ma, the “test pool,” to give a final concentration of 100 mg/L (10-fold the estimated therapeutic dose). The “control pool” consisted of freshly pooled plasma diluted with the same volume of distilled water. The test and control pools were then analyzed alternatively five times for total bilirubin and unconjugated and conjugated bilirubin fractions (BUBC; Kodak). Interference was calculated as the difference between the means of the test and control pools with 95% confidence interval computed as:

\( (\text{mean}_{\text{test}} - \text{mean}_{\text{control}}) \pm 1.96 \frac{\sigma}{\sqrt{n}} \)

where \( s \) is the within-run imprecision of the method and \( n \) is the number of replicates per sample. Therefore, at an ICG concentration of 100 mg/L, we calculated analytical interference in the Ektachem 700 method for total bilirubin to be 56 ± 1.087 mmol/L. ICG caused no interference with the BUBC method on the Kodak Ektachem 700.

We further investigated ICG interference in the total bilirubin assay by the dose–response method (4). We mixed various volumes of two pools containing 0 and 10 mg of ICG per liter, and analyzed each test sample three times for total bilirubin. The analytical interference of ICG in Kodak Ektachem total bilirubin assay can be described by the slope (0.53; \( r = 0.978, n = 15 \)) of a linear regression plot of the concentration of ICG (x) versus the concentration of total bilirubin (y). Determining bilirubin concentration with the BUBC method with the Kodak Ektachem 700 avoids the false increase in total bilirubin results in the presence of ICG. Alternatively, the blood specimen should be drawn after the ICG has been cleared from the plasma. This occurs approximately 2 h after administration of a 0.5-mg dose of ICG per kilogram body weight (1).

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Sensitivity of CYCLO-Trac SP (Specific Monoclonal Immunoassay for Cyclosporin A)

To the Editor:
Kneipl and McPhillips (1) suggest that, in INCSTAR’s CYCLO-Trac SP immunoassay for cyclosporin A, a 10:1 ratio of methanol to specimen be used instead of 4:1. Their suggested extraction procedures would diminish the sensitivity of the assay by more than twofold. Their desensitized dynamic range may be suitable for their pharmakokinetic studies. However, as their own data in Figure 1 show (1), the proposed modification would result in increased CVs for target concentrations of 150–300 µg/L when the specific monoclonal assay is used (2). Also, their desensitized assay protocol would not be suitable for determining concentrations below 50 µg/L, values often seen in triple or quadruple therapies and autoimmune applications of cyclosporin A. If clean sampling of the methanol extract is difficult owing to an unusually high hematocrit, the kit protocol suggests that both the amount of methanol and specimen be doubled to maintain the ratio 4:1; i.e., 800 µL of methanol is used to extract 200 µL of specimen.

References


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Is Electrophoresis a Reference Method for CK-MB in Comparison with an Immunoenzymometric Method?

To the Editor:
Hall et al. (1) compared the new “second-generation” immunoenzymometric assay ("Tandem-E CKMB II") for creatin kinase isoenzyme MB (CK-MB) with an electrophoretic method (Beckman’s "Paragon") with respect to their ability to detect acute myocardial infarction (AMI). There are some statistical flaws in this method comparison.

The reference interval (mean ± 2 SD) for CK-MB was 0–18.0 µg/L for non-AMI patients, and 0–6.0 µg/L for persons with no cardiac disorders as evaluated with the parametric method. In a recent document from the IFCC, however, a nonparametric method is recommended (2). The latter method will probably give different values.

Evaluation of predictive values were performed only at a CK-MB concentration of 18 µg/L. When diagnosing AMI, one should prefer high sensitivity to high specificity. Thus, evaluation should have been done on concentrations of 6 or 9 µg/L, the latter being the limit recommended by Hybritech. Consecutive sampling (e.g., every 6th hour) and (or) other diagnostic tools (e.g., LD isoenzymes) (3) should reveal the differential diagnosis between AMI and other cardiac and noncardiovascular diseases.

Hall et al. (1) classified questionable AMI cases as "non-AMI" patients. Thus, undiagnosed cases of AMI may have been included in this group. These patients would better have been classified as AMI or as a separate "questionable AMI" group. Values exceeding the reference interval should not be classified as "falsely positive," unless the diagnosis is confirmed by independent criteria.

The authors state that the correlation between the immunochemical and electrophoresis methods was "excel-