COLLEGE of AMERICAN PATHOLOGISTS

Educational Discussion: Free T4 Testing

2023-A Harmonized Thyroid (ABTH)

As noted in the discussion from our Participant Summary Report for the 2022 ABTH-B Survey, the TSH data, on presumably commutable specimens, indicate that there are some potentially clinically significant biases. Since there is no reference method available for TSH, it's not possible to determine which method(s) produce accurate results. One must rely on the reference intervals to determine which patients have "abnormal" high or low results.

Unfortunately, comparing two of the manufacturers on whom we had a sufficient number of participants, we saw that the direction of the bias was not matched by the difference in the manufacturers' proposed reference intervals (i.e., the assay whose results were higher had a lower reference interval). In other words, even without knowing which results are accurate (or even if either assay is accurate), these manufacturers' reference intervals would result in different categorizations of patients. Our findings matched those from a recent publication.[1]

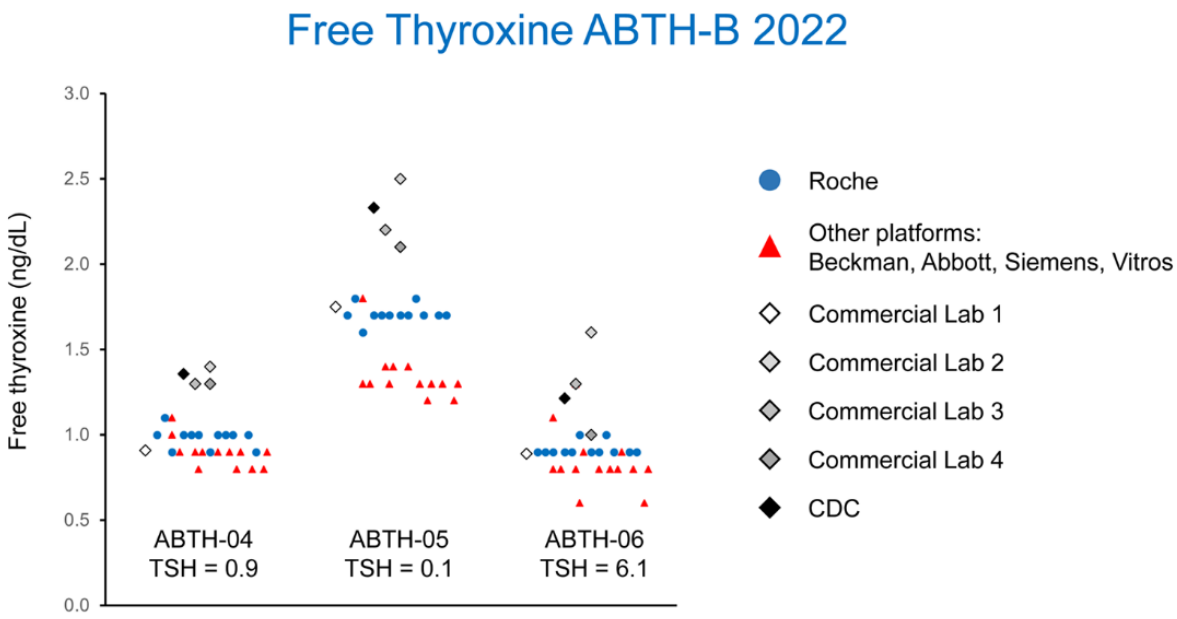
We had hoped that the situation with Free T4 would be more straightforward. In this case, there are two classes of assays available – immunoassays and mass-spectrometry. Mass spectrometry is considered a "gold standard" methodology, and a reference method is available. What complicates the results from mass spectrometry is that, even though T4 is a well-defined chemical compound, the pre-analytic steps can be variable.

In the accompanying figure, we have plotted the Free T4 results from the previous Survey (ABTH 2022-B). The values in blue represent the individual results from Roche (the only peer group with sufficient participants to form a peer group), and those in red represent the individual results from the other participants using immunoassays (Abbott, Beckman, Siemens, Vitros). There are five other points represented, one each from five reference/commercial laboratories, all of whom use equilibrium dialysis and liquid chromatography-tandem mass spectrometry.

For specimens ABTH-04 and ABTH-06, all of the immunoassay participants seem to get comparable results, versus specimen ABTH-05, where the Roche values are higher in most cases than the other immunoassays. Perhaps more interesting is that the mass spectrometry assays do not agree with one another as well as the immunoassays. Interestingly, one of the assays seems to track with the Roche values. On specimen ABTH-04, the other four mass spectrometry methods agree reasonably well. On specimen ABTH-05, three of the assays match pretty closely, with one other running considerably higher than the other four. All of the mass spectrometry results for sample ABTH-05 seem to suggest hyperthyroidism based on the individual laboratories' reference intervals, but it is disturbing that the values span such a wide range. Specimen ABTH-06 has a pattern similar to specimen ABTH-05. In this case, all values suggest euthyroidism by the laboratories' reference intervals, but again it is disturbing to see such a wide range of values from laboratories using the same detection method.

What can we take away from these studies? As was the case with TSH on the last Survey, it seems that the commercial/reference labs using mass spectrometry do not obtain consistent results. So, we're again left with the conclusion that it's critically important that all laboratories validate (and communicate) their reference intervals. Current "state of the art" for Free T4, even on commutable specimens, leaves much to be desired. It is nothing like cholesterol or hemoglobin A1c.

COLLEGE of AMERICAN PATHOLOGISTS



Reference:

1. Kalaria T, Sanders A, Fenn J, et al. The diagnosis and management of subclinical hypothyroidism is assay-dependent- Implications for clinical practice. *Clin Endocrinol*. 2021;94(6):1012-1016.

Gary Horowitz, MD, FCAP, Advisor
Andrew Hoofnagle, MD, PhD, FCAP, Chair
Accuracy-Based Programs Committee