respectively. None of the patients tested were being treated with digoxin.

Sera from 17 patients, newborn to 10 months old, were analyzed. Of these, 11 had values for digoxin at or above the assay sensitivity of 0.2 \( \mu \text{g/L} \), two of them (1.2 and 1.4 \( \mu \text{g/L} \)), being within the therapeutic range of 0.8 to 2.1 \( \mu \text{g/L} \). Twelve of the 17 values were from infants younger than six months and gave digoxin assay results ranging from 0 to 1.4 \( \mu \text{g/L} \) (mean 0.5 \( \mu \text{g/L} \)). The remaining three values for infants six months old or older ranged from 0 to 0.2 \( \mu \text{g/L} \) (mean 0.1 \( \mu \text{g/L} \)). Additionally, nine cord-blood samples gave values for apparent digoxin from 0.9 to 1.2 \( \mu \text{g/L} \). These data prompt us to no longer use the TDx assay to digoxin in samples from infants younger than six months.

We analyzed with the TDx serum samples from 28 patients with total bilirubin values from 20 to 190 mg/dL. Twenty-six (93%) had digoxin assay values at or above the assay sensitivity, including seven (25%) with values within the therapeutic range. Total bilirubin correlated only moderately \( r = 0.668 \); digoxin (\( \mu \text{g/L} \)) = 0.0055 bilirubin (mg/dL) + 0.12 \( \mu \text{g/L} \) with measured digoxin, while TDx sample-blank intensity correlated very poorly with digoxin \( r = 0.388 \). Addition of purified bilirubin to drug-free samples to give a final concentration of 200 mg/dL and assay indicated that, on average, only 0.2 \( \mu \text{g} \) of the apparent digoxin per liter could be accounted for by bilirubin interference. Although the correlation between sample bilirubin and apparent digoxin value is poor, we currently consider it unwise to assay any sample with total bilirubin >50 mg/dL by the TDx Digoxin II method.

Ten patients' samples with creatinine values from 45 to 201 mg/dL were also analyzed. Assay values for apparent digoxin ranged from 0 to 0.3 \( \mu \text{g/L} \). While 40% of the values were at or above the assay sensitivity, we do not consider this group to be a problem in the assay.

References

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Performance Criteria for Appropriate Characterization of "(Highly) Sensitive" Thyrotropin Assays

To the Editor:

During the past few years immuno-metric assays for thyrotropin (TSH) have been developed in which two or three monoclonal antibodies are used in a "sandwich" type of configuration. Typically, one or two monoclonal antibodies directed against the \( \beta \)-subunit of TSH and immobilized on a solid surface bind the TSH from the sample, and another monoclonal anti-TSH labeled with a radioisotope, enzyme, or fluorescent or chemiluminescent tag is used to quantify the bound TSH (1–2).

The key feature of this new generation of TSH assays is enhanced sensitivity, even with relatively short incubation times. Clinical investigations have shown that some, but not all, of these assays allow a complete discrimination between TSH concentrations in sera of hyperthyroid and euthyroid individuals and thus will have a major impact on the diagnosis of hyperthyroidism (3–5). This is in addition to the diagnostic value in hypothyroidism, which they share with the conventional competitive-binding RIAs.

Although the full clinical potential of "sensitive TSH" measurements remains to be assessed, it has been suggested that TSH may become the best single screening test for thyroid disorders (6, 7). As a result, more and more clinical laboratories are in the process of switching to this new technology, and more physicians can be expected to rely increasingly, or solely, on "sensitive" TSH measurements when hyperthyroidism is suspected on clinical grounds.

In the wake of these developments, specific and clinically meaningful criteria are needed for the performance of so-called highly- or ultra-sensitive TSH assays to identify those methods that can actually deliver the sensitivity necessary for their broader clinical application. In particular, criteria are needed so that clinical laboratories can determine, unequivocally, whether their in-house TSH measurements, under routine conditions, actually qualify as "sensitive TSH tests" and thus warrant use in the diagnosis of hyperthyroidism.

Unfortunately, the currently most widely used definition of assay sensitivity as the "detection limit" or "least detectable dose" (8) is of little clinical relevance or utility in characterizing routine clinical performance when sera are analyzed on different days, by different technologists, with use of different batches of reagents, etc.

Further, appropriate data on assay precision are also lacking in the literature. Nearly all reports state the precision only for the middle of the normal range (TSH >1.5 milli int. units/L), but not in the lower or subnormal range where it would be of greatest significance for clinical decision making.

Consequently, my major objective here is to advocate an alternative concept for analytical sensitivity and to present a practical procedure based on this concept for categorizing TSH assays as either "sensitive" or "regular."

First, drawing primarily on previous work by Ekins (9), Wide and Dahlberg (10), and Kalman et al. (11), I propose that the concept of "the lower limit of interassay quantitative measurement" be adopted for the characterization of sensitivity instead of the "detection limit." This new term reflects interassay variability and also, indirectly, intra-assay errors. It is a precise measure of the lower limit of the clinically usable working range of an assay, i.e., it indicates the minimum analyte concentration that can be reliably quantified from assay to assay with an acceptable interassay CV. As such, it has considerable clinical significance and should be used particularly in assays where high sensitivity is a sine qua non. It is readily obtained graphically from the precision profile (Figure 1). Second, in order to ensure adequate precision, "quantitative measurement" in the above expression ought to be defined by an interassay CV not exceeding 10% or, at most, 15%.

Third, once sensitivity is so defined, then TSH assays can be readily evaluated (or monitored) for their suitability in the diagnosis of thyrotoxicosis. Specifically, I recommend the following simple evaluation procedure for TSH assays, to avoid major diagnostic errors (Figure 1):
1. Determination of the "in-house interassay precision profile curve" (9). Five serum pools with TSH concentrations corresponding to the subnormal (at least two pools) and normal range are included in eight to 10 consecutive routine assays to assess the precision actually achieved by a laboratory over the normal and subnormal range.

2. Graphical assessment of the "in-house lower limit of interassay quantitative measurement" or "lower limit of the assay working range" under routine conditions: this is determined from the intercept of the interassay CV (y-axis) equal to 10% or 15% with the "in-house interassay precision profile" curve.

3. Determination of the "in-house reference range" for euthyroid individuals.

4. Assay classification: an assay can justifiably be called "sensitive" and deserves further clinical trials in hyperthyroid patients if and only if the "lower limit of quantitative measurement" falls clearly below the normal range. Or, in other words, a clear-cut, complete separation of hyperthyroid and euthyroid TSH values can be achieved only if the inherent sensitivity of the TSH assay and the technical expertise of the laboratory staff are such that TSH concentrations below the normal reference interval are still reproducible with a CV of <15%.

For instance, the enzyme immunoassay (EIA) from Abbott Laboratories as performed in our laboratory under routine conditions met the fundamental requirement for a "sensitive" assay (Figure 1), and subsequent clinical investigations have confirmed that initial conclusion. By contrast, various other TSH assays, also designated by their manufacturers as "sensitive," have failed in our hands to meet the criteria, suggesting that they cannot be used to diagnose hyperthyroidism.

References