False-positive Cultures After Native Knee Aspiration: True or False

Jason M. Jennings MD, DPT, Douglas A. Dennis MD, Raymond H. Kim MD,
Todd M. Miner MD, Charlie C. Yang MD, David C. McNabb MD

Published online: 9 December 2016
© The Association of Bone and Joint Surgeons® 2016

Abstract

Background Synovial fluid aspiration is a routine practice used by most orthopaedic surgeons to aid in the diagnosis of joint infection. In patients for whom there is a low pretest probability of infection, a positive culture—particularly if it is a broth-only culture—may be considered a contaminant, especially if the bacterial species are skin pathogens. To our

Each author certifies that he, or a member of his immediate family, has no funding or commercial associations (eg, consultancies, stock ownership, equity interest, patent/licensing arrangements, etc) that might pose a conflict of interest in connection with the submitted article.

All ICMJE Conflict of Interest Forms for authors and *Clinical Orthopaedics and Related Research*® editors and board members are on file with the publication and can be viewed on request. *Clinical Orthopaedics and Related Research*® neither advocates nor endorses the use of any treatment, drug, or device. Readers are encouraged to always seek additional information, including FDA-approval status, of any drug or device prior to clinical use. Each author certifies that his institution approved the human protocol for this investigation, that all investigations were conducted in conformity with ethical principles of research, and that informed consent for participation in the study was obtained. This work was performed at Colorado Joint Replacement, Denver, CO, USA.

J. M. Jennings, D. A. Dennis (✉), R. H. Kim, T. M. Miner,
C. C. Yang
Colorado Joint Replacement, 2535 S Downing Street, Suite 100,
Denver, CO 80210, USA
e-mail: roseannjohnson@centura.org

D. A. Dennis, R. H. Kim
Department of Biomedical Engineering, University of Denver,
Denver, CO, USA

D. A. Dennis
Department of Orthopedics, University of Colorado School of
Medicine, Aurora, CO, USA

knowledge no study has evaluated the incidence of contamination of aspirations from the native knee.

Questions/purposes What is the frequency of false-positive cultures among knee aspirations of the native knee?

Methods Two hundred patients, with a total of 200 knees, with the diagnosis of degenerative osteoarthritis undergoing a total knee arthroplasty (TKA) were identified for this study. None of these patients had symptoms, signs, or laboratory studies to suggest the presence of joint infection; a positive culture in this population therefore would be considered contaminated. Thirty-two (16%) patients were excluded secondary to a dry aspiration. One patient was enrolled in the study but did not have the knee aspirated and another patient's specimen was accidentally discarded. Each knee was aspirated under sterile conditions before performing the TKA. The fluid was sent for cell count and culture. If insufficient fluid was obtained for both cell count and culture, culture was performed rather than cell count.

Results There were no false-positive cultures (zero of 166 [0%]) in aspirations of native knees.

Conclusions Our study would indicate that when done properly under sterile technique, cultures taken from knee arthrocentesis in patients without prosthetic joints should

D. A. Dennis
Department of Biomedical Engineering, University of
Tennessee, Knoxville, TN, USA

R. H. Kim
Department of Orthopedic Surgery, Marshall University,
Huntington, WV, USA

D. C. McNabb
Raleigh Orthopedic Clinic, Raleigh, NC, USA

not be affected by perceived contaminant species. A positive specimen finding on culture should raise a strong suspicion of bacterial septic arthritis. Future studies should include more specimens as well as knees with prior TKA to help further identify the rate of false-positive cultures in knee arthrocentesis in both populations.

Level of Evidence Level I, diagnostic study.

Introduction

Synovial fluid aspiration is a routine practice used by most orthopaedic surgeons to aid in the diagnosis of joint infection. A preoperative aspiration of the knee has been advocated as the most helpful study for the diagnosis or exclusion of infection in the setting of revision total knee arthroplasty (TKA) [4]. Recently, the Musculoskeletal Infection Society developed a consensus statement [16] addressing the definition and diagnosis of a periprosthetic joint infection (Table 1). Two positive cultures (on two separate samples) are a major criterion and one positive culture qualifies as a minor criterion. Within the native knee setting, elevated white blood cell (WBC) count above 50,000 and/or a positive culture suggests a diagnosis of septic arthritis. In the absence of an elevated WBC count, a positive bacterial culture can produce a diagnostic dilemma. Should the clinician accept the culture result, reaspirate the joint, or consider the result to be falsely positive?

One concern is the potential for a false-positive aspiration secondary to contamination at the time of the arthrocentesis or in the microbiology laboratory. The incidence of false-positive intraoperative cultures in the setting of revision total hip and knee arthroplasty has a wide range of variability ranging from 4% to 96% [1, 2, 4, 9, 10, 18, 19]. In patients with low suspicion for infection and a low burden of bacteria discovered on culture, these findings may be considered a contaminant of the

aspiration or culture process, especially if the bacterial species are skin pathogens. To our knowledge, the false-positive incidence from a native knee aspiration in a large series has not been described.

Therefore, we asked: What is the frequency of false-positive cultures among knee aspiration of the native knee?

Patients and Methods

Approval for this study was obtained from our institution's ethical committee. Informed consent was obtained from patients before participation in this study. We prospectively evaluated the synovial fluid of patients with the diagnosis of degenerative osteoarthritis undergoing a primary TKA from February to July 2015. Exclusion criteria included patients who were immunocompromised, had a history of inflammatory arthritis or autoimmune disease, a prior knee infection, or current major or minor illness. Two hundred patients were enrolled in this prospective study, yielding 200 knees. The mean age of the subjects was 66 years (range, 33-95 years), and 92 (46%) were men. Thirty-two (16%) patients were excluded secondary to a dry aspiration. One patient was enrolled in the study but did not have the knee aspirated, and another patient's specimen was accidentally discarded. This left 166 patients remaining who had adequate fluid for culture analysis. Of these 166 patients, 17 (10%) did not have a sufficient quantity for both culture and cell count; thus, culture only was obtained in these patients.

In the operating room, the knee was cleaned with chlorhexidine wipes and then sterilely prepped with Duraprep® (3M, St Paul, MN, USA) and allowed to dry. The surgeon performing the aspiration was sterilely gloved and gowned with body exhaust suits at the time of aspiration. Before application of the iodophor-impregnated drape, an 18-gauge spinal needle with a stylet was placed into the knee through the superior lateral parapatellar position, the stylet was removed, and then the knee was aspirated. Immediately after aspiration, synovial fluid was applied on a leukocyte esterase strip as part of a previous study [11]. The remaining fluid was sent for synovial fluid WBC count and culture. If only minimal fluid was available, culture was preferentially obtained over cell count and fluid analysis. The synovial fluid was plated in aerobic conditions on sheep agar, chocolate agar, and MacConkey agar plates, which were held at 35° to 37° C. The anaerobic conditions included a Brucella blood agar with kanamycin and Vancomycin, both held in an anaerobic pouch at 35° to 37° C. All cultures were held for 14 days. We then calculated the percentage of potential synovial fluid aspirate contamination in patients undergoing primary TKA.

Table 1. Musculoskeletal Infection Society periprosthetic infection criteria

1. Major
a. Presence of sinus tract
b. Two positive cultures on two separate samples
2. Minor (three of five needed)
a. Erythrocyte sedimentation rate > 30 mm/hr and C-reactive protein > 10 mg/L
b. Synovial leukocyte count > 3000 cell/μL or ++ leukocyte esterase reagent testing
c. Synovial neutrophil > 65%
d. One positive culture
e. Greater than 5 neutrophils in five high-powered fields at × 400 magnification

Results

In all 166 specimens, cultures were negative. There were no false-positives. The average synovial fluid cell count (148 specimens) for the patient population in this study was 306 (range, 24–5030). Of the 149 available samples that allowed for leukocyte esterase testing, there was one false-positive. The synovial fluid WBC count was 290 and the culture remained negative without growth at 14 days in this patient.

Discussion

False-positive cultures from knee arthrocentesis in the setting of TKA have been described in previous studies [4, 19]. However, little is known about the incidence of potential contamination in the evaluation of native knee synovial fluid where the presumed pretest probability is approaching zero. We therefore conducted this study to determine the frequency of false-positive cultures among synovial fluid specimens of the native knee. Our results suggest in this patient population, when performed appropriately under sterile conditions, a positive culture should raise a strong suspicion of bacterial septic arthritis.

The results should be interpreted in light of limitations. Because all aspirations and cultures were performed at one facility, the variance in false-positive rates between microbiology laboratories could not be tested or evaluated in this study. This limits the generalizability of these results to other laboratories. As a result of the fact that the total number of positive cultures was zero, rather than an unexpectedly high rate, the authors feel our results were accurate for this study population. The number of subjects enrolled is also a limiting factor of this study. False-negative aspirations in a native joint population have no historical rate; therefore, addressing this study's ability to avoid a type II error is difficult. If a presumed rate of false-negative cultures on aspiration was 0.5%, then this study with its 166 specimens would be underpowered. To date, however, this remains the largest reported data set on culture results in native knee arthrocentesis. Lastly, we are unsure of the generalizability of these findings in patients with a TKA.

In an environment where the incidence of infection should be approaching zero, this study has shown that when done in a sterile environment, the probability of a false-positive culture in a synovial fluid aspiration of the native knee is rare. To our knowledge, this is the largest study population to demonstrate this finding in native knees. A positive specimen finding on culture should raise a strong suspicion of bacterial septic arthritis. Previous studies have reported false-positive joint aspirations rates

between 4% and 96% [1, 2, 4, 9, 10, 18, 19], but these studies were performed in a revision arthroplasty setting. Cultures that are positive as a result of common skin flora are more likely to be considered false-positives than more virulent and pathogenic organisms. *Propionibacterium acnes* is a common skin pathogen that is indicated as the cause of infection in open shoulder surgery [7, 8, 12]. *Staphylococcus aureus* and coagulase-negative *Staphylococcus* such as *Staphylococcus epidermidis* are both common skin flora but are also found in 55% to 70% of native and periprosthetic joint infections [13, 15]. *Streptococcus* species are also common skin flora but also rank second at 20% to 29% of native and periprosthetic joint infection pathogens [13, 15]. In this study we held all cultures for 14 days in an attempt to capture all organisms, particularly those of low virulence.

Various formulations can be used for surgical or procedure site skin preparation. Prior studies have revealed that alcohol-based solutions with emphasis on the use of chlorhexidine preparation shows a lower percentage of positive skin surface cultures [3, 5, 14, 17]. However, isopropyl alcohol has been shown to be the most important component with regard to surgical site skin preparation [5]. In this study, chlorhexidine was used to clean the leg before the sterile preparation with one alcohol-based solution, Duraprep®. An additional culprit in false-positive cultures could lie within the needle, because needles whether with and without a stylet have been shown to introduce skin plugs and even hair in the joint [6, 20]. This study was performed with an 18-gauge spinal needle with a stylet in place until the needle was in the knee.

The idea of a false-positive culture rests solely in an unexpected positive culture, usually of a skin flora pathogen, wherein other data available would suggest a diagnosis contrary to septic arthritis. Our findings suggest that the likelihood of false-positive cultures in aspirations of native knees performed under sterile conditions is exceedingly low, and a positive culture should raise a strong suspicion for septic arthritis. Additionally, we feel that liberal use of alcohol-based skin preparation may aid in reducing contamination during the process of arthrocentesis. Future studies should include a multicenter analysis to allow for more overall specimens lowering the risk of type II error and to explore the potential variance with laboratory error. Ideally specimens from patients with asymptomatic TKA would give a true false-positive rate in this patient population, but the risk of the potential introduction of a periprosthetic infection remains a concern.

Acknowledgments We thank Rose Johnson, Daniel Levy, and Catie Eschen for all of their help with data collection, manuscript formatting, and submission of this manuscript. We also thank the entire office staff and clinicians at Colorado Joint Replacement for their continued efforts in research.

References

1. Barrack RL, Aggarwal A, Burnett RSJ, Clohisey JC, Ghanem E, Sharkey P, Parvizi J. The fate of the unexpected positive intraoperative cultures after revision total knee arthroplasty. *J Arthroplasty*. 2007;22:94–99.
2. Barrack RL, Harris WH. The value of aspiration of the hip joint before revision total hip arthroplasty. *J Bone Joint Surg. Am*. 1993;75:66–76.
3. Bibbo C, Patel D V, Gehrmann RM, Lin SS. Chlorhexidine provides superior skin decontamination in foot and ankle surgery: a prospective randomized study. *Clin Orthop Relat Res*. 2005;438:204–208.
4. Duff GP, Lachiewicz PF, Kelley SS. Aspiration of the knee joint before revision arthroplasty. *Clin Orthop Relat Res*. 1996;331:132–139.
5. Dumville JC, McFarlane E, Edwards P, Lipp A, Holmes A, Liu Z. Preoperative skin antiseptics for preventing surgical wound infections after clean surgery. *Cochrane Database Syst Rev*. 2015;4:CD003949.
6. Glaser DL, Schildhor JC BA. The inadvertent introduction of skin into the joint during intra-articular knee injections: do you really know what is on the tip of your needle? *Proc Am Acad Orthop Surg*. 2001;68:130–131.
7. Horneff JG, Hsu JE, Huffman GR. *Propionibacterium acnes* infections in shoulder surgery. *Orthop Clin North Am*. 2014;45:515–521.
8. Hudek R, Sommer F, Kerwat M, Abdelkawi AF, Loos F, Gohlke F. *Propionibacterium acnes* in shoulder surgery: true infection, contamination, or commensal of the deep tissue? *J Shoulder Elbow Surg*. 2014;23:1763–1771.
9. Klatte TO, Meinicke R, O'Loughlin P, Rueger JM, Gehrke T, Kendoff D. Incidence of bacterial contamination in primary THA and combined hardware removal analysis of preoperative aspiration and intraoperative biopsies. *J Arthroplasty*. 2013;28:1677–1680.
10. Lachiewicz PF, Rogers GD, Thomason HC. Aspiration of the hip joint before revision total hip arthroplasty. Clinical and laboratory factors influencing attainment of a positive culture. *J Bone Joint Surg Am*. 1996;78:749–754.
11. McNabb DC, Dennis DA, Kim RH, Miner TM, Yang CC, Jennings JM. Determining false positive rates of leukocyte esterase reagent strip when used as a detection tool for joint infection. *J Arthroplasty*. 2016 Jun 7. [Epub ahead of print]
12. Mook WR, Klement MR, Green CL, Hazen KC, Garrigues GE. The incidence of propionibacterium acnes in open shoulder surgery: a controlled diagnostic study. *J Bone Joint Surg Am*. 2015;97:957–963.
13. Nolla JM, Lora-Tamayo J, Gómez Vaquero C, Narváez J, Murillo O, Pedrero S, Cabo J, Ariza J. Pyogenic arthritis of native joints in non-intravenous drug users: a detailed analysis of 268 cases attended in a tertiary hospital over a 22-year period. *Semin Arthritis Rheum*. 2015;45:94–102.
14. Ostrander R V, Botte MJ, Brage ME. Efficacy of surgical preparation solutions in foot and ankle surgery. *J Bone Joint Surg Am*. 2005;87:980–985.
15. Parvizi J, ed. *Periprosthetic Joint Infection: Practical Management Guide*. 1st ed. New Dehli, India: Jaypee Brothers Medical Publishers (P) Ltd; 2013.
16. Parvizi J, Gehrke T. Definition of periprosthetic joint infection. *J Arthroplasty*. 2014;29:1331.
17. Saltzman MD, Nuber GW, Gryzlo SM, Marecek GS, Koh JL. Efficacy of surgical preparation solutions in shoulder surgery. *J Bone Joint Surg Am*. 2009;91:1949–1953.
18. Spangehl MJ, Masri BA, O'Connell JX, Duncan CP. Prospective analysis of preoperative and intraoperative investigations for the diagnosis of infection at the sites of two hundred and two revision total hip arthroplasties. *J Bone Joint Surg Am*. 1999;81:672–683.
19. Van den Bekerom MPJ, Stuyck J. The value of pre-operative aspiration in the diagnosis of an infected prosthetic knee: a retrospective study and review of literature. *Acta Orthop. Belg*. 2006;72:441–447.
20. Wahl K, Adams SB, Moore GE. Contamination of joints with tissue debris and hair after arthrocentesis: the effect of needle insertion angle, spinal needle gauge, and insertion of spinal needles with and without a stylet. *Vet Surg*. 2012;41:391–398.