

# The Leukocyte Esterase Test for Periprosthetic Joint Infection is Not Affected by Prior Antibiotic Administration

### ABSTRACT

*Background:* Previous studies have demonstrated that administration of antibiotics prior to performing diagnostic testing for periprosthetic joint infection (PJI) can interfere with the accuracy of the standard diagnostic test results. Therefore, the purpose of this study was to evaluate the effects of antibiotic administration prior to performing the synovial leukocyte esterase (LE) strip test for PJI.

*Methods:* We identified 121 patients undergoing revision hip and knee **arthroplasty** for a **Musculoskeletal Infection Society** (MSIS)-confirmed diagnosis of PJI. All patients also had an LE strip test performed. Patients in one group (A)(32%) were on antibiotics prior to the diagnostic work-up, whereas patients in another group (B)(68%) did not receive antibiotics within 2 weeks of the diagnostic work-up. The LE strip test results (++/+), ESR (mm/h), serum CRP (mg/L), synovial WBC counts (cells/ul), and PMN (%) were collected and compared between the two groups.

*Results:* The median serum ESR (85 mm/h compared to 67 mm/h;  $p=0.009$ ), CRP(16.5 compared to 12.9;  $p=0.032$ ), synovial WBC(45,675 9,650;  $p<0.0001$ ), and PMN%(93% compared to 88%;  $p=0.004$ ) were all significantly lower for patients receiving antibiotics. Furthermore, administration of antibiotics resulted in a significant decrease in the sensitivity of all tests, except LE. ESR [92.7% compared to 79.5%, relative risk (RR)=2.8,  $p=0.04$ ], CRP (81.8% compared to 64.2%;  $RR=1.9$ ,  $p=0.03$ ), WBC (93.4% compared to 69.3%,  $RR=5.0$ ,  $p=0.001$ ), PMN (91.5% compared to 74.4%,  $RR=3.0$ ,  $p=0.01$ ), LE (83% compared to 77%,  $RR=1.6$ ,  $p=0.17$ ). Moreover, the rate of negative cultures were higher among group A patients at 30.7% compared to group B at 12.1% ( $p=0.021$ ).

*Conclusion:* This, and previous studies, have demonstrated that administration of premature antibiotics can compromise the results of standard diagnostic tests for PJI, causing lower median results and significant increases in false-negative results. However, the LE strip test

28 for PJI maintains its performance even in the setting of antibiotic administration. Antibiotic  
29 administration prior to diagnostic workups for PJI does not improve the care of these patients  
30 and stands to interfere with timely diagnosis. LE strip test can be used as a reliable diagnostic  
31 marker for diagnosing PJI even when prior antibiotics are administered.  
32 *Level of Evidence:* Diagnostic. Level II

## INTRODUCTION

Periprosthetic joint infection (PJI) is arguably one of the most complex complications of total joint arthroplasty (TJA). The burden of this devastating complication on both patients<sup>1-4</sup> and healthcare providers<sup>5,6</sup> has been well documented. Furthermore, this appears to be the tip of the iceberg given the projected increase in the number of primary and revision TJAs being performed in the future<sup>7</sup>. It is therefore imperative that suspected cases of PJI are diagnosed promptly and accurately in an attempt to optimize outcome for patients. Despite major progress in the diagnosis and management of PJI over the past decade, diagnostic uncertainty continues to trouble clinicians at the point of care delivery. A key development has been the introduction of the criteria for defining PJI by the Musculoskeletal Infection Society (MSIS)<sup>8</sup>. These guidelines were later modified at the International Consensus Meeting (ICM) on PJI in 2013 and are now the most widely used by the orthopaedic community (Table 1)<sup>9</sup>.

We have previously shown that premature antibiotic administration has a detrimental effect on the sensitivity of serum erythrocyte sedimentation rate (ESR) and c-reactive protein (CRP) as well as synovial white blood cell (WBC) count and polymorphonuclear neutrophil (PMN) percentage<sup>14</sup>. Moreover, it is well recognized that prior antibiotics treatment reduces culture yield and can result in culture-negative infections<sup>10-13</sup>. The diagnostic inaccuracies that can ensue in such situations will no doubt result in a delay in treatment of PJI and may compromise patient care. As such, the American Academy of Orthopaedic Surgeons (AAOS) clinical practice guidelines recommend that antibiotics are withheld for at least two weeks in order to prevent the aforementioned scenarios<sup>15</sup>.

Despite the importance of this issue, we are aware of no previous study which has yet investigated the susceptibility of the synovial leucocyte esterase (LE) strip test to prior antibiotic administration. This inexpensive and simple test forms one of the minor ICM

criteria for defining PJI and previous studies have demonstrated its role as an accurate point-of care test<sup>16-22</sup>. A recent Systematic Review and Meta-analysis estimated that the pooled diagnostic sensitivity and specificity of LE were 0.81 (95% confidence interval [CI], 0.49 to 0.95) and 0.97 (95% CI, 0.82 to 0.99)<sup>23</sup>. Additionally, the area under curve (AUC) for LE and PJI was 0.97 (95% CI, 0.95 to 0.98) in this study. LE is an enzyme, which is secreted by activated neutrophils and has been used for many years to detect infection in urine<sup>24,25</sup>, as well as other bodily fluids<sup>26-31</sup>.

The purpose of this study was therefore to evaluate the utility of synovial LE strip test for the diagnosis of PJI in the face of prior antibiotic administration and compare this with the other serological and serum markers recommended by the ICM diagnostic criteria (serum ESR, serum CRP, synovial WBCs, and synovial PMNs).

## **MATERIALS AND METHODS**

Institutional review board approval was obtained prior to the commencement of this retrospective study. All patients who had undergone revision hip or knee arthroplasty for PJI from October 2009 to July 2014 were identified from our institution's prospectively collected arthroplasty database. A total of 3,742 revision arthroplasties (1,618 hip and 2,124 knee) were performed during this period – of which 14.4% were performed for suspected PJI. There were 213 MSIS-confirmed PJI cases (99 hip; 114 knee) among this cohort and since LE testing is standard practice in all revision cases at our institution, all of these cases also had LE strip test results available. The clinical records of these patients were then searched to ascertain whether or not they had received oral and/or intravenous antibiotics in the two weeks preceding their clinical workup for PJI, that is, serum ESR/CRP and synovial fluid WBC count, and synovial fluid PMN%. The inclusion criteria for the study were; 1) the presence of a total hip or knee arthroplasty; 2) sufficient data to categorize PJI based on the

modified MSIS criteria; 3) available LE test strip result; and 4) available pre-investigation antibiotic information. Patients with insufficient laboratory and antibiotic status information and the ones who developed an early post-operative infection were excluded from the study..

Prior antibiotic administration information was available for a total of 121 of the MSIS-confirmed PJI cases. There were 39 (32%) patients who had received antibiotics within two weeks of baseline diagnostic workup for PJI and these patients made up the “Abx” cohort. These patients had received antibiotics by different referring organizations (emergency departments, family physicians and other orthopaedic departments) prior to being referred to our tertiary service. Hence, we were unable to determine the reasoning for commencement of antibiotics prior to diagnostic work-up for PJI. **However, based on electronic chart reviews it was noted that the referring institution initiated antibiotics for suspected hip or knee PJI in all included patients. However, we** were unable to obtain information regarding the type of antibiotic administered, the dosage, or duration. There were 82 (68%) patients who had not received any antibiotics prior to their diagnostic work-up for PJI and these patients made up the “No-Abx” cohort. In addition to baseline demographic data (age, sex, and body mass index [BMI]), the medical records of all the patients were reviewed for details of clinical and microbiological data (Table 2). There was no statistical difference between Abx and No-Abx cohorts in terms of age, gender, and joint type ( $p > 0.05$  for all variables). There was also no statistical difference between Abx and No-Abx cohorts in terms of frequency of gram-positive organism organisms ( $p=0.156$ ). There were 28 (72%) cases, which grew gram-positive bacteria in the Abx cohort compared to 64 (78%) cases in the No-Abx cohort.

Synovial fluid LE testing for all cases was performed using the Chemstrip 7 urine test strip (Roche Diagnostics, Indianapolis, Indiana) by trained orthopedic research fellows - either in the office or intra-operatively. This was carried out using a standardized protocol

previously described<sup>20</sup>. Importantly, this included centrifuging all synovial fluid samples prior to testing and reading the calorimetric test strip results at exactly one minute. The LE results can be classed as either negative, trace, +, or ++ with the latter two being considered a positive result in the current study. In addition to the LE test strip results, serum ESR, serum CRP, synovial fluid WBC count, and synovial fluid PMN% were recorded for each patient. The median for each of these laboratory tests was calculated for patients in the Abx and No-Abx cohort. The modified MSIS criteria thresholds (Table 1)<sup>9</sup> were used to calculate the sensitivity and false-negative rates for each of the diagnostic tests.

### *Statistical Analysis*

Descriptive statistics were used for reporting demographic data and all laboratory values. **Because the data were not normally distributed in the study groups a nonparametric test (Mann-Whitney test)** was used to compare median laboratory values between the Abx and No-Abx cohort. The Fisher's exact test was used to calculate the sensitivity of the LE and the diagnostic laboratory tests (ESR, CRP, WBC count, and PMN%) as well as the rate of false-negative results. Statistical analysis was performed using GraphPad Prism 7 (GraphPad Software, La Jolla, CA, USA). A p-value of  $\leq 0.05$  was considered statistically significant.

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## **RESULTS**

The administration of antibiotics resulted in a significant decrease in the sensitivity of all tests, except LE. The sensitivity of LE was 77% in Abx cohort compared to 83% in No-Abx cohort (relative risk [RR] =1.6, p=0.17). In contrast, serum ESR had a sensitivity of

79.5% in Abx cohort compared to 92.7% in No-Abx cohort (RR=2.8, p=0.04). Serum CRP had a sensitivity of 64.2% in Abx cohort compared to 81.8% in No-Abx cohort (RR=1.9, p=0.03). SF WBC count was 69.3% in Abx cohort compared to 93.4% in No-Abx cohort (RR=5.0, p=0.001). SF PMN% was 74.4% in Abx cohort compared to 91.5% in No-Abx cohort (RR=3.0, p=0.01)(Table 3, Figure1). Moreover, the use of premature antibiotics decreased the synovial fluid culture yield significantly from 87.9% in the No-Abx group to 69.3% in the Abx group (RR=2.5, p=0.015)

In addition, the median values for the diagnostic laboratory tests were significantly lower in patients who had received antibiotics (Table 4). The median serum ESR was 67mm/hr (range: 5-140) for Abx group compared with 85mm/hr (range: 11-142) for No-Abx group (p=0.009). The median serum CRP was 12.9mg/L (range: 1-45.4) for Abx cohort compared to 16.5mg/L (range: 2-70) for No-Abx cohort (p=0.032). Similar differences were present for synovial fluid tests. The median synovial fluid (SF) WBC count was 9,650 cells/ $\mu$ L (range: 30-529,200) in Abx cohort compared with 45,675 cells/ $\mu$ L (range: 27-325,500) in No-Abx cohort (p<0.0001). The median SF PMN% was 88% (range: 3-100) in Abx cohort compared with 93% (range 32-99) in No-Abx cohort (p=0.004).

## DISCUSSION

This study evaluated the utility of the synovial LE strip test for the diagnosis of PJI in cases where antibiotics had been administered prior to diagnostic work-up. It compared the LE test with the other serological and serum markers used in the ICM criteria (serum ESR, serum CRP, synovial WBCs, and synovial PMNs). It is the first of its kind to investigate the effect of premature antibiotic administration on the accuracy of the LE strip test for diagnosing PJI. Our findings suggest that the LE strip test maintains its performance even in the setting of previous antibiotic administration. Unlike serum ESR, CRP, synovial fluid



WBC count, and PMN%; LE remains a reliable test for diagnosing PJI in these unfortunate but not uncommon circumstances. **Furthermore, none of the latter diagnostic tests are available as a “point of care” test. From a practical point of view, the strength of LE testing lies in the fact that it can be used in the office or bedside with immediate results, thereby enabling rapid management decisions to be made.** In addition, our findings were in line with the previous studies and provided further evidence that prior antibiotic administration can reduce the reliability of the standard diagnostic laboratory tests used in the MSIS criteria<sup>14</sup> as well as the culture yield<sup>32</sup>.

Given the significant morbidity and mortality associated with PJI<sup>1,2</sup>, an accurate and timely diagnosis remains one the most crucial aspects of the management. The LE strip test is a reliable diagnostic tool, even when antibiotics have been administered prior to diagnostic work-up. Although the sensitivity of the LE test is not high enough for it to be used as a stand-alone screening test for PJI, the findings of the current study support its continued use in conjunction with other traditional tests due to its excellent specificity and negative predictive value<sup>23</sup>. A recent study has demonstrated that when LE results (+/++) are concordant with serum ESR and CRP, it can be used to confirm or rule out infection with a greater than 95% certainty<sup>33</sup>.

A previous study has also demonstrated that alpha-defensin, as a promising synovial biomarker, also maintains its concentration and sensitivity in the setting of prior antibiotic administration<sup>34</sup>. As with the LE test, alpha-defensin had higher sensitivity in detecting PJI when compared with traditional laboratory tests and culture. However, what makes the LE test potentially more attractive is that it is considerably less expensive (especially in Europe where the alpha-defensin test is available as a qualitative point-of-care test. Future studies are needed to focus on comparing the cost-effectiveness of these synovial biomarkers.

There are some limitations that must be considered when interpreting the findings of the current study. This was a retrospective study, hence was carried with inherited weaknesses such as variability in data collection. Secondly, due to the lack of information about the antibiotic types, dosages, and routes of administration, we were unable to establish an association between these variables and the laboratory results. **We did not have the antibiotic susceptibility of the infecting organisms. It was therefore theoretically possible that some patients received antibiotics to which the infecting organisms were resistant, or even antibiotics that did not cover the infecting organism. However, based on the chart reviews, all patients were commenced on antibiotics to cover presumed hip and knee PJI. Nevertheless, there is no doubt that the availability of more information on antibiotics susceptibility of the infecting organisms may have helped to further refine the reliability of the tests that were evaluated in the current study.** Finally, by selecting only the MSIS-confirmed PJI cases, we may have inadvertently missed some patients who had PJI and had received prior antibiotics, but were not included in the study, as they did not reach enough positive MSIS criteria. This potential scenario may have resulted in underestimation of the affect of antibiotics on the diagnostic tests.

In conclusion, this study once again endorses LE strip test as a reliable, widely available, and inexpensive test for diagnosing PJI even when premature therapeutic antibiotics are used. Thus, based on the findings of this study and building on the prior recommendations by the AAOS we strongly urge the medical community to refrain from administration of antibiotics prior to infection work up in patients who are suspected to have PJI i.e. any TJA patient with a red painful knee or similar symptoms. This ill-advised practice does not improve the care of these patients and significantly compromises the accuracy of the standard diagnostic tests resulting in interference with timely diagnosis. However, in case clinicians faced such patients, LE strip test can be used as a reliable tool to confirm or refute

207 diagnosis in conjunction with the patients' clinical picture and the rest of the battery of the  
208 diagnostic tests.

## References

1. Zmistowski B, Karam JA, Durinka JB, Casper DS, Parvizi J. Periprosthetic joint infection increases the risk of one-year mortality. *J Bone Joint Surg Am* 2013; **95**(24): 2177-84.
2. Berend KR, Lombardi AV, Jr., Morris MJ, Bergeson AG, Adams JB, Sneller MA. Two-stage treatment of hip periprosthetic joint infection is associated with a high rate of infection control but high mortality. *Clin Orthop Relat Res* 2013; **471**(2): 510-8.
3. Choi HR, Bedair H. Mortality following revision total knee arthroplasty: a matched cohort study of septic versus aseptic revisions. *J Arthroplasty* 2014; **29**(6): 1216-8.
4. Choi HR, Beecher B, Bedair H. Mortality after septic versus aseptic revision total hip arthroplasty: a matched-cohort study. *J Arthroplasty* 2013; **28**(8 Suppl): 56-8.
5. Kurtz SM, Lau E, Watson H, Schmier JK, Parvizi J. Economic burden of periprosthetic joint infection in the United States. *J Arthroplasty* 2012; **27**(8 Suppl): 61-5 e1.
6. Parisi TJ, Konopka JF, Bedair HS. What is the Long-term Economic Societal Effect of Periprosthetic Infections After THA? A Markov Analysis. *Clin Orthop Relat Res* 2017.
7. Kurtz S, Ong K, Lau E, Mowat F, Halpern M. Projections of primary and revision hip and knee arthroplasty in the United States from 2005 to 2030. *J Bone Joint Surg Am* 2007; **89**(4): 780-5.
8. Parvizi J, Zmistowski B, Berbari EF, et al. New definition for periprosthetic joint infection: from the Workgroup of the Musculoskeletal Infection Society. *Clin Orthop Relat Res* 2011; **469**(11): 2992-4.
9. Zmistowski B, Della Valle C, Bauer TW, et al. Diagnosis of periprosthetic joint infection. *J Arthroplasty* 2014; **29**(2 Suppl): 77-83.
10. Berbari EF, Marculescu C, Sia I, et al. Culture-negative prosthetic joint infection. *Clin Infect Dis* 2007; **45**(9): 1113-9.

- 234 11. Malekzadeh D, Osmon DR, Lahr BD, Hanssen AD, Berbari EF. Prior use of  
235 antimicrobial therapy is a risk factor for culture-negative prosthetic joint infection. *Clin*  
236 *Orthop Relat Res* 2010; **468**(8): 2039-45.
- 237 12. Trampuz A, Piper KE, Hanssen AD, et al. Sonication of explanted prosthetic  
238 components in bags for diagnosis of prosthetic joint infection is associated with risk of  
239 contamination. *J Clin Microbiol* 2006; **44**(2): 628-31.
- 240 13. Trampuz A, Piper KE, Jacobson MJ, et al. Sonication of removed hip and knee  
241 prostheses for diagnosis of infection. *N Engl J Med* 2007; **357**(7): 654-63.
- 242 14. \*\*\* **Blinded by JBJS** \*\*\*
- 243 15. Della Valle C, Parvizi J, Bauer TW, et al. American Academy of Orthopaedic  
244 Surgeons clinical practice guideline on: the diagnosis of periprosthetic joint infections of the  
245 hip and knee. *J Bone Joint Surg Am* 2011; **93**(14): 1355-7.
- 246 16. Wang C, Li R, Wang Q, Duan J, Wang C. Leukocyte Esterase as a Biomarker in the  
247 Diagnosis of Periprosthetic Joint Infection. *Med Sci Monit* 2017; **23**: 353-8.
- 248 17. Shafafy R, McClatchie W, Chettiar K, et al. Use of leucocyte esterase reagent strips in  
249 the diagnosis or exclusion of prosthetic joint infection. *Bone Joint J* 2015; **97-B**(9): 1232-6.
- 250 18. Deirmengian C, Kardos K, Kilmartin P, et al. The alpha-defensin test for  
251 periprosthetic joint infection outperforms the leukocyte esterase test strip. *Clin Orthop Relat*  
252 *Res* 2015; **473**(1): 198-203.
- 253 19. Colvin OC, Kransdorf MJ, Roberts CC, et al. Leukocyte esterase analysis in the  
254 diagnosis of joint infection: can we make a diagnosis using a simple urine dipstick? *Skeletal*  
255 *Radiol* 2015; **44**(5): 673-7.
- 256 20. Tischler EH, Cavanaugh PK, Parvizi J. Leukocyte esterase strip test: matched for  
257 musculoskeletal infection society criteria. *J Bone Joint Surg Am* 2014; **96**(22): 1917-20.

- 258 21. Wetters NG, Berend KR, Lombardi AV, Morris MJ, Tucker TL, Della Valle CJ.  
259 Leukocyte esterase reagent strips for the rapid diagnosis of periprosthetic joint infection. *J*  
260 *Arthroplasty* 2012; **27**(8 Suppl): 8-11.
- 261 22. Parvizi J, Jacovides C, Antoci V, Ghanem E. Diagnosis of periprosthetic joint  
262 infection: the utility of a simple yet unappreciated enzyme. *J Bone Joint Surg Am* 2011;  
263 **93**(24): 2242-8.
- 264 23. Wyatt MC, Beswick AD, Kunutsor SK, Wilson MJ, Whitehouse MR, Blom AW. The  
265 Alpha-Defensin Immunoassay and Leukocyte Esterase Colorimetric Strip Test for the  
266 Diagnosis of Periprosthetic Infection: A Systematic Review and Meta-Analysis. *J Bone Joint*  
267 *Surg Am* 2016; **98**(12): 992-1000.
- 268 24. Kusumi RK, Grover PJ, Kunin CM. Rapid detection of pyuria by leukocyte esterase  
269 activity. *JAMA* 1981; **245**(16): 1653-5.
- 270 25. Perry JL, Matthews JS, Weesner DE. Evaluation of leukocyte esterase activity as a  
271 rapid screening technique for bacteriuria. *J Clin Microbiol* 1982; **15**(5): 852-4.
- 272 26. Azoulay E, Fartoukh M, Galliot R, et al. Rapid diagnosis of infectious pleural  
273 effusions by use of reagent strips. *Clin Infect Dis* 2000; **31**(4): 914-9.
- 274 27. Gal-Oz A, Kassis I, Shprecher H, Beck R, Bentur L. Correlation between rapid strip  
275 test and the quality of sputum. *Chest* 2004; **126**(5): 1667-71.
- 276 28. Lebovics RS, Murthy VV, Karmen A. Leukocyte esterase activity in effusion fluid of  
277 patients with otitis media. *Otolaryngol Head Neck Surg* 1993; **108**(3): 248-50.
- 278 29. Jacobs JA, De Brauwier EI, Cornelissen EI, Drent M. Correlation of leukocyte  
279 esterase detection by reagent strips and the presence of neutrophils: a study in BAL fluid.  
280 *Chest* 2000; **118**(5): 1450-4.
- 281 30. Sam R, Sahani M, Ulozas E, Leehey DJ, Ing TS, Gandhi VC. Utility of a peritoneal  
282 dialysis leukocyte test strip in the diagnosis of peritonitis. *Artif Organs* 2002; **26**(6): 546-8.

- 283 31. Matsuda M, Noda Y, Takemori Y. Novel diagnostic method of testing for  
284 *Helicobacter pylori* infection using the rapid leukocyte strip test, Leukostix. *J Gastroenterol*  
285 *Hepatol* 2003; **18**(10): 1196-201.
- 286 32. Parvizi J, Erkocak OF, Della Valle CJ. Culture-negative periprosthetic joint infection.  
287 *J Bone Joint Surg Am* 2014; **96**(5): 430-6.
- 288 33. Tarabichi M, Fleischman AN, Shahi A, Tian S, Parvizi J. Interpretation of Leukocyte  
289 Esterase for the Detection of PJI based on Serological Markers. *The Journal of Arthroplasty*.
- 290 34. Shahi A, Parvizi J, Kazarian GS, et al. The Alpha-defensin Test for Periprosthetic  
291 Joint Infections Is Not Affected by Prior Antibiotic Administration. *Clin Orthop Relat Res*  
292 2016; **474**(7): 1610-5.

293 **Figure Legend**

294 **Figure 1.** Comparison of false-negative rates of laboratory tests of patients in the Abx and  
295 No-Abx groups for the presence of PJI.