

## Q&A column

### Editor: Frederick L. Kiechle, MD, PhD

Submit your pathology-related question for reply by appropriate medical consultants. CAP TODAY will make every effort to answer all relevant questions. However, those questions that are not of general interest may not receive a reply. For your question to be considered, you must include your name and address; this information will be omitted if your question is published in CAP TODAY.

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**Q. Our lab does not have reference ranges established for body fluid manual differentials. Is it acceptable to use ranges from a reference material and include a disclaimer citing the source of the ranges?**

A. July 2021—It is acceptable to use ranges from reference material for body fluid manual differentials, including cell counts from peritoneal, pleural, pericardial, and synovial fluids. This is reflected in the CAP hematology and coagulation checklist requirement HEM.36820 Reference Intervals. This checklist requirement was revised in September 2019 to include the following statement: “Reference interval citations from the manufacturer’s insert or published literature citations may be used to determine the reference interval.”<sup>1</sup> This may be most relevant for laboratories with relatively low test volumes for which establishing institutional reference intervals is not feasible.

Note that some published intervals from body fluid reference material may be based on total white blood cells rather than total nucleated cells. There are a number of clinical and practical reasons why the TNC count is preferred over the WBC count, including the desire to identify malignant non-WBCs and simplify chamber (hemocytometer) counts for low cellularity specimens. A checklist item recently underwent revision to reflect this; it will be published this fall. Please refer to a CAP TODAY Q&A by Megan Nakashima, MD, for a discussion of how automated body fluid cell counts should be reported.<sup>2</sup>

Reference intervals are not available for some tests, including for many body fluid counts, because they depend on complex dynamic factors related to a patient’s physiologic and disease states.<sup>3</sup> For example, reference intervals may not be available for bronchoalveolar lavage, dialysis, and drainage fluid specimens. In these instances, a comment accompanying the result is required. HEM.36820 provides the following suggested language: “The reference interval(s) and other method performance specifications are unavailable for this body fluid. Comparison of the result with concentration in the blood, serum, or plasma is recommended.”

1. College of American Pathologists. HEM.36820 Reference intervals. In: Hematology and coagulation checklist. June 4, 2020.
2. Nakashima MO. Q&A Column. CAP TODAY. 2020;34(1):47-48.
3. Kjeldsberg CR, Knight JA. *Body Fluids: Laboratory Examination of Amniotic, Cerebrospinal, Seminal, Serous and Synovial Fluids*. American Society of Clinical Pathologists; 1993.

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**Q. In our lab, we perform semen analysis and make slides to send out for sperm morphology using Kruger's strict criteria. We get quite a few results back as swollen sperm head for probable contamination. The reference lab insisted that liquefying agent was added, but when we reviewed the results, the sample was normal, so liquefying agent wasn't used. What can cause a sperm head to swell, other than liquefying agent?**

A. Swollen sperm heads may result from a fresh semen sample being exposed to a hypoosmotic solution, which permits fluid to enter through the membranes and create swelling. A hypoosmotic swelling test evaluates the functional integrity of the sperm's plasma membrane and the fertility potential of sperm.<sup>1</sup> If the sample was exposed to a hypoosmotic liquefying agent prior to preparing the morphology slides, it is possible that the heads would swell. If the sample was not exposed to any solution, the laboratory should ensure that an ocular micrometer is used during the morphology analysis and that the reference lab is not merely eyeballing the slide. A normal head, according to Kruger analysis, ranges from 3  $\mu\text{m}$  to 5  $\mu\text{m}$  in width and 5  $\mu\text{m}$  to 7  $\mu\text{m}$  in length.<sup>2</sup>

Your lab may want to consider splitting sample slides between the aforementioned reference lab and a different reference lab to see if the latter reports swollen heads.

1. Ramu S, Jeyendran R. The hypo-osmotic swelling test for evaluation of sperm membrane integrity. *Methods Mol Biol.* 2013;927:21-25.
2. Menkveld R, Stander FS, Kotze TJ, Kruger TF, van Zyl JA. The evaluation of morphological characteristics of human spermatozoa according to stricter criteria. *Hum Reprod.* 1990;5(5):586-592.

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