HK34: What thresholds for neutrophils in histopathological sections should we use in the diagnosis of periprosthetic joint infections?

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Response/Recommendation: The optimal threshold of polymorphonuclear neutrophils (PMN) in histopathological sections for diagnosing periprosthetic joint infections (PJI) is still a matter of debate. A threshold of ≥ five PMN/HPF in each of at least five high-power fields (HPFs) is recommended to differentiate infected from aseptic hip and knee cases in frozen and permanent histopathological sections of deep tissue samples.

Level of Evidence: Moderate
Delegate Vote:

Rationale:

Thresholds of either five PMN/HPF or 10 PMN/HPF in each of five HPFs were recommended in the ICM guideline of 2018 for diagnosing periprosthetic joint infection (PJI) [1], and a threshold of ≥5 PMN/HPF in each of five HPFs in the EBJIS definition of PJI [2]. The three most commonly used thresholds in the literature are ≥23 PMN/10HPF, ≥ five PMN/HPF, and ≥10 PMN/HPF. However, high-quality studies on the optimal threshold for polymorphonuclear neutrophils (PMN) in histopathological sections for diagnosing hip and knee PJI are scarce.

To address the posed question, we conducted a comprehensive systematic review. Relevant studies were identified using the MeSH terms developed by librarians. After screening the initial studies, articles that met the inclusion criteria were reviewed in detail, and data was extracted.

Permanent sections

In our meta-analysis of 16 studies (2,308 patients undergoing revision surgery following total hip or knee arthroplasty) reporting the used threshold for permanent sections of intraoperatively collected samples [3-18], five studies [3-7] presented the diagnostic value of histopathological analysis when using the threshold of ≥23 PMN/10HPF, eleven studies [7-17] when applying the threshold of ≥ five PMN/HPF, and four studies [7, 8, 17, 18] when using the threshold of ≥10 PMN/HPF. The threshold of ≥23 PMN/10HPF showed a pooled sensitivity, specificity, diagnostic odds ratio (DOR), and summarized area under the curve (sAUC) of 80.7% (95% CI: 78.0-83.3), 94.9% (95% CI: 93.3-96.3), 162.4 (95% CI: 36.4-725.7), and 0.957 (SE 0.02). When considering the threshold of ≥ five PMN/HPF, they were 82.0% (95% CI: 80.0-84.0), 94.7% (95% CI:93.5-95.8), 133.5 (95% CI: 41.6-428.6), and 0.963 (SE: 0.02); and 88.8% (95% CI: 84.6-92.2), 89.2% (95%CI: 85.0-92.5), 67.1 (95% CI: 39.0-115.3), and 0.950 (SE: 0.01) when the threshold of ≥10 PMN/HPF was used. In this meta-analysis, a threshold of ≥ five PMN/HPF showed the best performance. However, several limitations need to be highlighted: The majority of studies (13 of 16 did not use a standardized infection definition. The gold standard reference in five studies [8, 9, 14, 16, 18] was a positive culture, and in eight studies [3-6, 10, 11, 15, 17], institutional criteria (including clinical features, microbiology, and/or histology). In only three studies [7, 12, 13], a standardized infection definition was applied. Many studies included histology in their infection definition, leading to an incorporation bias. Furthermore, no uniform histopathological investigation (e.g., different numbers of high-power fields) was performed, making a comparison between studies difficult. These limitations may also explain the untypically higher pooled sensitivity and lower pooled specificity in studies

utilizing the higher threshold of ≥10 PMN/HPF compared to studies using the threshold of ≥ five PMN/HPF indicating low reliability of the available studies.
Only one study investigated different thresholds with standardized infection definitions and histopathological analyses [7]. In this retrospective study of 119 patients undergoing revision hip or knee arthroplasty, a threshold of ≥ five PMN/HPF was recommended. Nevertheless, the effective threshold depended importantly on the chosen infection definition, with more PJI cases identified when using the EBJIS definition. When the ICM or IDSA definitions were used as a reference for PJI, ≥10 PMN/HPF showed a better diagnostic value compared to ≥ five PMN/HPF. However, when applying the EBJIS definition, no difference was observed between the latter two thresholds (AUC 0.89 versus 0.90; *P*=0.916), but higher sensitivities were seen with a threshold of ≥ five PMN/HPF (93 versus 86%) at the cost of lower specificity (84 versus 93%). Due to the better sensitivity when applying the more sensitive EBJIS infection definition, the threshold of ≥ five PMN/HPF was recommended to ensure accurate infection diagnosis. In addition, the threshold of ≥23 PMN/10HPF showed a significantly poorer outcome in comparison to ≥ five PMN/HPF and ≥10 PMN/HPF regardless of the used infection definition (EBJIS, IDSA, ICM 2018) [7].

Frozen sections

In our systematic review and meta-analysis of 22 studies (2,904 patients who have septic or aseptic failure after a total hip or knee arthroplasty) describing the used threshold for frozen sections [13, 14, 17-36], three studies [19-21] reported the diagnostic value of histopathological analysis when using the threshold of ≥ 23 PMN/10HPF, 16 [13, 14, 17, 22-34] when applying the threshold of ≥ 5 PMN/HPF, and six studies [14, 17, 18, 33, 35, 36] when using the threshold of ≥10 PMN/HPF. The threshold of ≥ 23 PMN/10HPF showed a pooled sensitivity, specificity, diagnostic odds ratio (DOR), and summarized area under the curve (sAUC) of 81.0% (95% CI: 74.9-86.1), 95.6% (95% CI: 91.8-98.0), 83.3 (95% CI: 19.7-352.9), and 0.940 (SE 0.12). When using the threshold of ≥ five PMN/HPF, performance values were 69.4% (95% CI: 67.5-71.3), 94.4% (95% CI: 93.3-95.3), 56.2 (95% CI: 30.4-103.7), and 0.964 (SE: 0.01); and when applying the threshold of ≥ 10 PMN/HPF 62.0% (95% CI: 58.3-65.6), 96.1% (95% CI: 94.4-97.4), 50.7 (95% CI: 20.4-126.1), and 0.892 (SE: 0.05), respectively. However, similar limitations were observed in these studies: No standardized infection definition was used in 15 studies (only positive culture: n = 10, institutional criteria: n = 5) [14, 17, 18, 21-24, 26, 29-33, 35, 36]. Many studies included histology in their infection definition and no uniform histopathological analysis (e.g., different numbers of high-power fields) was performed among studies. It needs to be highlighted that only three studies with overall 205 patients including 60 PJIs investigated the threshold of ≥ 23 PMN/10HPF, while 16 studies with 2,256 patients (378 PJIs) evaluated the threshold of ≥ five PMN/HPF and seven studies with 697 patients (133 PJIs) the threshold of ≥ 10 PMN/HPF. All these drawbacks make a comparison between studies difficult.
High accuracies are reported in the literature, and despite the listed limitations, a threshold of ≥ five PMN/HPF in each of at least five high power fields (HPFs) can be recommended to differentiate septic from aseptic failure in frozen and permanent histopathological sections of deep tissue samples. At least five high power fields should be investigated and the mean PMN count calculated [7]. However, it needs to be emphasized that a threshold of ≥ 10 PMN/HPF can be used as well, but with a risk of missing some (low-grade) PJIs. In addition, the presence of only one to five PMN/HPF cannot exclude infection. In these cases, a careful interpretation in conjunction with other diagnostic test methods within the infection definition is recommended.

References:

1. Shohat, N., et al., *Hip and Knee Section, What is the Definition of a Periprosthetic Joint Infection (PJI) of the Knee and the Hip? Can the Same Criteria be Used for Both*

Joints?: Proceedings of International Consensus on Orthopedic Infections. J Arthroplasty, 2019. **34**(2s): p. S325-s327.
2. McNally, M., et al., *The EBJIS definition of periprosthetic joint infection.* Bone Joint J, 2021. **103-b**(1): p. 18-25.
3. Fernández-Sampedro, M., et al., *Accuracy of different diagnostic tests for early, delayed and late prosthetic joint infection.* BMC Infect Dis, 2017. **17**(1): p. 592.
4. Janz, V., et al., *Evaluation of sonicate fluid cultures in comparison to histological analysis of the periprosthetic membrane for the detection of periprosthetic joint infection.* Int Orthop, 2013. **37**(5): p. 931-6.
5. Morawietz, L., et al., *Twenty-three neutrophil granulocytes in 10 high-power fields is the best histopathological threshold to differentiate between aseptic and septic endoprosthesis loosening.* Histopathology, 2009. **54**(7): p. 847-53.
6. Müller, M., et al., *Diagnosis of periprosthetic infection following total hip arthroplasty--evaluation of the diagnostic values of pre- and intraoperative parameters and the associated strategy to preoperatively select patients with a high probability of joint infection.* J Orthop Surg Res, 2008. **3**: p. 31.
7. Sigmund, I.K., et al., *Diagnostic accuracy of neutrophil counts in histopathological tissue analysis in periprosthetic joint infection using the ICM, IDSA, and EBJIS criteria.* Bone Joint Res, 2021. **10**(8): p. 536-547.
8. Boettner, F., et al., *The Rule of Histology in the Diagnosis of Periprosthetic Infection: Specific Granulocyte Counting Methods and New Immunohistologic Staining Techniques may Increase the Diagnostic Value.* Open Orthop J, 2016. **10**: p. 457-465.
9. Bori, G., et al., *Interface membrane is the best sample for histological study to diagnose prosthetic joint infection.* Mod Pathol, 2011. **24**(4): p. 579-84.
10. Inagaki, Y., et al., *Correlation of histological and microbiological findings in septic and aseptic knee implant failure.* Arch Orthop Trauma Surg, 2019. **139**(5): p. 717-722.
11. Kashima, T.G., et al., *Use of chloroacetate esterase staining for the histological diagnosis of prosthetic joint infection.* Virchows Arch, 2015. **466**(5): p. 595-601.
12. Kuo, F.C., et al., *Which Minor Criteria is the Most Accurate Predictor for the Diagnosis of Hip and Knee Periprosthetic Joint Infection in the Asian Population?* J Arthroplasty, 2022. **37**(10): p. 2076-2081.
13. Li, R., et al., *Comparison of leukocyte esterase testing of synovial fluid with synovial histology for the diagnosis of periprosthetic joint infection.* Medical Science Monitor, 2017. **23**: p. 4440-4446.
14. Lonner, J.H., et al., *The reliability of analysis of intraoperative frozen sections for identifying active infection during revision hip or knee arthroplasty.* J Bone Joint Surg Am, 1996. **78**(10): p. 1553-8.
15. Montoya-delaTorre, C., et al., *Usefulness of histology for predicting infection at the time of hip and knee revision in patients with rheumatoid arthritis.* Arch Orthop Trauma Surg, 2022. **142**(10): p. 2489-2495.
16. Pandey, R., E. Drakoulakis, and N.A. Athanasou, *An assessment of the histological criteria used to diagnose infection in hip revision arthroplasty tissues.* J Clin Pathol, 1999. **52**(2): p. 118-23.
17. Wong, Y.C., et al., *Intraoperative frozen section for detecting active infection in failed hip and knee arthroplasties.* J Arthroplasty, 2005. **20**(8): p. 1015-20.
18. Miyamae, Y., et al., *Different diagnostic properties of C-reactive protein, real-time PCR, and histopathology of frozen and permanent sections in diagnosis of periprosthetic joint infection.* Acta Orthop, 2013. **84**(6): p. 524-9.
19. Sigmund, I.K., et al., *A comparative study of intraoperative frozen section and alpha defensin lateral flow test in the diagnosis of periprosthetic joint infection.* Acta Orthop, 2019: p. 1-11.

20. Kasparek, M.F., et al., *Intraoperative Diagnosis of Periprosthetic Joint Infection Using a Novel Alpha-Defensin Lateral Flow Assay.* J Arthroplasty, 2016.
21. Tohtz, S.W., et al., *Validity of frozen sections for analysis of periprosthetic loosening membranes.* Clin Orthop Relat Res, 2010. **468**(3): p. 762-8.
22. Bori, G., et al., *Low sensitivity of histology to predict the presence of microorganisms in suspected aseptic loosening of a joint prosthesis.* Mod Pathol, 2006. **19**(6): p. 874-7.
23. Feldman, D.S., et al., *The role of intraoperative frozen sections in revision total joint arthroplasty.* J Bone Joint Surg Am, 1995. **77**(12): p. 1807-13.
24. Kanner, W.A., K.J. Saleh, and H.F. Frierson, Jr., *Reassessment of the usefulness of frozen section analysis for hip and knee joint revisions.* Am J Clin Pathol, 2008. **130**(3): p. 363-8.
25. Kelly, M.E., et al., *The Utility of Frozen Section Histology in Diagnosing Periprosthetic Joint Infection in Revision Total Joint Arthroplasty.* Journal of Arthroplasty, 2021. **36**(6): p. 2137-2143.
26. Ko, P.S., et al., *The role of intraoperative frozen section in decision making in revision hip and knee arthroplasties in a local community hospital.* J Arthroplasty, 2005. **20**(2): p. 189-95.
27. Kwiecien, G., et al., *Intraoperative Frozen Section Histology: Matched for Musculoskeletal Infection Society Criteria.* J Arthroplasty, 2017. **32**(1): p. 223-227.
28. Luyckx, L., et al., *Intraoperative frozen section histopathology for the diagnosis of periprosthetic joint infection in hip revision surgery: the influence of recent dislocation and/or periprosthetic fracture.* HIP International, 2022. **32**(1): p. 87-93.
29. Nuñez, L.V., et al., *Frozen sections of samples taken intraoperatively for diagnosis of infection in revision hip surgery.* Acta Orthop, 2007. **78**(2): p. 226-30.
30. Pace, T.B., K.J. Jeray, and J.T. Latham, Jr., *Synovial tissue examination by frozen section as an indicator of infection in hip and knee arthroplasty in community hospitals.* J Arthroplasty, 1997. **12**(1): p. 64-9.
31. Pons, M., et al., *Infected total hip arthroplasty--the value of intraoperative histology.* Int Orthop, 1999. **23**(1): p. 34-6.
32. Spangehl, M.J., et al., *The role of intraoperative gram stain in the diagnosis of infection during revision total hip arthroplasty.* J Arthroplasty, 1999. **14**(8): p. 952-6.
33. Wu, C., et al., *Utility of intraoperative frozen section in the diagnosis of periprosthetic joint infection.* PLoS One, 2014. **9**(7): p. e102346.
34. Zagra, L., et al., *Can leucocyte esterase replace frozen sections in the intraoperative diagnosis of prosthetic hip infection?* Bone Joint J, 2019. **101-b**(4): p. 372-377.
35. Banit, D.M., H. Kaufer, and J.M. Hartford, *Intraoperative frozen section analysis in revision total joint arthroplasty.* Clin Orthop Relat Res, 2002(401): p. 230-8.
36. Francés Borrego, A., et al., *Diagnosis of infection in hip and knee revision surgery: intraoperative frozen section analysis.* Int Orthop, 2007. **31**(1): p. 33-7.