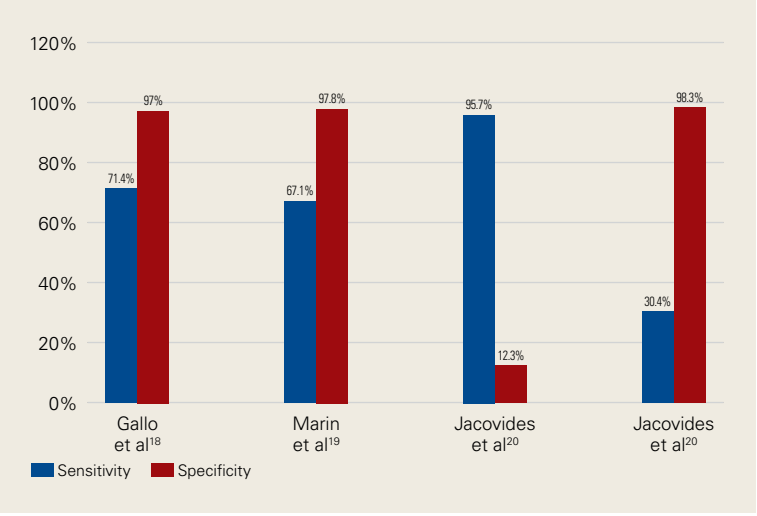

Figure 5. Summary of polymerase chain reaction primary outcomes of interest.

are especially troublesome, as they can present with normal serum inflammatory markers and negative synovial fluid analysis and cultures from joint aspiration.[22]

AD is a highly sensitive and specific synovial fluid biomarker in detecting common PJIs. AD has a higher sensitivity and specificity for detecting PJI, as compared to synovial fluid cell count, culture, ESR, and CRP.[15,16,19] Moreover, it has been shown that as many as 38% to 88% of patients diagnosed with aseptic loosening have PJIs with low-grade organisms,[23,24] such as Coagulase-negative *S acnes* and *P acnes*. Several studies reviewed in this article have demonstrated that AD can detect infections with these low virulence organisms. Our systematic review supports the claim that AD can potentially be used as a screening tool for PJI with common, as well as difficult-to-detect, organisms. Our findings also support the claim that novel synovial fluid biomarkers have the potential to become of significant diagnostic use and help improve the ability to diagnose PJIs when combined with current laboratory and clinical diagnostic criteria.

In summary, 5 AD studies[5-9] had sensitivity ranging from 63% to 100% and specificity ranging from 95% to 100%; 3 IL-6 studies[10-12] had sensitivity ranging from 46.8% to 90.9% and specificity ranging from 85.7% to 97.6%; 4 LE studies[13-16] had sensitivity ranging from 28.6% to 100% and specificity ranging from 63.6% to 96.5%; and 3 PCR studies[18-20] had sensitivity ranging from 67.1% to 95.7% and specificity ranging from 12.3% to 97.8%. Sensitivity and specificity were consistently higher for AD than for IL-6, LE, and PCR, though there was significant overlap, heterogeneity, and variation across all the included studies. Moreover, the outlier study with the lowest sensitivity for AD (63%) was in patients undergoing TSA, where *P acnes* infection is more common and has been reported to be more difficult to detect by standard diagnostic tools. **Tables 5, 6 and Figures 6, 7** provide the data for each of these studies.

Although the overall incidence of PJI is low, infected revisions remain a substantial financial burden to hospitals, as annual costs of infected revisions is estimated to exceed $1.62 billion by 2020.[25] The usefulness of novel biomarkers and PCR in diagnosing PJI can be found in their ability to diagnose infections and facilitate appropriate early treatment. Several of these tests are readily available commercially and have the potential to be cost-effective diagnostic tools. The price to perform an AD test from Synovasure™ (Zimmer Biomet) ranges from $93 to $143. LE also provides an economic option for diagnosing PJI, as LE strips are commercially available for the cost of about 25 cents. PCR has also become an economic option, as costs can average $15.50 per sample extraction or PCR assay and $42.50 per amplicon sequence as reported in a study by Vandercam and colleagues.[26] Future studies are needed to determine a diagnostic algorithm which incorporates these novel synovial markers to improve diagnostic accuracy of PJI in the most cost effective manner.

The current literature supports that AD can

Table 5. **Sensitivities of All Index Tests**

Index Test, Authors	Sensitivity, %
α-Defensin	
Frangiamore et al[5]	63
Deirmengian et al[6]	97.3
Bingham et al[7]	100
Deirmengian et al[8]	100
Frangiamore et al[9]	100
Interleukin 6	
Frangiamore et al[10]	87,[a] 86[b]
Lenski & Scherer[11]	90.9
Randau et al[12]	62.5,[c] 46.8[d]
Leukocyte esterase	
Guenther et al[13]	100
Wetters et al[14]	92.9,[e] 93.3[f]
Nelson et al[15]	28.6
Tischler et al[16]	79.2
Polymerase chain reaction	
Gallo et al[18]	71.4
Marin et al[19]	67.1
Jacovides et al[20]	95.7

[a]These are the results of interleukin 6 (IL-6) compared to the authors' definition of prosthetic joint infection (PJI). [b]These are the results of IL-6 compared to the definition of PJI by the Musculoskeletal Infection Society. [c]Cutoff, >2100 pg/mL. [d]Cutoff, >9000 pg/mL. [e]Compared with white blood cell count. [f]Compared with positive cultures or present sinus tract.

Table 6. **Specificities of All Index Tests**

Index Test, Authors	Specificity, %
α-Defensin	
Frangiamore et al[5]	95
Deirmengian et al[6]	95.5
Bingham et al[7]	95
Deirmengian et al[8]	100
Frangiamore et al[9]	98
Interleukin 6	
Frangiamore et al[10]	90,[a] 95[b]
Lenski & Scherer[11]	94.7
Randau et al[12]	85.7,[c] 97.6[d]
Leukocyte esterase	
Guenther et al[13]	96.5
Wetters et al[14]	88.8,[e] 77[f]
Nelson et al[15]	63.6
Tischler et al[16]	80.8
Polymerase chain reaction	
Gallo et al[18]	97
Marin et al[19]	97.8
Jacovides et al[20]	12.3

[a]These are the results of interleukin 6 (IL-6) compared to the authors' definition of prosthetic joint infection (PJI). [b]These are the results of IL-6 compared to the definition of PJI by the Musculoskeletal Infection Society. [c]Cutoff, >2100 pg/mL. [d]Cutoff, >9000 pg/mL. [e]Compared with white blood cell count. [f]Compared with positive cultures or present sinus tract.

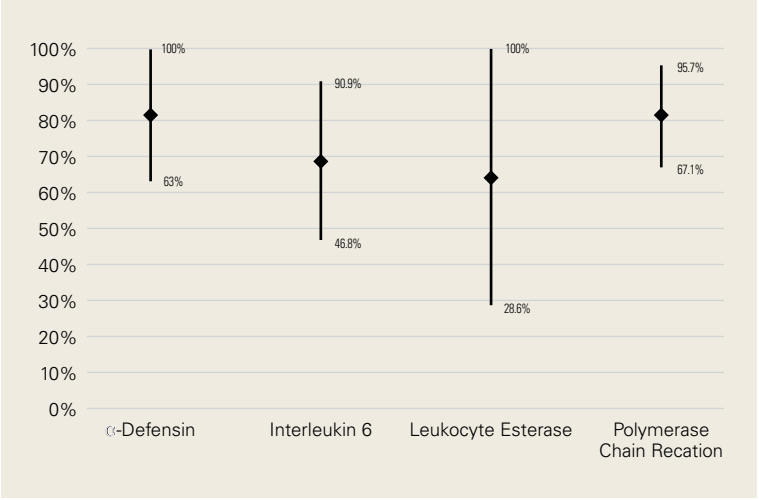

Figure 6. Sensitivity variation in diagnostic tests.

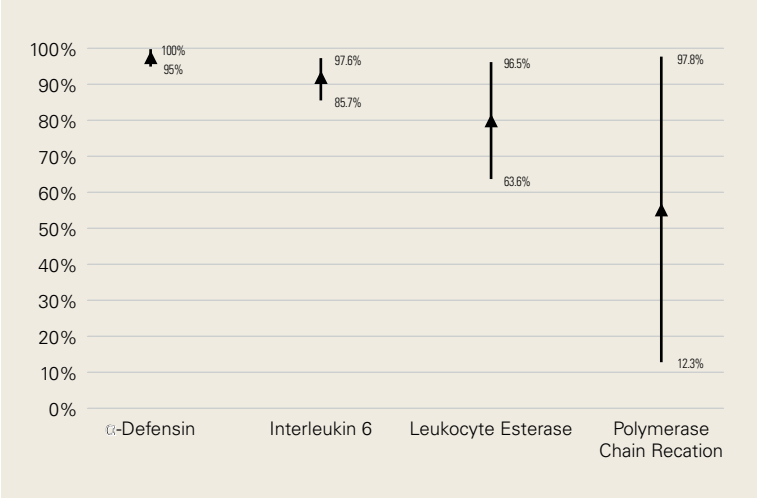

Figure 7. Specificity variation in diagnostic tests.

potentially be used to screen for PJI. Our findings suggest novel synovial fluid biomarkers may become of significant diagnostic use when combined with current laboratory and clinical diagnostic criteria. We recommend use of AD in cases in which pain, stiffness, and poor TJA outcome cannot be explained by errors in surgical technique, and infection is suspected despite MSIS criteria not being met.

The studies reviewed in this manuscript were limited in that none presented level I evidence (12 had level II evidence, and 3 had level III evidence), and there was significant heterogeneity (some studies used their own diagnostic standard, and others used the MSIS criteria). Larger scale prospective studies comparing serum ESR/CRP

level and synovial fluid analysis to novel synovial markers are needed.

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