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Intraoperative Synovial C-reactive Protein Is as Useful as Frozen Section to Detect Periprosthetic Hip Infection

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Abstract

Background Synovial quantification of C-reactive protein (SCRP) has been recently published with high sensitivity and specificity in the diagnosis of periprosthetic joint infection. However, to our knowledge, no studies have compared the use of this test with intraoperative frozen section, which is considered by many to be the best intraoperative test now available.

Questions/purposes We asked whether intraoperative SCRP could lead to comparable sensitivity, specificity, and predictive values as intraoperative frozen section in revision total hip arthroplasty.

Methods A prospective study was performed including 76 patients who underwent hip revision for any cause. SCRP quantification (using 9.5 mg/L as denoting infection) and the analysis of frozen section of intraoperative samples (five or more polymorphonuclear leukocytes under high magnification in 10 fields) were performed in all the patients. The definitive diagnosis of an infection was determined according to the Musculoskeletal Infection Society (MSIS). In this group, 30% of the patients were diagnosed with infection using the MSIS criteria (23 of 76 patients).

Results With the numbers available, there were no differences between SCRP and frozen section in terms of their ability to diagnose infection. The sensitivity of SCRP was 90% (95% confidence interval [CI], 70.8%–98.6%), the specificity was 94% (95% CI, 84.5%–98.7%), the positive predictive value was 87% (95% CI, 66.3%–97%), and the negative predictive value was 96% (95% CI, 87%–99.4%); the sensitivity, specificity, positive predictive value, and negative predictive value were the same using frozen sections to diagnose infection. The positive likelihood ratio was 16.36 (95% CI, 5.4–49.5), indicating a low probability of an individual without the condition having a positive test, and the negative likelihood ratio was 0.10 (95% CI, 0.03–0.36), indicating low probability of an individual without the condition having a negative test.

Conclusions We found that quantitative SCRP had similar diagnostic value as intraoperative frozen section with comparable sensitivity, specificity, and predictive value in a group of patients undergoing revision total hip arthroplasty. In our institution, SCRP is easier to obtain, less expensive, and less dependent on the technique of obtaining and interpreting a frozen section. If our findings are confirmed by other groups, we suggest that quantitative SCRP be considered as a viable alternative to frozen section.

Level of Evidence Level I, diagnostic study.

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Introduction

Periprosthetic joint infection (PJI) is one of the most frequent and devastating complications of total joint arthroplasty. Sometimes, the distinction between mechanical loosening and infection is not clear. However, this is of paramount importance because the treatments are so different. The surgeon has a number of different tools to diagnose an infection, including history, clinical findings, radiographs, scintigraphy, blood studies, and aspiration. The sensitivity and specificity of the different methods have been questioned, ranging from 37% to 100% and from 83% to 100%, respectively, and no single laboratory test accurately detects infection before revision arthroplasty [4, 9–11, 16, 17, 19] (Table 1).

Intraoperative analysis of frozen sections is commonly used to diagnose periprosthetic infection [1, 2, 6, 12]. We have been using it in our service for 30 years and have previously reported frozen section was in agreement with the observations on standard histology in 134 of 136 cases [12]. However, frozen section is not a universally accepted method, results depend on the tissue that has been taken by the surgeon, and it requires a pathologist trained in musculoskeletal diagnosis. Such a specialist may not always available, in particular given that many PJI-related procedures at many institutions are performed toward the end of the surgical day. By contrast, synovial C-reactive protein (SCRP) is a simple, inexpensive test that has shown sensitivity of 85% with 95% specificity at a threshold of 9.5 mg/L in 55 revision hip and knee procedures in one series [14]. However, SCRP has not been widely used to detect infection, is relatively unknown, is nonspecific, and may increase in response to several diseases with acute inflammatory reactions and so comparing these two diagnostic tests is potentially important.

We therefore asked whether intraoperative SCRP could lead to comparable sensitivity, specificity, and predictive values as intraoperative frozen section in revision THA.

Patients and Methods

We studied 76 patients with a THA undergoing reoperation or revision surgery between November 2011 and December 2012. We excluded patients with chronic inflammatory diseases (three patients), Paget's disease (one patient), and immunodeficiency syndromes (one patient), because the SCRP level is reportedly elevated in these conditions [15]; apart from those exclusions, the study cohort represented all patients undergoing all revision surgeries during the study period. The study group included 43 men and 33 three women with a mean age of 67 years (range, 31–90 years). The study was approved by our institutional review board, and the patients gave informed consent. Revision surgery was indicated as a result of infection in 38 cases (débridement and retention, first- or second-stage reimplantation surgery), aseptic loosening in 27 cases, recurrent dislocation in seven cases, and periprosthetic fractures in four cases. All patients except six had undergone their original surgery elsewhere.

Synovial fluid for SCRP detection was taken with a 14-G intravenous needle before opening the capsule and immediately sent to the laboratory. Results generally were available after 20 minutes. Determination of high-sensitivity SCRP was performed with LX20-Beckman Coulter (Beckman Coulter, Brea, CA, USA) instruments using

Table 1. Analysis of diagnostic parameters according to different authors

Study	Diagnostic parameter	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Schinsky et al. [16]	ESR and CRP	90	91	95	82
Schinsky et al. [16]	Aspiration	82	83	69	90
Tohtz et al. [20]	Soft tissue culture	37	91	67	77
Nuñez et al. [12]	FS	98	99	98	99
Buttaro et al. [5]	IL-6 and CRP	57	100	100	94
Parvizi et al. [14]	SCRP	85	95	NR	NR
Parvizi et al. [15]	α-defensin and SCRP	99	100	NR	NR
Tetreault et al. [19]	sCRP	97	76	60	99
Omar et al. [13]	sCRP	95	93	NR	NR
Omar et al. [13]	WBC	85	86	NR	NR
Omar et al. [13]	PMN	90	90	NR	NR
Present study	SCRP and FS	90	94	87	96

PPV = positive predictive value; NPV = negative predictive value; ESR = erythrocyte sedimentation rate; CRP = C-reactive protein; FS = frozen section; IL-6 = interleukin-6; SCRP = synovial C-reactive protein, sCRP = serum C-reactive protein; WBC = white blood cell count; PMN = polymorphonuclears; NR = not reported.

turbidimetry and SCRP Beckman Coulter reactives (Beckman Coulter). This reactive is based on a high-sensitivity immunoassay kinetic infrared method. The particles covered with antibody anti-SCRP are bound to the patient's SCRP, forming nonsoluble aggregates that cause turbidity. The analytic sensitivity is defined as the less measurable concentration that can be distinguished from zero with a confidence of 95% and for the determination of SCRP is 0.02 mg/dL (0.2 mg/L).

Two to five samples of tissues to be analyzed were taken during surgery from the pseudocapsule, the cement-bone interface of the femur and acetabulum, and any other tissues involved according to the surgeon's judgment. All samples were referred for frozen section and to be processed on a routine basis [12] and were analyzed by the pathology unit of our hospital. Histological analysis was performed on all material. Smear tissue staining was hematoxylin-phloxine. Serial sections (4 mm thick) were processed in a freezing chamber and stained with hematoxylin-phloxine and toluidine blue. The material for standard processing was fixed in 10% formol and paraffin and embedded in an automatic tissue processor. Six microsections stained with hematoxylin-eosin and Mason's trichrome were observed with an optical microscope under high magnification (total magnification $\times$ 400) and a polarized light lens (Axiostar; Zeiss, Oberkochen, Germany). All samples were analyzed by two pathologists (MD, HGR) who counted the number of cells in 10 fields. Neutrophils, lymphocytes, plasma cells, macrophages, multinuclear giant cells, acrylic material, and polystyrene particles were included. For the specific purposes of this study, only neutrophils were considered. The limit to considering a sample to be infected was five or more polymorphonuclear leukocytes in 10 fields [11].

The definitive diagnosis of an infection was determined according to the Musculoskeletal Infection Society (MSIS) on the basis of two positive periprosthetic cultures with phenotypically identical organisms, a sinus tract communicating with the joint, or having three of the following minor criteria: elevated serum C-reactive protein (CRP) and erythrocyte sedimentation rate, elevated synovial fluid white blood cell count, elevated synovial fluid polymorphonuclear neutrophil percentage, a positive histological analysis of periprosthetic tissue, or a single positive culture [15]. In this group, 30% of the patients were diagnosed with infection using the MSIS criteria (23 of 76 patients; 95% confidence interval [CI], 12–40; Table 2). In 21 patients, infection was suspected and in two patients, it was not.

The threshold laboratory value to be positive for infection was assigned for the SCRP level (9.5 mg/L) to determine the sensitivity, specificity, positive predictive value, and negative predictive value, as previously published by Parvizi et al. [14].

Table 2. Type of procedures performed during revision surgery

Procedure	Number of cases
Both-component exchange	28
Reimplantation surgery	15
One-component exchange	12
Polyethylene exchange	1
Spacer	14
Débridement and retention	6

The functional sensitivity was defined as the lowest measurable concentration with a deviation between essays of 20% [18]. The estimated functional sensitivity is $\leq$ 0.018 mg/dL ($\leq$ 0.18 mg/L). The cost for each test in our hospital was USD 19.

Statistical analysis expressed infection prevalence, sensitivity, specificity, positive and negative predictive values, and positive and negative likelihood ratio in percentages with confidence intervals of 95%. The positive likelihood ratio indicates the probability of an individual without the condition having a positive test, and the negative likelihood ratio expresses the probability of an individual without the condition having a negative test.

The ability of the method was expressed with the area under the curve with confidence intervals of 95%. A multivariate statistical analysis was performed using the SPSS Version 19.0 program (SPSS, Inc, Chicago, IL, USA). Results were analyzed with the t-test and Pearson's chi-square according to the type of variable, considering a statistically significant $p \leq 0.05$.

Results

With the numbers available, there were no differences between SCRP and frozen section in terms of their ability to diagnose infection (Tables 3, 4). At a threshold of SCRP of 9.5 mg/L, the sensitivity was 90% (95% confidence interval [CI], 70.8%–98.6%), the specificity was 94% (95% CI, 84.5%–98.7%), the positive predictive value was 87% (95% CI, 66.3%–97%), and the negative predictive value was 96% (95% CI, 87%–99.4%); the sensitivity, specificity, positive predictive value, and negative predictive value were the same using frozen sections to diagnose the presence of infection.

The positive likelihood ratio was 16.36 (95% CI, 5.4–49.5), indicating a low probability of an individual without the condition having a positive test, and the negative likelihood ratio was 0.10 (95% CI, 0.03–0.36), indicating a low probability of an individual without the

Table 3. Culture results

Pathogen	Number of cases
MSSA	5
CNS	4
MRSA	3
Staphylococcus epidermidis	3
Enterococcus faecalis	2
Pseudomonas aeruginosa	1
Corynebacterium	1
Enterobacter cloacae	1
Negative	3

MSSA = methicillin-sensitive *Staphylococcus aureus*; CNS = coagulase-negative Staphylococcus; MRSA = methicillin-resistant *S aureus*.

Table 4. Results comparing SCRP with definitive pathology

Positive SCRP/histology	Infection present	Infection absent	Total
Positive test	20	4	24
Negative test	2	50	52
Total	22	54	76

SCRP = synovial C-reactive protein.

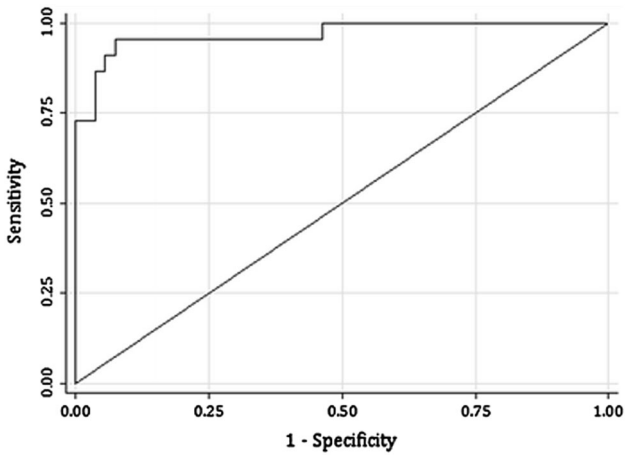

Fig. 1 Receiver operator curve showing that SCRP provided similar sensitivity, specificity, and positive predictive values as frozen section.

condition having a negative test. The area under the curve was 0.968 (95% CI, 0.924–1) (Fig. 1).

Discussion

PJI is a frequent cause of revision hip surgery and in some series trails only loosening and instability and dislocation [15, 16]. The currently used laboratory tests for infection are notable in failing to provide a sufficiently accurate diagnosis of this devastating complication. Sensitivity and specificity of each method have been questioned by different authors and there are no consistently agreed-on strategies that provide a reliable, expeditious, or highly accurate diagnosis [3] (Table 5). This is particularly true for intraoperative evaluation. We therefore sought to compare the use of intraoperative SCRP with that of intraoperative frozen section in the setting of revision THA.

Table 5. Results comparing frozen section with definitive pathology

Positive frozen section/histology	Infection present	Infection absent	Total
Positive test	21	0	21
Negative test	1	54	55
Total	22	54	76

The main limitation of our study includes the relatively small number of patients with different diagnoses that may affect the statistical power of our conclusions. However, previous publications describing SCRP as a diagnostic tool include a similar number of patients with hip and knee revision surgeries and do not compare SCRP with frozen section. Thus, we did not feel that a power calculation would be particularly helpful, because the sample sizes required for statistical exactitude would be unrealistic. Parvizi et al. found an 85% sensitivity and a 95% specificity to detect a PJI in 55 revision surgeries [14]. We observed high positive and negative predictive values using intraoperative SCRP. However, these values should be taken with caution, because they depend on the prevalence of the disease in the sample being studied. In addition, because we receive many infected THAs from other institutions, there were 23 patients with septic failures among our 76 patients, which is not the prevalence of infection in the general population. This raises the possibility of a lack of external validity, in which our results may not be reproducible in another practice setting. Because frozen section analysis depends on the surgeon and the pathologist, this test may be related to more errors than a laboratory test. The first error could be made by the surgeon by not taking samples from the areas of greatest suspicion. Another error might be made by the pathologist when handling the samples. We tried to minimize this as a source of error because the patients were operated on by three staff surgeons with more than 10 years in practice (MAB, FC, FP), and the histologic analysis was performed by two senior pathologists in a hospital that has 30 years of experience in this diagnostic method. The SCRP test is imperfect, because it is nonspecific, may increase in response to several diseases with acute inflammatory

reactions, and may be unusable in "dry" joints. However, all the cases in these series presented synovial fluid to test SCRP. Potential advantages of the SCRP test are that it may be used to monitor patients in second-stage reimplantations, in which frozen section has shown to be of little help to detect a persistent infection [8] and cultures from aspirations are affected by the elution of antibiotics from the cement spacers. Another practical limitation is that the threshold CRP level of 9.5 may not be appropriate at all centers.

We have previously studied the combination of serum CRP and interleukin-6 to detect PJI and compared with frozen section. Serum CRP and interleukin-6 provided similar sensitivity, specificity, and positive predictive values as the frozen section [5]. However, limitations of the interleukin-6 diagnostic method include high cost and relative lack of availability: in our institution, for logistic reasons, all IL-6 samples were saved and performed intermittently in batches. Thus, tests for individual patients were not accessible in a timely fashion when we started our investigations. A recent publication showed that the combination of SCRP with synovial α-defensin demonstrated a sensitivity of 97% and a specificity of 100% for the diagnosis of PJI. Synovial fluid α-defensin tests alone demonstrated a sensitivity of 97% and a specificity of 96% for the diagnosis of PJI [7]. However, α-defensin is not yet available in our country and the cost of this diagnostic method may be much higher than SCRP and not accessible to most of the institutions. Although Tetreault et al. have recently found that measurement of CRP in synovial fluid rather than serum using readily available assay equipment does not offer a diagnostic advantage in detection of PJIs [19], serum CRP may be elevated in many conditions other than PJI such as chronic inflammatory diseases, Paget's disease, and immunodeficiency syndromes. Thus, SCRP may be more specific to detect PJI, because serum CRP has been associated with a sensitivity of 86% and a specificity of 92% [17] and we observed higher values using SCRP (90% and 94%, respectively). Tetrault et al. also found that 15% of the cases had to be discarded because the synovial fluid was too viscous or hemolyzed; this was not the case in our series.

The ideal intraoperative test for PJI diagnosis should be accurate, convenient to the patient, cause minimal morbidity, and be cost-effective. SCRP could provide objective, analytical, and consistent results for all surgeons with no need for test interpretation. In this study, the quantification of SCRP presented comparable sensitivity, specificity, and predictive values as frozen section with the advantages of being a simple and economic test, widely available at any time in different centers, where frozen section analysis may not be accessible.

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