



Culture-negative periprosthetic joint infections

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- Culture-negative periprosthetic joint infections (CN-PJI) pose a significant challenge in terms of diagnosis and management. The reported incidence of CN-PJI is reported to be between 7% and 15%.
- Fungi and mycobacterium are thought to be responsible for over 85% of such cases with more fastidious bacteria accounting for the rest.
- With the advent of polymerase chain reaction, mass spectrometry and next generation sequencing, identifying the causative organism(s) may become easier but such techniques are not readily available and are very costly.
- There are a number of more straightforward and relatively low-cost methods to help surgeons maximize the chances of diagnosing a PJI and identify the organisms responsible.
- This review article summarizes the main diagnostic tests currently available as well as providing a simple diagnostic clinical algorithm for CN-PJI.

Keywords: alpha-defensin; culture-negative periprosthetic joint infection; Dithiothreitol; interleukin 6; leucocyte esterase; next generation sequencing; polymerase chain reaction; sonication

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Introduction

Periprosthetic joint infections (PJI) remain a difficult and challenging complication of all joint arthroplasty surgery. The incidence of a PJI after a primary total hip replacement (THR) or knee replacement (TKR) is reported as between 2% and 2.4%.¹ In Europe, the mean PJI rate is 0.8% for TKR and 1.2% for THR but considerable variation exists between countries.² The management of a PJI is complex and expensive, requiring, in most cases, revision surgery

and long-term use of antibiotics, and is associated with significant morbidity and mortality. The annual cost of PJI in the United States is expected to be around \$1.6 billion by 2020¹ although this figure is likely to be a gross underestimate in that it only addresses direct hospital costs.³ In the majority of cases, the diagnosis of a PJI is relatively straightforward with clear clinical evidence of an infection, raised inflammatory markers such as C-reactive protein (CRP), evidence of loosening on the plain radiographs and a positive culture result from sampling.

There are a number of diagnostic criteria used to diagnose a PJI, including those of the Musculoskeletal Infection Society (MSIS),⁴ the International Consensus Meeting (ICM) in 2013⁵ and the European Bone and Joint Infection Society (EBJIS).⁶ They all share some common criteria (Table 1) with a few individual variations. More recently, Parvizi et al published a diagnostic score for PJI taking into account some of the new diagnostic assays currently in use such as the alpha-1 defensin or D-dimer assays.⁷

The cut-off value for leucocyte cell count previously used in diagnosing PJI has varied from study to study and between THR and TKR. The 2013 ICM determined that a figure of > 3000 leu/ μ L be used in diagnosing PJI.⁵ With regard to cases of PJI after THR, Schinsky et al concluded that the optimal cut off for leucocyte level was 4200 leu/ μ L. This level could be lowered to 3000 leu/ μ L when used in conjunction with serum inflammatory markers giving a sensitivity of 92% and a specificity of 86%.⁸ A recent paper looking to define PJI again used a level of > 3000 leu/ μ L and showed a higher sensitivity and similar specificity when compared to MSIS and ICM definitions.⁷

In a small number of cases, the diagnosis of a PJI remains unclear and the microbiological cultures are negative. Culture-negative periprosthetic joint infections (CN-PJI) were originally defined by Berbari et al⁹ as no growth of either aerobic or anaerobic cultures taken from periprosthetic tissue with either the presence of periprosthetic purulence, presence of acute inflammation (on histopathological

Table 1. Diagnostic criteria for a periprosthetic joint infection (PJI)

Criterion	Definition of PJI				
	MSIS ^a		ICM (2013) ^b		EBJIS ^c
	Definitive	Supportive	Definitive	Supportive	Definitive
<i>Sinus tract communicating with the prosthesis</i>	x		x		x
<i>Identical micro-organisms isolated from > 2 cultures</i>	x		x		x
<i>Purulence surrounding prosthesis</i>		x			x
<i>Acute inflammation (> 5 neutrophils per high-power field in 5 high-power fields observed from histological analysis of periprosthetic tissue at 400x magnification)</i>		x		x	x
<i>Single culture of any micro-organism (including a virulent organism)</i>		x		x	x
<i>Elevated synovial fluid leucocyte count (>3000 µL)**</i>		x		x	x
<i>Elevated synovial fluid neutrophil percentage* (> 80% PMN)</i>		x		x	x
<i>Elevated serum ESR and CRP values</i>		x		x	

Note. MSIS, Musculoskeletal Infection Society; ICM, International Consensus Meeting; EBJIS, European Bone and Joint Infection Society.

^aMSIS definition requires 1 definitive criterion or 4 (out of 6) supportive criteria.

^bICM (2013) definition requires 1 definitive or 3 (out of 5) supportive criteria; * > 80–90% PMN; ** or +/++ on leucocytes esterase testing (ICM 2013).

^cEBJIS definition requires 1 or more definitive criteria.

Table 2. Definition of culture-negative periprosthetic joint infection (any one of the following features below)⁹

Periprosthetic purulence observed at the time of operation
Histopathological features consistent with acute inflammation
Elevated synovial white cell count (> 1.7 x 10 ³ /µL ³) or elevated synovial neutrophil (PMN) percentage (> 65% PMNs)
Sinus track in direct communication with the joint

tissue samples) or a sinus tract communicating with the prosthesis (see Table 2). There is currently no gold standard for diagnosing a CN-PJI. It is important to differentiate whether a culture-negative PJI is a true negative (i.e. aseptic loosening) or a false negative result (diagnostic tests have failed to identify an organism but there is a PJI). There are two broad groups of patients who may have a CN-PJI:

Group A – A PJI which is clearly infected but microbiological cultures remain negative. This situation usually arises as a result of the administration of antibiotics prior to sampling. The other reason there may be a failure to culture an organism is the fact that most bacteria responsible for a PJI form biofilms and simply do not produce colonies when they are transferred to the surfaces of agar plates.¹⁰

Group B – A potential PJI, but cultures are negative and there is no obvious clinical evidence of infection such as a sinus or purulent discharge (as per the Berbari definition of a CN-PJI). This situation may represent the presence of a low-grade infection caused by fungal or atypical pathogens such as *coxiella burnetti*, *propionibacter* or *mycobacterium*. Some (if not most) of the cases of so-called aseptic loosening may actually represent low-grade PJIs which have not been diagnosed.^{10,11}

The first step is to confirm whether there is a PJI using the appropriate serological, microbiological and radiological

tests (as described below). The next step, having established that there is a PJI, is to determine whether newer diagnostic means can help identify organism(s) causing the PJI together with their sensitivities and resistance to antimicrobials (antibiogram) if initial, more traditional microbiological cultures are negative.

Epidemiology

The prevalence of culture-negative PJI ranges from 5% to 42% (see Table 3). Most of the studies on this topic are retrospective in nature. The consensus appears to be that the true incidence of culture-negative PJI is between 7% and 15%.³ By far the overwhelming majority (98%) of CN-PJI relates to total hip and knee replacements.⁹

Microbiology of CN-PJI

The pathogenesis of CN-PJI is thought to be due to fungal and mycobacterial infections in over 85% of all cases.²³ The remainder of the causative organisms are bacterial, including fastidious bacteria which are difficult to culture and require specialized microbiological techniques to detect (see Table 4 for the range of organisms responsible for CN-PJI). In particular, *Brucella* and *Coxiella burnetii* are responsible for the majority (> 50%) of bacterial CN-PJI and standard culture methods can fail to detect these bacteria.^{23,24}

Table 3. Description of studies and the prevalence of culture-negative periprosthetic joint infection

Study title	Author	Joint	Prevalence (%)	Study type	Study design	No. cases in study	Year
Two-stage revision for the culture-negative infected total hip arthroplasty	Ibrahim MS et al ¹²	Hip	N/A	Case control	Prospective	50	2018
Two-stage revisions for culture-negative infected total knee arthroplasties: a five-year outcome in comparison with one-stage and two-stage revisions for culture-positive cases	Li H et al ¹³	Knee	14.2	Case control	Retrospective	129	2017
Comparison of infection control rates and clinical outcomes in culture-positive and culture-negative infected total-knee arthroplasty	Kim Y-H et al ¹⁴	Knee	42.1	Case control	Retrospective	242	2015
The outcome of infected total knee arthroplasty: culture-positive versus culture-negative	Kim Y-H et al ¹⁵	Knee	26.7	Case control	Retrospective	191	2015
Organism profile in periprosthetic joint infection: pathogens differ at two arthroplasty referral centers in Europe and in the United States	Aggarwal VK et al ¹⁶	USA: Hip (63%) Knee (37%) Europe: Hip (46%) Knee (54%)	USA: 15.8 Europe: 16.1	Case control	Retrospective	USA: 772 Europe: 898	2014
Periprosthetic joint infections treated with two-stage revision over 14 years: an evolving microbiology profile	Bjerke-Kroll BT et al ¹⁷	Hip (50%) Knee (50%)	21.3	Case control	Retrospective	785	2014
Periprosthetic joint infection with negative culture results: clinical characteristics and treatment outcome	Choi HR et al ¹⁸	Hip (50%) Knee (50%)	23.0	Case control	Retrospective	175	2013
Culture negative prosthetic joint infection: a description of current treatment and outcomes	Peel TN et al ¹⁹	Hip (21%) Knee (79%)	N/A	Case cohort	Retrospective	19	2013
Culture-negative periprosthetic joint infection does not preclude infection control	Huang R et al ²⁰	Hip (46%) Knee (54%)	16.3	Case control	Retrospective	295	2012
Prior use of antimicrobial therapy is a risk factor for culture-negative prosthetic joint infection	Malekzadeh D et al ²¹	Hip (50%) Knee (50%)	10.5	Case control	Retrospective	1413	2010
Two-stage revision arthroplasty of the hip for infection using an interim articulated Prostalac hip spacer	Biring GS et al ²²	Hip	5.1	Case control	Retrospective	99	2009
Culture-negative prosthetic joint infection	Berbari EF et al ⁹	Hip (45%) Knee (55%)	7.0	Case control	Retrospective	897	2007

Risk factors for CN-PJI

The risk factors for CN-PJI are similar to those for culture-positive PJI²⁴ and include obesity, age > 65 years, male gender and co-morbidities (chronic renal disease, diabetes mellitus, rheumatoid arthritis, liver disease and vascular insufficiency).²¹ Obesity in particular is a strong risk factor for PJI in patients with a BMI \geq 30 kg/m.²⁵ For CN-PJI, specific risk factors include previous history of a PJI or surgical site infection (SSI), previous revision surgery and prior antibiotic use. In a retrospective case control study of 135 patients with CN-PJI, the authors found that 64% of these patients had prior antibiotic use and the odds ratio for a CN-PJI was 4.7. The odds ratio was 3.5 with postoperative wound discharge.²¹

Diagnosing a CN-PJI

Diagnosing a CN-PJI is difficult and may require a combination of clinical, radiological, serological (inflammatory markers), histopathological and microbiological assessments.

This review will focus specifically on inflammatory markers currently in use for the diagnosis of a PJI.

C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR)

Serum CRP and ESR are widely used serum inflammatory markers in the diagnosis of a PJI. For early PJI, a CRP > 10 mg/L is the threshold, and for ESR it is > 30 mm/hr. The CRP rises after surgery reaching a peak level on day two before returning to preoperative levels after three weeks. The ESR reaches a peak level on day five after surgery and remains elevated for up to 12 months postoperatively.^{26,27} They are non-specific for infection and can be elevated for other reasons such as in rheumatoid arthritis or if there is concurrent infection in the patient elsewhere such as a chest or urinary tract infection.³ The pooled sensitivity for CRP is 88% with a specificity of 74% based on a meta-analysis of 23 papers by Berbari et al.²⁶ The pooled sensitivity for ESR was lower at 75% with a specificity of 70%.²⁶ A normal CRP (< 10 mg/L) and ESR (< 30 mm/hr) had a sensitivity of 96% for excluding a PJI.²⁸

Leucocyte esterase

Leucocyte esterase (LE) is an enzyme secreted by neutrophils in the presence of infection. Testing for LE has been used in the diagnosis of urinary tract infections for many years. Parvizi et al first described its potential in diagnosing PJI in 2011, with a specificity and sensitivity of 100% and 80.6% respectively among 30 cases of PJI undergoing revision arthroplasty.²⁹ This test is cheap and easy to perform. A positive test occurs in seconds in the presence of LE, although most studies allow two to three minutes before recording results. Detection relies upon a hydrolytic reaction resulting in a colour change on colorimetric reagent testing strips catalysed by LE which can be performed intraoperatively with ease.³ This is graded by degree of colour change occurring and different studies have used different thresholds to represent a positive result.³⁰ Blood-stained synovial fluid has been shown to affect interpretation of results and is therefore a limitation of this test. Using a centrifuge has been shown to counter this problem without compromising accuracy.³¹ A meta-analysis performed by Wyatt et al looked at 545 patients undergoing predominantly hip and knee arthroplasty. There were 151 cases of PJI included and the pooled sensitivity and specificity were 81% and 97% respectively.⁷ Higher sensitivities in other studies have been reported although with smaller sample sizes.⁸ Another limitation of this test is that it does not identify the causative organisms. LE is a cheap screening tool which, due to its specificity, has been shown to be accurate at excluding cases of infection in cases of PJI.

Alpha-defensin assay

There has been increasing interest in the use of the alpha-defensin assay as a diagnostic tool for PJI. The alpha-defensin protein is an antimicrobial peptide that is naturally released by neutrophils responding to a pathogen in the synovial fluid.³² The alpha-defensin assay can be either performed in the laboratory as an immunological assay or performed at the bedside using the lateral flow alpha-defensin test (Synovasure, Zimmer). This is a qualitative immunoassay optimized and validated for synovial fluid and it takes ten minutes to provide a result. The alpha-defensin concentration is shown in terms of a signal-to-cut-off ratio (S/CO).³² The alpha-defensin test results are clinically reported as 'positive' or 'negative', although the S/CO value reflects the concentration of alpha-defensin in each synovial fluid sample. The alpha defensin assays have been shown to have a high sensitivity (97%), high specificity (97%), high positive predictive value (88%) and negative predictive value (99%).³³ The systematic review and meta-analysis by Wyatt et al in 2016 found that the alpha-defensin assay had a sensitivity of 100% and specificity of 96%.³⁴ The laboratory-based

assay has been shown to be superior in terms of sensitivity and specificity compared with the lateral flow assay.³⁵ In their meta-analysis, Suen et al showed that the laboratory-based assay had a sensitivity of 95.0% and specificity of 96.5%.³⁵ The lateral flow assay had a sensitivity of 77.4% and a specificity of 91.3%. Sigmund et al. showed that Synovasure had a sensitivity of 69% and a specificity of 94% in 50 patients.³⁶ A very recent study in May 2018 by Renz et al has cast some doubt on how sensitive the lateral flow assay is. In their study of 212 patients, of whom 151 (71%) had had a knee replacement and 61 (29%) had had a hip replacement, the lateral flow assay had a sensitivity of between 54% to 84% depending on which criterion was used to diagnose a periprosthetic joint infection. The specificity of the assay was very high (96–99%).³⁷ The authors suggest that the lateral flow assay should not be used as a screening test but rather to confirm a PJI. A major advantage of the alpha-defensin assay is that it is unaffected by prior antibiotic use in the presence of a PJI.³⁸ The major limitation of the test is that it does not provide a microbiological target in terms of a positive microbiological culture and associated sensitivities to antibiotics.

D-dimer

D-dimers are fibrin degradation products formed when a fibrin clot is dissolved by plasmin. They are elevated in a number of conditions, most notably in the presence of a venous thromboembolic event (VTE) such as a deep vein thrombosis or pulmonary embolus. They are also raised in the presence of malignancy, infection and after surgery. Lately, there has been interest in how D-dimer levels are influenced in the presence of a PJI. A study from South Korea showed that D-dimer concentration levels rise and fall rapidly within two days postoperatively after THR and TKR and concluded by suggesting that D-dimer levels together with CRP and ESR may be a useful marker for early PJI.³⁹ D-dimer levels were found to have a better sensitivity (89%) and specificity (93%) compared to CRP and ESR.⁴⁰ D-dimer is a cheap and easy test to perform. An immediate rise in levels postoperatively, followed by a sharp return to normal offers the promise of using D-dimer as a marker of both early and late PJI.

Interleukin-6 (IL-6) assay

Lymphoid and non-lymphoid cells produce IL-6 which is part of the inflammatory response, increasing with trauma, infection, and surgery. In patients with aseptic prosthetic loosening, IL-6 levels decrease to the normal level within 48 hours after arthroplasty. However, following infection, IL-6 activates the release of CRP. Therefore, the increase of IL-6 precedes that of CRP after infection; thus, IL-6 may be a more sensitive marker for PJI.⁴¹ In a study of 40 patients suspected of having a PJI

Table 4. Organisms responsible for culture-negative periprosthetic joint infection²³

Organism	Percentage (%)
Fungi	46
Mycobacteria	43
Bacteria	11
Brucella	35
Coxiella burnetii	16
Listeria monocytogenes	3
Propionibacterium acnes	6
Staphylococcus*	6
Streptococcus*	10
Lactobacillus spp.	–
Pasteurella multocida	–
Pseudomonas spp	–
Serratia marcescens	–

*These organisms typically form biofilms and this may account for the fact that microbiological cultures are negative (even if antibiotics have not been used before sampling has taken place).

after hip and knee arthroplasties, 11 had a proven PJI with elevated serum IL-6 (> 10.4 pg/ml). IL-6 had a sensitivity of 100%, specificity of 90.9%, PPV of 79%, NPV of 100%, and accuracy of 92.5%.⁴² Lenski and Scherer found that synovial fluid IL-6 had a sensitivity of 90% and a specificity of 94.7%.⁴³ A systematic review and meta-analysis by Yoon et al in 2018 looked at 16 studies using IL-6 as a diagnostic tool for PJI, involving 1327 subjects. The authors found a pooled sensitivity for synovial fluid IL-6 of 83%, specificity of 91%, positive likelihood ratio of 9.3 and negative likelihood ratio of 0.19 and concluded that IL-6 was a useful diagnostic tool for PJI and IL-6 assays are readily available.⁴⁴

Synovial fluid D-lactate

There has been increasing interest in looking at D-lactate in synovial fluid as a specific bacterial biomarker for PJI as it is a product of bacterial metabolism. In a small study of 58 patients with a PJI, Karbysheva et al found that D-lactate had a sensitivity of 96.5% and could be performed as a 'point-of-service' test (taking under one hour to get a result).⁴⁵ It has been suggested that the D-lactate test would be especially useful when a synovial differential leucocyte count was uninterpretable.⁴⁶

Identifying the organism

Microbiological culture (extended)

The commonest reason for a culture-negative PJI is the use of broad-spectrum antibiotics prior to synovial fluid or tissue samples being obtained for microbiological and histological analysis. Barbari et al found that 53% of CN-PJI had preceding antibiotic therapy before a diagnosis of PJI had been established.⁹ Another reason is that the current length of time for bacterial cultures may be insufficiently short (five days rather than up to two weeks) and/or atypical

organisms (such as fungi, mycobacterium or Coxiella) are not tested for.²⁴ Fastidious organisms such as *Cutibacterium acnes* (formerly known as *Propionibacterium acnes*) can require incubation for 14–21 days to isolate.^{47,48} *Cutibacterium acnes* is a gram-positive anaerobe which is most commonly associated with shoulder PJI due to an association with sebaceous follicles located in the axilla.⁴⁹ Its role in PJI is thought to be underestimated due to the need for an extended incubation period after taking cultures. Cases of PJI caused by *Cutibacterium acnes* are on the increase and this is likely due to improved diagnostic methods, and an awareness of the need for extended cultures. In a study comparing routine cultures with next generation sequencing (NGS) in patients undergoing revision shoulder replacement surgery, the authors found fair concordance between routine cultures and NGS techniques. The most common organism found in their study was *Cutibacterium acnes*, accounting for almost 62% of cases. Next generation sequencing techniques suggest that, for revision shoulder replacements, polymicrobial infections are more likely than monomicrobial infections.⁵⁰ Organisms such as *Coxiella burnetii* have been reported in the literature as being a cause of a PJI but special microbiological tests are required to identify this organism.²³ The same authors also reported that up to 46% of culture-negative PJI were due to fungal infections and 43% were due to mycobacterium.²³

Sonication

Synovial fluid cultures and intraoperative tissue cultures are the current standard for detecting bacterial speciation but can have a high false-negative rate (17% to 53%).⁵¹ Organisms associated with PJI often form a biofilm on the surface of the implant. A biofilm is a structured aggregation of bacterial cells of one or more species, encased in a self-produced matrix and adherent to the implant surface.⁵² Sonication helps increase the number of bacterial cells available for culture by disrupting the bacteria found in the biofilm (Fig. 1). The sensitivity of sonication has been shown to range from 60% to 97% and the specificity to range from 90% to 99%.^{53–59} The meta-analysis by Liu et al of 16 studies, involving 2390 cases, found a pooled sensitivity for sonication of 79% and a specificity of 97%.⁶⁰ The area under curve (AUC) for the summary receiver operating characteristic (SROC) was 0.9. The studies included used sonication with or without vortexing. Given the time required and expense incurred in performing sonication, our recommendation is that it should not be used routinely but reserved for select cases of CN-PJI where sonication may increase the chances of identifying a causative organism.^{61,62}

Dithiothreitol

Dithiothreitol (DTT) is a sulfhydryl compound acting as strong reducing agent and protein denaturant, thanks to



Fig. 1 MicroDTTect analysis bag (photo courtesy of Heraeus Medical GmbH).

its ability to reduce disulphide bridges and prevent the formation of intramolecular and intermolecular bonds between cysteine residues. Very recently, a few studies have looked into the use of DTT in dissolving biofilm, helping improve the bacterial yield from implants in the presence of a PJI.⁶³ These studies have all been from a single institution and using a commercially available product (MicroDTTect, Heraeus) (Fig. 1). The authors of one study report a sensitivity of 88.0% and a specificity of 97.8%,⁶⁴ whilst in a separate study comparing the use of DTT versus sonication, the sensitivity of the DTT test was found to be 85.7% and the specificity was 94.1%.⁵⁹ The use of DTT needs further studies to corroborate the findings from Drago's team at the Galeazzi Orthopaedic Institute, Milan.⁵⁹

Polymerase chain reaction (PCR) dsRNA analysis

A meta-analysis of 14 studies (1480 subjects) looking at the diagnostic capabilities of PCR-based assays in PJI showed a pooled sensitivity of 86% and a specificity of 91%. The commonest form of PCR assay was the 16S rRNA type.⁶⁵ The use of PCR assays will become increasingly common, especially in the presence of CN-PJI, in order to optimize the chances of identifying the causative organism(s).

Ibis PLEX-ID

A new sophisticated modality (the Ibis Biosciences T5000 biosensor system) has been introduced that uses pan-domain primers in a series of PCRs to identify the species of all bacteria and fungi as well as to identify key antibiotic resistance genes.¹¹ The Ibis database contains the base ratios of thousands of known bacterial pathogens, and the base ratios of the bacteria in samples can be determined and matched to those in the database, to determine the

presence of any of these organisms.¹⁰ The Ibis technology has been shown to detect organisms in cases of culture-negative PJI but also cases of presumed aseptic loosening in revision hip and knee arthroplasty. In a series of 57 cases of revision arthroplasty in which aseptic loosening was thought to be the reason for failure, 50 (88%) of these cases had one or more organisms detected using Ibis system.¹¹

PCR mass spectrometry

PCR-electrospray ionization mass spectrometry (PCR-ESI/MS) was first described in 2005 as a novel diagnostic tool for detecting infections. In 2008, the technology was advanced and has been used to detect a broad range of bacterial, fungal and yeast-based infections.⁶⁶ PCR-ESI/MS sensitivity (77.6%) for detecting PJI from sonicate fluid is similar to the sensitivity (77.1%) of a genus-/group-specific rapid PCR panel assay targeting PJI bacteria and superior to that of a 16S rRNA gene PCR assay (sensitivity of 70.4%).⁶⁶ PCR-ESI/MS is less affected than culture by prior antimicrobials: PCR-ESI/MS sensitivity was 85.7% among subjects receiving antimicrobial therapy within 14 or 28 days of surgery.⁶⁶ A similar study also showed that the sensitivity and specificity of PCR-ESI/MS were 81% and 95% respectively in diagnosing a PJI.⁶⁷

Next generation sequencing

The field of genomics has advanced significantly with the use of next generation sequencing (NGS), a DNA sequencing technology which has revolutionized research in genetics and enables the entire human genome to be sequenced in a day.⁶⁸ Within the field of microbiology and with respect to PJI, the use of NGS techniques may be very useful in cases of CN-PJI where more traditional microbiological techniques of culturing organisms have failed to identify the causative organism, by providing a genomic definition of the organism.⁶⁸ The genomes of pathogens define what they are, and can provide further information about their drug sensitivity and the relationship of different pathogens with each other.⁶⁸ In a study of 86 anonymized synovial fluid samples, the use of NGS was useful in detecting the pathogen in cases of PJI with a high (> 96%) concordance rate with routine microbiological culture and was also useful in detecting organisms in CN-PJI.⁶⁹ In a separate study, NGS techniques were used to identify organisms in 168 failed total knee replacements and, in 16% of the CN-PJI (25 cases), yielded a potential organism.⁷⁰ The authors concluded that NGS may have a valuable role to play in CN-PJI cases. The concern remains, however, that such techniques may provide false-positive results because these molecular techniques are so sensitive and may indeed identify contaminant organisms, leading to over treatment of patients who do not have a

Table 5. Direct costs of different diagnostic tests

Test	Approximate direct cost per unit (£)
D-dimer	£20
Leucocyte esterase (with mini centrifuge)	£0.58/strip ³¹ (£328.12 for mini centrifuge)
Alpha-defensin (Synovasure)	£500
Traditional tissue culture	£274 ⁷¹
MicroDTTect	£350 ⁷¹
Sonication	£353 ⁷¹

PJI.⁶⁹ Meticulous care in handling synovial fluid specimens, from patient to laboratory, is required in order to minimize the risk of specimen contamination.

Financial costs

Inevitably, the emergence of newer diagnostic techniques for PJI and identification of the organisms responsible can be costly. There is an enormous variation in costs from relatively low-cost tests such as the leucocyte esterase strips to the expensive alpha-defensin assays, MicroDTTect kits, sonication and indeed molecular and genetic techniques (Table 5). It still remains unclear as to whether the relative expense of these newer tests can be justified, especially in a

publicly funded healthcare system such as the NHS in the UK. Further research is needed in order to determine the health economic benefit of such expensive tests.

Conclusions

In summary:

- There needs to be a greater awareness that ‘aseptic loosening’ may instead be a low-grade infection.
- In the event that a CN-PJI is suspected, repeated sampling should be carried out (with the patient having been off antibiotics for as long as possible, at least two weeks) and any samples should be cultured for 14–21 days and potentially checked for fungi and atypical bacteria (see Figure 2).
- PJI cases may be better managed in a centre with experience and expertise, within a multi-disciplinary team setting, where cases can be discussed amongst experienced arthroplasty surgeons, microbiologists, radiologists and other healthcare professionals with an interest in PJI, with access to the newer diagnostic techniques.

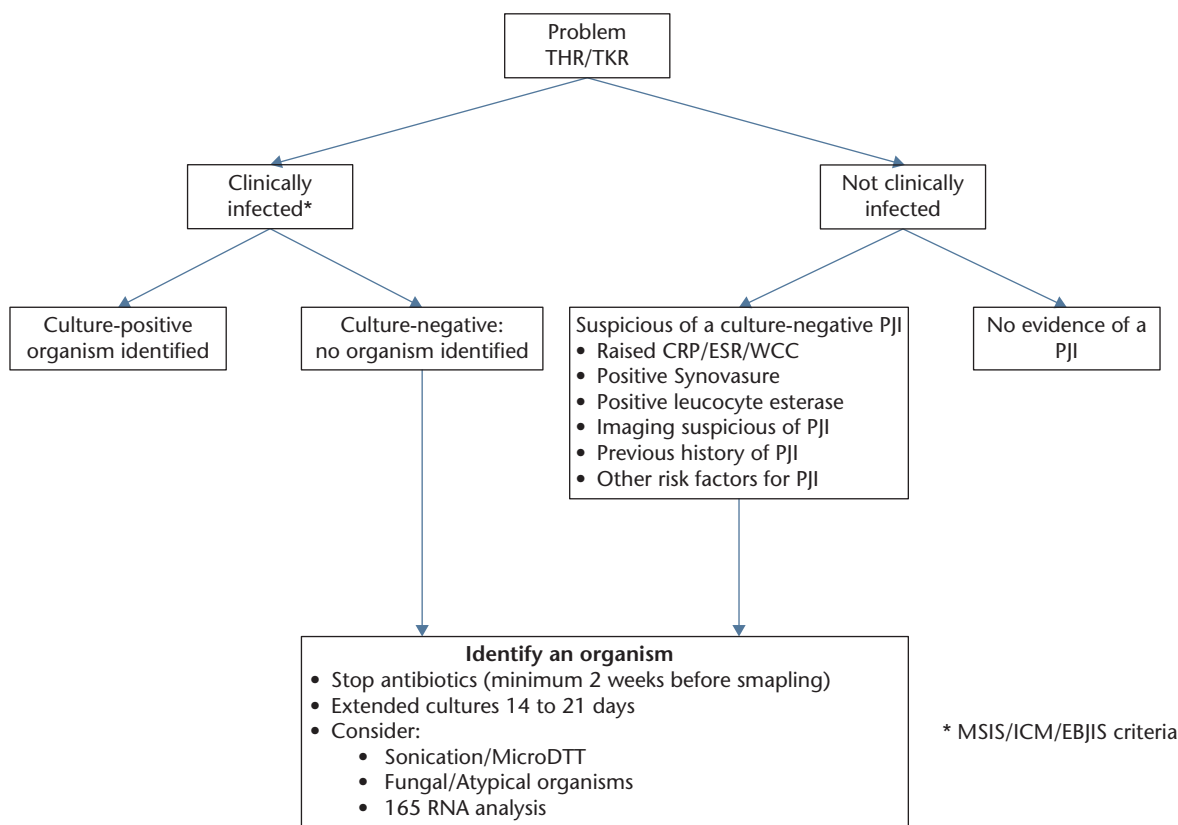


Fig. 2 Diagnostic algorithm for culture-negative periprosthetic joint infection (PJI). *Note.* THR, Total Hip Replacement; TKR, Total Knee Replacement; CRP, C-Reactive Protein; ESR, Erythrocyte Sedimentation Rate; WCC, White Cell Count; Hx, History.

- Meticulous care when sampling should be taken by the orthopaedic surgeon ensuring that, where possible, antibiotics have been stopped for at least two weeks prior to sampling.

The diagnosis and management of PJI is challenging and especially when a CN-PJI is suspected. Such cases should be treated in much the same way as oncological conditions, within a multi-disciplinary setting with clinicians who have a specialist interest in PJI. With the advent of newer serum biomarkers, more sophisticated imaging modalities and molecular-based microbiological techniques for identifying organisms, there is a greater armamentarium available to the orthopaedic surgeon. Some of these advances are still in their development phase and may prove costly to use. Furthermore, the use of such molecular techniques may identify many organisms creating ‘noise’, making it difficult to differentiate whether the results are due to contamination or are true. There are, however, more straightforward ways to quickly and inexpensively improve clinical practice.

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ICMJE CONFLICT OF INTEREST STATEMENT

RW reports an educational contract for teaching on Exeter Hip Courses for Stryker Europe outside the submitted work.

RK reports consultancy and education activities for Corin Group PLC; employment by the NHS as a place of clinical practice; payment for lectures on annual course for Heraeus; payment for development of educational presentations for Corin Group PLC; and accommodation and travel expenses provided for two educational courses for Stryker, all outside the submitted work.

PF reports consultancy for Microport, Corin, Zimmer/Biomet, MBA, 3M; payment for lectures including service on speakers bureaus for Microport, Corin, 3M; payment for development of educational presentations for Corin and 3M; and travel/accommodations/meeting expenses unrelated to activities listed for Corin, MBA and 3M, all outside the submitted work.

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LICENCE

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