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BASIC SCIENCE

Presence of *Propionibacterium acnes* in primary shoulder arthroscopy: results of aspiration and tissue cultures

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Background: Infection after shoulder surgery has a serious impact on patient outcome and costs associated with care. *Propionibacterium acnes* infection may be insidious and manifest years after index surgery with resultant joint arthropathy or prosthesis infection. Our goal was to evaluate the presence of *P. acnes* in a group of patients undergoing primary arthroscopic shoulder surgery to better understand this organism.
Methods: Samples were collected from 57 patients undergoing first-time shoulder arthroscopy. Demographic data and medical comorbidities were collected. A control, 2 skin swabs, synovial fluid, and 3 tissue samples were obtained. All samples were placed on aerobic plates, on anaerobic plates, and in thioglycolate broth and held for 28 days.
Results: Fifty-seven patients underwent arthroscopic shoulder surgery. The mean age was 51 years. Eighty-one samples (21.8%) were positive for *P. acnes* when cultures were held 14 days; 32 subjects (56%) had at least 1 culture that grew *P. acnes*. Positive skin cultures for *P. acnes* increased from 15.8% before incision to 40.4% at closure. This was even more pronounced in men as positive skin cultures increased from 31.3% before incision to 63.0% at closure. Thirteen patients (22.8%) had more than 3 cultures positive. None of the patients in this study have had signs or symptoms to suggest clinical *P. acnes* infection.
Conclusions: Of all subjects studied, 56% had at least 1 positive culture; 21% (of all 371 culture specimens obtained) grew *P. acnes*. We suspect that it is a consequence of true positive cultures from imperfect skin preparation and dermal contamination.
Level of evidence: Basic Science Study, Microbiology.
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Keywords: Shoulder; infection; *P. acnes*; culture; arthroscopy; aspiration

Propionibacterium acnes, a gram-positive anaerobic bacillus, resides nonpathogenically in the sebaceous glands associated with hair follicles.[8,9,13,14,16,18,22,23] This organism is a frequent pathogen in infections after arthroscopic

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and open surgical shoulder procedures.[1,3,6,8,9-11,13,18,21,24] When it is unrecognized or untreated, indolent infection with *P. acnes* may result in persistent shoulder pain and glenohumeral arthrosis, sometimes many years after the index procedure.[13] The diagnosis of *P. acnes* infection is challenging, given the slow growth rate and a lack of consensus about how to identify a true infection.[7,8,13]

Because this bacterium provides such significant challenges in shoulder surgery, we sought to identify risk factors for *P. acnes* infection and to take measures to limit these risks. One potential area for improvement is to address surgical skin preparation; the skin has residual *P. acnes* in 7% of cases, and the dermal layer has been suggested to be an important source of wound contamination.[15,19] Studies have demonstrated the presence of *P. acnes* on the skin as well as in arthroplasty and revision arthroplasty surgeries.[15,16,18] The purpose of our study was to evaluate the presence of *P. acnes* on surgically cleansed skin, in glenohumeral aspiration fluid, and in tissue culture specimens from shoulders previously not operated on. Our hypothesis was that *P. acnes* would not be found at a high rate in patients who had not previously had shoulder surgery.

Materials and methods

Sterile skin swabs, aspirated joint fluid, and tissue specimens were collected from June 2013 to September 2013 from 57 patients undergoing shoulder arthroscopy without any previous shoulder surgery. Demographic data, visual analog scale (VAS) for pain, and medical comorbidities were also collected. Our inclusion criterion was shoulder disease necessitating primary shoulder arthroscopy. Our exclusion criteria were previous shoulder surgery and antibiotic treatment within 4 weeks before surgery.

All patients consented to participate in this study. Preoperatively, 2 g of cefazolin was routinely given, or 900 mg of clindamycin if cephalosporin allergy was present. Patients were draped and sanitized before surgery in our routine institutional method by scrubbing the arm, shoulder, and axilla with a scrub brush containing 3.3% chloroxylenol cleansing solution. The skin was then patted dry with a sterile towel. The surgical site was prepared with 3 applications of 2% chlorhexidine gluconate (ChloraPrep; CareFusion, San Diego, CA, USA). After preparations and draping, the skin of the anterior deltoid at the site of the anterior arthroscopic portal was swabbed with a skin swab. This skin swab was then placed into a charcoal medium.

After incision for trocar placement, the glenohumeral joint was aspirated. If no fluid was available, the glenohumeral joint was flushed with 5 mL of saline, which was then collected in a sterile specimen container. Three samples of débrided tissue were collected through a cannula. The first tissue sample was collected from the middle glenohumeral ligament. The second tissue sample came from the rotator interval and the third tissue sample from the bursa. A sample of the cuff was taken instead of the bursa in patients with torn rotator cuffs. For a patient undergoing a labrum repair, the second tissue sample was collected from the high rotator interval and the third tissue sample from the low rotator interval.

For 47 patients, a second skin swab was collected from the anterior deltoid at the site of the anterior portal at the conclusion of the surgical procedure; we added this culture after the study had started. This skin swab was also placed into a charcoal medium. All tissue samples were placed in sterile specimen containers. Each sample container was placed in an individual sterile transport bag and transported to the laboratory within 1 hour of collection.

All samples for each patient were delivered to the microbiology laboratory within 1 hour, where they were placed in a fume hood that had been sterilized with 1.4% hydrogen peroxide spray for 1 minute. All samples were processed by medical technicians following sterile procedure. Each sample was plated on a blood agar plate to facilitate the growth of any organism, a MacConkey agar plate to select for gram-negative rods, a colistin and nalidixic acid plate to identify gram-positive organisms, and a CDC blood agar plate kept in anaerobic conditions with a gentamicin disk (ANA CDC) to select for anaerobic organisms. Each sample was also placed in a test tube of thioglycolate broth to encourage the growth of any organisms present. Microbiology laboratory procedures were followed according to Figure 1 and detailed there.

Each control and skin swabs were removed from their transport bag and streaked across a microscope slide to check immediately for contaminating organisms with a Gram stain. The swabs were next streaked across the first quadrant of each agar plate and were also placed in a test tube of thioglycolate broth and wrapped in parafilm. Individual, disposable sterile loops were used to streak the remaining quadrants of each plate.

One drop of the aspirate joint fluid sample was transferred to each agar plate as well as to a microscope slide for immediate analysis. Four or 5 drops of joint fluid were transferred to thioglycolate broth test tubes. Each drop of joint fluid was streaked across the agar plates with individual, disposable sterile loops.

Each tissue sample was added with 1 mL of thioglycolate broth to a sterile tissue grinder (Precision Disposable Tissue Grinder; Covidien, Dublin, Ireland) and ground for 1 minute or until homogenized. One drop of the dissolved tissue sample was transferred to each agar plate and to a microscope slide. Individual, disposable sterile loops were used to streak the agar plates. Four or 5 drops of the tissue sample were then placed into a test tube of thioglycolate broth.

The blood agar plate, MacConkey agar plate, and colistin and nalidixic acid plate were incubated aerobically at 37°C for 48 hours and then checked for growth of bacteria. The ANA CDC plates with gentamicin disks were kept in sterile bags with a carbon dioxide pouch system to maintain anaerobic conditions. Each sterile bag included an oxygen indicator to ensure that anaerobic conditions were met. The ANA CDC plates were incubated at 37°C for 7 days and then analyzed for *P. acnes*. Thioglycolate broth test tubes were sealed with parafilm to protect against contamination. These samples were incubated at 37°C for 28 days or until a positive *P. acnes* diagnosis could be made. All cultures were checked daily for bacterial growth.

Bacterial colonies suspected of being *P. acnes* were smeared onto microscope slides and Gram stained. Isolated *P. acnes* was tested for biotype with a MicroScan Rapid Anaerobe ID (MicroScan; Siemens, Erlangen, Germany). Isolated *P. acnes* was then streaked on brucella agar/ANA CDC agar to measure hemolysis. Hemolysis was defined as when there was at least 2 mm of hemolysis around bacterial colonies.[17]

A positive culture was defined by growth on the anaerobic plate within 7 days or if the thioglycolate broth became turbid and subtyped as *P. acnes* within 28 days.

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