

# National periprosthetic joint infection sampling and culture guide

Orthopaedic surgery

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## Document purpose

This guide from the Health Quality & Safety Commission outlines a contemporary approach to the laboratory diagnosis of periprosthetic joint infection (PJI).

Joint replacement surgery can greatly improve a person's quality of life. With an ageing population, more replacement procedures will be performed.

PJI is uncommon (~1 percent for primary arthroplasties) but causes significant patient morbidity.

The diagnosis of PJI can be challenging and different approaches have been taken, for example, the number of specimens obtained. Although the literature on the diagnosis of PJI continues to evolve, there is now sufficient data available to guide us on how to optimise the laboratory diagnosis of PJI.

We hope this guide brings greater consistency in approaches to diagnosing this complication of joint replacement surgery.

## Introduction

Microbiological sampling remains a cornerstone for both the diagnosis of PJI and the subsequent antimicrobial management.

Where microbiological investigations are performed to a high level, the culture results can help surgeons to:

- exclude infection and thereby avoid the morbidity of unnecessary surgery and prolonged antibiotic courses
- diagnose infection with a specific pathogen(s), allowing:
  - targeted local antimicrobial measures (eg, antibiotic beads or antibiotic-loaded cement spacers)
  - targeted systemic antimicrobial therapy (eg, intravenous and oral therapy, including assessment of suitability for rifampicin use)
  - consideration of patients suitable for one-stage exchange surgery.

As with any test, microbiological sampling is neither 100 percent sensitive nor 100 percent specific. In order to optimise the quality of information from microbiological sampling, all efforts should be made to obtain appropriate specimen types in sufficient numbers from the most appropriate sites.

This document provides guidance on standard of care microbiological sampling and culture approaches for suspected PJIs.

## Timing of empiric treatment antibiotics with respect to microbiological sampling

In this section, we discuss the timing of empiric antibiotics for the treatment of suspected/confirmed PJI. For the timing of administration of usual surgical antibiotic prophylaxis, see [Intraoperative microbiological sampling](#) below.

There are two reasons for the use of empiric antibiotics to treat patients with PJI:

1. Stabilisation of the acutely septic patient.
2. Maximising the chances of cure (or suppression) of the PJI.

Only a minority of patients with PJIs require administration of acute, empiric antibiotic treatment before microbiological samples are obtained. In general, these patients present with acute sepsis, haemodynamic instability and/or new end-organ dysfunction. Even in these situations, take at least two sets of blood cultures and obtain microbiological samples as soon as possible after initiating empiric antibiotic treatment (for example, joint aspirates for easily accessible joints or intraoperative samples from acute operative interventions).

For the majority of patients with PJIs, antibiotics can be safely withheld until **after** microbiological sampling has been **completed**. This generally includes two sets of blood cultures, joint aspiration (eg, for knee joints) and intraoperative sampling.

### Patients undergoing surgery with debridement, antibiotics and implant retention (DAIR)

Patients undergoing DAIR procedures with the intention of cure (as opposed to long-term antibiotic suppression) have been shown to have lower chances of cure the longer operative intervention is delayed.<sup>1</sup> Therefore, there should be a degree of urgency for these patients to have expedited operative debridement and microbiological sampling. Patients should receive empiric antibiotic treatment immediately after the completion of intraoperative microbiological sampling.

Most guidelines would suggest limiting the use of DAIR strategies with a curative intent to patients with acute infections (< 3 weeks' duration) with stable prostheses and no sinus tract.<sup>2,3</sup>

### Patients with chronic PJIs and/or undergoing implant exchange surgery

For patients with chronic PJIs and/or who are undergoing implant exchange surgery, **withhold** all antibiotics **until after** completion of all microbiological sampling. Ideally withhold antibiotics for a minimum of 14 days before sampling. This includes patients who have culture-positive, preoperative joint aspirates.

The rationale for delayed antibiotics in this scenario is as follows:

1. Administration of antibiotics before operative debridement (and sampling) is unlikely to have any significant impact on the chances of infective cure for chronic PJIs and/or those undergoing implant exchange surgery.
2. Intraoperative cultures remain the most sensitive and specific culture technique. Administration of antibiotics before sampling will decrease the culture yield, particularly in chronic PJIs where organism turnover is dramatically reduced.

3. The specificity of preoperative aspirate culture results using intraoperative culture as the gold standard approximates 91 percent.<sup>4</sup> Intraoperative cultures are therefore critically important to confirm the validity of aspirate results.
4. Patients with chronic PJIs and/or who are undergoing elective implant exchange surgeries have a low risk of developing sepsis while awaiting surgery.

## Preoperative joint aspiration

### What is the role of preoperative joint aspiration in PJIs?

Preoperative joint aspiration can provide invaluable information in cases of suspected PJI.

Although not conclusive alone, culture-negative aspirates with low-level synovial cell counts (see [suggested synovial cell count cut-offs](#) below) provide additional evidence for the exclusion of PJIs.

Culture-positive preoperative aspiration cultures can inform surgical options (eg, one-stage exchange surgery) and perioperative antibiotic plans (targeted local and systemic antibiotic choice). In patients where operative mortality risks are too high, joint aspiration can sometimes represent the only opportunity to obtain joint samples for culture.

Due to a significant rate of false-negative and false-positive (contaminated) cultures, preoperative joint aspirations do not replace the need for high-quality intraoperative sampling.<sup>4</sup>

### Is there a role for intraoperative Gram stain results in the exclusion of PJI?

No. Gram stain is a rapid microscopy technique used for identifying the presence of bacteria. A positive Gram stain result requires a significant burden of infection to be present. When positive, it is very specific for infection (97–100 percent) however it is a poor test for exclusion of PJI due to its low sensitivity (19–44 percent).<sup>5</sup> Gram staining should therefore never be used to exclude PJI.

### What synovial cell counts are indicative of PJI?

In the absence of an inflammatory arthropathy (eg, rheumatoid arthritis), a synovial white cell count  $\geq 1,700$  cells  $\times 10^6/L$  or a neutrophil percentage  $> 65$  percent is highly suggestive of a PJI in the non-acute setting.<sup>6</sup> In one study of prosthetic knees, a synovial white cell count  $\geq 1,700$  cells  $\times 10^6/L$  was associated with a sensitivity of 94 percent and specificity of 88 percent for PJI.<sup>7</sup>

In acute infection (within six weeks of operation or haematogenous infection), the cell count and neutrophil percentage cut-offs have not yet been determined. One study suggests a synovial white cell count  $\geq 27,800$  cells  $\times 10^6/L$  (sensitivity 84 percent, specificity 99 percent) or a polymorph percentage  $\geq 89$  percent (sensitivity 84 percent, specificity 69 percent) as optimal cut-offs within the first six weeks of primary total knee arthroplasty; however, further studies are required to validate these results.<sup>8</sup>

## Intraoperative microbiological sampling

Intraoperative microbiological sampling represents the gold standard for investigation of suspected PJI. It is the only available technique for determining the antibiotic susceptibility of infecting pathogens. Due to the ability to take multiple samples, this reduces the risk of false-positive and false-negative results compared with preoperative joint aspirates (a single sample).

### Should usual surgical antibiotic prophylaxis be withheld before intraoperative microbiological sampling?

A recent systematic review found no negative impact on culture results in patients with suspected but unconfirmed chronic PJI before surgery.<sup>9</sup> As surgical site infection (SSI) is more common after revision arthroplasty, especially in those with a high BMI, any negative impact on cultures, by giving prophylaxis on time, must be weighed against the need to reduce SSI risk following revision. On balance, prophylaxis should be given on time in most situations.

1. Administer usual surgical antibiotic prophylaxis **on time**, before microbiological sampling when:
  - a. there is a low suspicion of PJI
  - b. PJI has been confirmed and the microbiology is known
  - c. chronic PJI is suspected, but not confirmed, before surgery.

When there is a low suspicion of PJI and culture results are likely to be negative, give usual surgical antibiotic prophylaxis to minimise the risk of SSI. Where the microbiology of a confirmed PJI is already known, the benefit of withholding usual surgical antibiotic prophylaxis is felt to be negligible.

2. Withhold usual surgical antibiotic prophylaxis **until after** microbiological sampling for patients with a moderate–high suspicion of PJI. Where there is a moderate–high suspicion of PJI (in the absence of an identified pathogen), accurate microbiology is critical for optimising the patient’s surgical and antibiotic treatment plan. Every attempt should be made to identify the infecting pathogen(s). Only delay prophylaxis after full evaluation of the risk (higher SSI rate) versus benefit (better culture sensitivity).

These recommendations are in line with existing international PJI guidelines.<sup>5,10</sup>

### What specimen types should be taken?

Intraoperative microbiology samples should only consist of deep periprosthetic tissue samples and deep joint aspirates (intra-articular fluid/pus). Common samples and their yield in a recent large multicentre study were: cancellous bone (77 percent); bone in contact with cement (79 percent); capsule (85 percent); cortical bone (87 percent); synovial tissue (88 percent); sub-fascial tissue (89 percent); tissue in contact with material (92 percent); and joint fluid (92 percent).<sup>11</sup>

The following specimen types should **not** be performed.

### 1. Sinus tract samples

Sinus tracts inevitably become colonised with bacteria. Sinus tract cultures do not accurately reflect pathogens causing deep infection.<sup>7</sup>

### 2. Swabs

Swabs collect a lower infective burden than tissue samples and are more prone to desiccation. Swab cultures are less accurate than tissue cultures, which should be taken in preference.<sup>12</sup> Laboratories should inform local surgeons that swabs are not acceptable specimens for diagnosing PJI.

### 3. Superficial samples

Superficial cultures should not be used to infer the results of deep cultures and have no role in the investigation of deep PJI. If taking superficial samples (eg, you suspect only superficial infection), they must be clearly labelled as superficial and ideally should indicate why a superficial sample was taken. The interpretation of microbiological results is often performed by clinicians not present in theatre and for whom the site of sampling is critical.

## How many samples should be taken?

The isolation of a recognised pathogen from an appropriate specimen is persuasive evidence for infection. Such pathogens, like *Staphylococcus aureus*, *Streptococcus agalactiae* and Enterobacteriaceae, are rarely culture contaminants. Many PJIs are, however, caused by skin commensals, such as coagulase-negative staphylococci and, for shoulders, *Propionibacterium acnes*, which are recognised culture contaminants. The clinical interpretation of such isolates therefore rests on the number of independent cultures they are present in.

The longest-standing and most frequently quoted reference for the number of samples is that from the Oxford unit.<sup>13</sup> The culture method used in that study comprised agar media and a cooked meat broth. The authors recommended five or six specimens be obtained and the cut-off for definite PJI be three or more cultures with an indistinguishable isolate.<sup>13</sup> A number of recent large studies, using contemporary automated blood culture methods, concluded that 3–4 specimens are sufficient to allow the recovery of an infecting organism in two or more cultures.<sup>11,14,15</sup> See [Appendix 1](#) for a summary of the key references used as guidance on culture approaches and the number of specimens to collect.<sup>11,13–16</sup>

## Cultures not using contemporary automated blood culture bottles

The **minimum** microbiological sampling for suspected PJI is **five deep tissue samples** +/- a joint aspirate.

Take five deep tissue samples regardless of whether the clinical suspicion of PJI is high or low. Some centres have found that a lower number of samples submitted may reflect a lower suspicion of PJI,<sup>13</sup> however:

- where suspicion is low, repeatedly negative cultures strongly support exclusion of PJI and mitigate the risk of wrongly considering contaminants as evidence of infection
- where there is a high suspicion of PJI, multiple samples optimise the chances of identifying the infecting organism(s).

## Sensitivity and specificity of five deep tissue samples

### 1. Sensitivity

The sensitivity of microbiological culture relies on the laboratory receiving samples with adequate bacterial load for culture. Because of the heterogeneous distribution of bacteria in PJIs, multiple cultures minimise false negatives from sampling bias (eg, taking a sample from a site of low infective burden). Multiple samples are particularly important for chronic PJIs where the infective burden is lower and biofilm-associated bacteria are more difficult to culture due to a reduced metabolic turnover.

When five separate tissue cultures are taken and all are culture-negative, histological evidence of infection is present in only approximately 3 percent of cases.<sup>13</sup>

### 2. Specificity

The organisms that cause PJI include common culture contaminants (eg, coagulase-negative staphylococci). False-positive rates with a single positive culture approach 30 percent. When five separate tissue cultures are taken, growth of an indistinguishable organism from  $\geq 3$  samples has a sensitivity of 66 percent and a specificity of 99.6 percent.<sup>13</sup>

## Cultures using blood culture bottles incubated in automated systems

**Four samples** are sufficient when using contemporary blood culture systems.<sup>11,15</sup> While one study observed the best accuracy with three specimens, taking both sensitivity and specificity into account, four specimens achieved higher sensitivity.<sup>15</sup> Until more data is available on the reliability of fewer cultures, we recommend **four samples** when using an automated blood culture system for tissue samples. There should be local agreement on the minimum number of samples required based, in part, on the culture approach used in the serving laboratory.

There is not enough data to recommend the use of a blood culture set, ie, containing both an aerobic and anaerobic bottle, over a single paediatric bottle or vice versa. They are likely to be equivalent given the pathogens being sought in this setting. The choice will in part depend on the capacity of the laboratory's culture system, as will the decision to use blood cultures over other culture media. When evaluated, the use of an automated blood culture system was less costly.<sup>11,17</sup>

## How should samples be taken and from what sites?

Withhold antiseptic and antibiotic lavage until completion of microbiological sampling.

Wherever possible, use a dedicated PJI sampling surgical pack so you have the necessary equipment for sampling (see [Appendix 2](#) for a suggested list). Use fresh equipment (ie, new forceps or scalpel) for collection of each successive tissue sample, with assisting nursing staff handing off forceps to avoid contamination.

Take all the deep tissue samples deep to the fascia and accurately label them. We recommended using a dedicated form where possible (see [Appendix 3](#) for an example). Exact sites of biopsy will vary depending on operation type (debridement versus explantation will offer very different sampling sites), however, samples should always be taken from the most abnormal sites. Appropriate targets include synovial membrane, pseudocapsule and bone at the prosthesis interface. See above for [specimen types and their yield](#).<sup>11</sup> The larger

the tissue sample the better the yield; however, avoid cement debris in samples and **do not** send the prosthesis itself. Send all samples fresh in sterile containers.

Where there is synovial fluid/pus, take an aspirate. If possible, place at least 4 mL into a purple-top tube for cell count and differential, and place the remainder of the sample into a sterile container. The larger the sample, the higher the culture yield. If less than 4 mL are available, use 1 mL for the count tube and the rest for culture. Microbiology lab staff will perform the subsequent processing (inoculation onto agar and into blood culture bottles).

### **What is an acceptable timeframe for samples to reach the laboratory?**

Microbiological samples should follow routine transport times from the operating theatre to the laboratory (ie, hours rather than minutes). There is no role for urgent microbiological investigations to guide intraoperative management. Ideally samples should be processed the day they are collected. If this is not possible they may be stored at 4°C overnight before transport to the laboratory early the next day.

### **How should samples be processed in the microbiology laboratory?**

Following arrival in the laboratory, specimens should be processed as soon as practicable (ie, given similar priority to other sterile site samples). Handle each specimen separately. Perform manipulation of samples in a Biosafety cabinet Class 2 to protect the sample from contamination.

Homogenise tissue samples (eg, using mechanical tissue grinders and sterile equipment). Inoculate samples onto non-selective media (eg, blood and chocolate agar in CO<sub>2</sub> and blood +/- enriched anaerobic agar incubated in anaerobic conditions).

Perform cultures on both solid agar and in blood/broth cultures. Blood/broth culture has been shown to be more sensitive than solid media alone, however, mixed infections may be less easily detected with broth cultures. Blood/broth cultures must never be the only type of culture used; they are in addition to cultures on solid media.

The preferred enrichment broth for tissue samples is not defined, although Robertson's cooked meat or thioglycolate broth are commonly used.<sup>13,18</sup> Blood culture bottles are also suitable ([see above](#)).<sup>11,14,16</sup>

Blood culture bottles are the recommended enrichment media for joint aspirates. The bottles inoculated will depend on the volume of sample remaining after cell count, differential and inoculation of solid agar. Suggested division of samples by volume are as follows.

<b>Aspirate volume</b>	<b>Division of sample</b>
8–20 mL	Divide evenly between aerobic and anaerobic adult blood culture bottles or, if there are capacity limitations, an aerobic adult bottle
2–7 mL	Divide evenly between paediatric and anaerobic adult blood culture bottles or, if there are capacity limitations, an aerobic adult bottle
1 mL	Paediatric blood culture bottle or an adult aerobic bottle

### **How long should samples be cultured for?**

If no potential pathogen has been isolated after five days' incubation, incubate plates for a 7–10 days. Enrichment broths should be subcultured when visually turbid. Visually negative broths (except monitored blood culture bottles) should be terminally subcultured (eg, subcultured on day 4–5 with subcultured plates held for a further 7–10 days). Bemer et al found a combination of three media, with different duration incubation times, had the best diagnostic yield, ie, blood culture bottle (5 days), chocolate agar plate (7 days) and anaerobic broth (14 days).<sup>11</sup>

To assist with identifying mixed cultures, we suggest using selective agar (eg, CNA, MacConkey agar and selective anaerobic plates) for broth subcultures when the direct cultures have already grown staphylococci or Gram-negative bacilli.

### **Identification and susceptibility testing**

All organisms growing from prosthetic joint samples should be identified to species level.

When the same species is present in two or more samples, perform susceptibility testing.

### **Shoulder joint PJI and *P. acnes***

*P. acnes* is a common cause of shoulder joint PJI. While classified as an anaerobic Gram-positive bacillus, many strains are aerotolerant, eg, good recovery has been reported with chocolate agar incubated in CO<sub>2</sub>.<sup>11</sup> The replication time of *P. acnes* is much slower than common pathogens and cultures need to be held an adequate amount of time to ensure its isolation. When specimens are transported in air and agar, and brain heart infusion broth cultures held for 28 days, all true *P. acnes* isolates are recovered by 13 days.<sup>19</sup> Holding cultures longer only recovered contaminant *P. acnes* isolates. Shorter incubation is, however, possible if samples are transported in an anaerobic fluid transport medium (not in common use in New Zealand) and inoculated into anaerobic thioglycolate broth. Under these circumstances > 96 percent of cultures are positive at day seven.<sup>18</sup> Any culture approach should have a suitable medium incubated for an appropriate time to recover *P. acnes*. This is essential for samples from suspected shoulder PJI. In most instances this means at least 14 days' incubation of at least one medium.

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10. Parvizi J, Gehrke T, Chen AF. 2013. Proceedings of the International Consensus on Periprosthetic Joint Infection. *Bone Joint J* 95-B(11): 1450–2.
11. Bemmer P, Leger J, Tande D, et al. 2016. How many samples and how many culture media to diagnose a periprosthetic joint infection: a clinical and microbiological prospective multicenter study. *J Clin Microbiol* 54: 385–91.
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14. Peel TN, Dylla BL, Hughes JG, et al. 2016. Improved diagnosis of prosthetic joint infection by culturing periprosthetic tissue specimens in blood culture bottles. *mBio* 7:e01776-15. doi: 10.1128/nBio.01776–15.
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16. Hughes HC, Newnham R, Athanasou N, et al. 2011. Microbiological diagnosis of prosthetic joint infections: a prospective evaluation of four bacterial culture media in the routine laboratory. *Clin Microbiol Infect* 17: 1528–30.
17. Peel TN, Sedarski JA, Dylla L, et al. 2017. Laboratory workflow analysis of culture of periprosthetic tissues in blood culture bottles. *J Clin Microbiol* 55: 2817–26.
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## Appendix 1: Annotated bibliography of key references used for guidance on culture methods and sample number

11. Bemer P, Leger J, Tande D, et al. 2016. How many samples and how many culture media to diagnose a periprosthetic joint infection: a clinical and microbiological prospective multicenter study. *J Clin Microbiol* 54: 385–91.
- Prospective multicentre study, West France
  - Five samples cultured from patients suspected of having PJI
  - Samples disrupted by stainless steel beads using bead mill
  - Looked at yield of 2/3/4 samples vs 5 samples
  - Looked at yield 2/3 media vs 5 media
  - Modified IDSA microbiological criteria, only 1 culture had to be positive with strict pathogen, eg, *S. aureus*, Enterobacteriaceae, otherwise  $\geq 2$  specimens with indistinguishable organism
  - Mathematical modelling to estimate yield of differing specimen numbers
  - Cultures evaluated:
    - paediatric BC bottle, 1 mL, 14 d
    - Schaedler broth (SB) (an AnO<sub>2</sub> broth), 1 mL, 14 d then subculture
    - BA and Choc, 50  $\mu$ L, CO<sub>2</sub>, 7 d
    - BA AnO<sub>2</sub>, 50  $\mu$ L, 7 d

Sensitivity %	IDSA definition	Microbiological
2 samples	98	85.2
3 samples	99.2	93.9
4 samples	99.7	98.1

- SB far more sensitive than BA AnO<sub>2</sub> for recovering anaerobes
  - Paediatric BC best for aerobic culture
  - Many *P. acnes* grew on choc CO<sub>2</sub>
  - Best yield from 3 media: BC (5 d), choc (7 d), SB (14 d). More media didn't increase yield significantly
  - Conclusion: 'Four samples adequate'.
13. Atkins BL, Athanasou N, Deeks JJ, et al. 1998. Prospective evaluation of criteria for microbiological diagnosis of prosthetic-joint infection at revision arthroplasty. The OSIRIS Collaborative Study Group. *J Clin Microbiol* 36: 2932–9.
- Prospective, single reference centre study, Oxford
  - Evaluable episodes – 297
  - PJIs – 41 (14 percent)
  - Histopathology used as reference standard
  - Standard set of samples recommended to be taken
  - Tissue disrupted with glass beads
  - Aliquoted (volume not stated) onto 3 media: chocolate agar (Choc) (CO<sub>2</sub>), Blood agar (BA) (CO<sub>2</sub>) and anaerobic (AnO<sub>2</sub>), Robertson's cooked meat broth (RCMB)
  - Plates read for 7 days, RCMB subbed at 5 days if not cloudy before
  - Sixty-five percent of samples from PJIs were culture positive

- Mathematical modelling used to determine the number of samples needed to result in given number of positive specimens with indistinguishable organism
- The isolation of an indistinguishable organism in 3 or more specimens was highly predictive of infection (sens 65 percent, spec 99.6 percent, LR 169)
- Gram stain was insensitive (sens 12 percent, spec 98 percent, LR 10)
- Conclusion: 'We recommend that five or six specimens be sent, that the cutoff for a definitive diagnosis of infection be three or more specimens that yield an indistinguishable organism.'

14. Peel TN, Dylla BL, Hughes JG, et al. 2016. Improved diagnosis of prosthetic joint infection by culturing periprosthetic tissue specimens in blood culture bottles. *mBio* 7:e01776-15. doi: 10.1128/nBio.01776-15.

- Prospective single centre study, Mayo Clinic
- Compare sens/spec of blood culture set vs conventional approach, ie, thioglycolate broth (Thio) and agar plates
- Compared culture approaches vs IDSA definition ( $\geq 2$  specimens with indistinguishable organism)
- Also used Bayesian latent class (LCM) modelling, which assumes no gold standard, to compare culture approaches
- 369 patients
- 117 (32 percent) met IDSA definition for PJI, 83 percent chronic PJI
- Tissue homogenised in Stomacher®
- Standard culture: 0.1 mL BA + choc CO<sub>2</sub> (5 d), 0.1 mL BA AnO<sub>2</sub> (14 d), 1 mL Thio (14 d)
- Blood culture (BC) set: 1 mL each into BACTEC O<sub>2</sub> and AnO<sub>2</sub> bottles

Sensitivity (%)	BC set	Conventional
LCM	92	63
IDSA	61	44, p=0.003

- BCs were positive earliest,  $\leq 22$ hrs
- AnO<sub>2</sub> BC had a non-significant higher recovery of organisms vs IDSA definition, 48 percent vs 43 percent
- No O<sub>2</sub> BC positive after 7 d
- Extending AnO<sub>2</sub> bottle to 14 d picked up 3 true and 3 false *P. acnes* isolates
- Conclusion: 'BCs higher sensitivity for detection of PJIs.'

15. Peel TN, Spelman T, Dylla BL, et al. 2017. Optimal periprosthetic tissue specimen number for the diagnosis of prosthetic joint infection. *J Clin Microbiol* 55: 234-43.

- Prospective single centre study, Mayo Clinic
- Same data set as reference 14 above
- Modified MSIS definition; includes  $\geq 2$  specimens with indistinguishable organism
- Greatest accuracy for conventional culture, 91 percent, was with 4 specimens
- Greatest accuracy for BCs, 92 percent, was with 3 specimens

Sensitivity %	Conventional	BC
≤ 3 specimens	68	71
≤ 4 specimens	77	86

- Four specimens in BC had high sensitivity
  - Conclusion: 'Greatest accuracy of diagnosis of PJI was when 3 specimens were cultured into BCs. Increasing the number to ≥ 5, as currently recommended, does not improve the accuracy of diagnosis of PJI.'
16. Hughes HC, Newnham R, Athanasou N, et al. 2011. Microbiological diagnosis of prosthetic joint infections: a prospective evaluation of four bacterial culture media in the routine laboratory. *Clin Microbiol Infect* 17: 1528–30.
- Same Oxford group as for reference 13
  - Evaluable patients – 141
  - Mean number specimens/patient 4.9, mode 5
  - PJIs based on histopathology – 23
  - Four approaches; 3 liquid media and set of 4 agar media
  - 1 mL into: RCMB, fastidious anaerobic broth (FA), BACTEC O<sub>2</sub> + BACTEC AnO<sub>2</sub> bottles
  - 0.25 mL onto: Choc and BA CO<sub>2</sub>, BA AnO<sub>2</sub> one with one without a metronidazole disc
  - Ten BACTEC sets positive in 1 bottle only, use of both critical for 9 percent PJIs
  - Sensitivity: BACTEC set 87 percent, RCMB 83 percent, FA 57 percent, agar set 39 percent
  - Conclusion: 'Use of BACTEC for these specimens is attractive.'

## Appendix 2: Example equipment list for a PJI sampling pack

<b>Instrument</b>	<b>Quantity</b>
Russian Mayo Forceps	5
BP Handle NO: 4 L	3
Mayfield Bone Rongeur	1
Rushkin Bone Rongeur	1
Hatt Spoon Curette, large 20 mm	1
Hatt Spoon Curette, small 10 mm	1

### Appendix 3: Periprosthetic joint infection sampling form (example)

<b>Peri-prosthetic Joint Sample Form</b>		Patient ID label here
	Time	Taken by
Lab Use Only	Antibiotic prophylaxis withheld before sampling: (circle) Yes                  No	
<b>B. Joint: (circle)</b> Left    Right Hip    Knee Other _____		<b>A. Sample number: (circle)</b> 1      2      3      4      5 Other _____
<b>D. Sample type: (circle)</b> Tissue          Aspirate Other _____		<b>C. Sample site: (circle)</b> Peri-prosthetic membrane Pseudocapsule/deep tissue Superficial Other _____
<b>E. Tests</b> Routine MC&S <input type="checkbox"/>		<b>Clinical details:</b> (no need to be repeat for samples 2-5)
Other _____		Other _____
Requestor:		Contact No:



Peri-prosthetic Joint Sample Form