Heparin’s ability to chelate calcium has been documented (1). Zoppi et al. (2) suggested similar effects on sodium analyses, but only for sodium heparin. The intended use of the Chiron capillary tubes (model no. 478504) is the measurement of pH and blood gases; the package insert made no mention of the use of these capillary tubes for electrolyte determinations.

Dry lithium heparin decreases measured sodium in adult blood (Fig. 1B), producing a mean decrease of −2.1 to −3.1 mmol/L at 150 kIU/L heparin and as much as −5.8 to −8.6 mmol/L at 500 kIU/L heparin (consistent with incomplete filling of the Chiron capillary tube) on the i-STAT and the ABL500 analyzer, respectively, but only minimally on the Vitros analyzer.

When adult venous blood samples (n = 30) were collected in heparinized tubes (<50 kIU/L), i-STAT, ABL500 (both using whole blood), and Vitros 750 (using plasma) agreed well. Deming regression analyses for the i-STAT vs the Vitros or ABL yielded slopes of 1.09 for the Vitros and 1.04 for the ABL, with y-intercepts of −12.2 mmol/L for Vitros and −4.40 mmol/L for ABL. Bland–Altman plots revealed no significant nonlinear trend.

We analyzed venous blood (500 μL) from discarded samples obtained with butterflies on 32 newborn infants with postnatal age <1 month picked at random from the NICU and collected in heparinized tubes (<50 kIU/L); whole blood samples (95 and 200 μL) were analyzed simultaneously on the i-STAT and ABL, respectively, but only minimally on the Vitros analyzer. Bland–Altman analysis (Fig. 1C) of the i-STAT and Vitros sodium values exhibited a difference up to −7 mmol/L (mean, −2.3 mmol/L). No difference (mean, 0.32 mmol/L) was observed between the i-STAT analyzer and the ABL500. There was no correlation between hematocrit (r = −0.29; P > 0.9) or protein (r = 0.28; P > 0.9) and the difference between the i-STAT and Vitros analyzers.

In summary, we report evidence of a negative bias in sodium values between the i-STAT or the ABL systems and the Vitros analyzer similar to the bias described previously between Corning and Vitros (3,4) in neonates but not observed in older children (5) or adults. Sodium measurements on blood collected in capillary tubes that contain high lithium heparin concentrations may produce an apparent bias up to −8 mmol/L. If lithium heparin must be used for sodium analysis with the i-STAT or ABL500 instruments, we recommend the use of full capillary tubes with low concentrations of lithium heparin.

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References

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To The Editor:
Recent correspondence to Clinical Chemistry (1,2) addressed the performance of the second generation Tacrolimus assay for the IMx analyzer (Abbott Diagnostics), with the former letter identifying nonequivalence in results from its predecessor and the latter considering performance approaching the lower limits of detection. Our own published results (3–5) have considered these points, and we report here our additional experience.

In their comparison of the second vs the first-generation assays, Garg et al. (1) described comparable coefficients of variation (CVs), but this is not the case when identical control samples are used in each assay at low concentrations (≈5 μg/L), e.g., 14.2% vs 42.4% at 4.2 μg of tacrolimus per liter of blood (1). In common with previous findings (3–5), Garg et al. (1) reported lower values with the second-generation assay using 36 samples of undefined origin. The slopes and intercepts reported for these various comparisons differed (as did the comparison methods applied), but Garg et al. (1) did not relate these data into the practical measurement of mean differences in assay results. Mean underestimates of 1–2 μg/L were reported for tacrolimus concentrations of 3–35 μg/L both by Wallemacq et al. (4), who used renal and liver recipients, and ourselves (3) (adult and pediatric liver and adult renal transplant recipients and patients with autoim-
mune disease). Of the explanations proposed for these differences by the various investigators, the lower recovery of the second-generation assay experienced by Garg et al. (1) in 1998 is not consistent with 1998 data from the Tacrolimus International Proficiency Testing Scheme (coordinator, Dr. D.W. Holt, St George’s Hospital Medical School, London, UK), from which can be calculated a positive bias and mean recovery of 114% (range, 102.1–121.7%) in 20 samples to which 3–28 μg/L tacrolimus was added and a similar underestimate relative to the results reported by the small number of centers using HPLC/mass spectrometry. A bias in assay calibrators (3) or differences in the contribution of tacrolimus metabolites (3, 4) may be a more likely explanation for the differences between the first- and second-generation assay results. Given the inherent variability in assay performance and tacrolimus pharmacokinetics, we still doubt whether a difference of 1–2 μg/L in assay results would have major practical impact on management by the realistic clinician. This is true both early after transplantation, when tacrolimus trough concentrations usually exceed 10 μg/L (but are subject to variability because of alterations in graft function, drug dosage, and coadministered medication), and later in clinically stable patients, when tacrolimus concentrations are often below 10 μg/L (and pharmacokinetic variability is lower but still subject to the influence of food intake) (6). In this lower range, where the increased sensitivity of the second-generation assay is advantageous, the CVs in tacrolimus measurements will span the differences of 1–2 μg/L between assay results.

Applying the concept of functional sensitivity (analyte concentration at 20% interassay CV) to the second-generation tacrolimus assay, Schambeck et al. (2) have defined a value of 3.1 μg/L for single measurements and recommend the use of two replicates at such concentrations. However, it is difficult to justify duplicate measurements realistically in terms of cost-benefit, the inherent variability in biological determinants of drug concentrations referred to above, or the use of concentrations as an adjunct to indicators of graft function and clinical condition in regulating dosage. The functional sensitivity also may not necessarily equate with the practical lower limit of quantification of the assay in routine use, particularly because two mode 1 calibrators (tacrolimus-free samples) are used to adjust the calibration curve in every assay. Thus, in defining a practical lower limit for routine assay of tacrolimus, it could be argued that intraassay variability (i.e., in relation to the mode 1 or tacrolimus-free calibrators run on the same carousel) may be of greater relevance than the corresponding interassay variability used in determining functional sensitivity. Within an assay, and like Schambeck et al. (2), we reported that tacrolimus concentrations of 1–2 μg/L blood are distinguishable from tacrolimus-free samples at high significance and without overlap (3). Moreover, recent data from the Tacrolimus International Proficiency Testing Scheme (coordinator, Dr. D.W. Holt) reported a CV of 17.1% on a sample containing 3.0 μg/L circulated to 184 centers and a CV of 21.7% on a corresponding sample of 2.0 μg/L analyzed at 172 centers. In addition to reporting a minimum detection limit of ~1.5 μg/L (3), our own routine experience using low concentrations of tacrolimus has been acquired from 179 interestassay measurements of control material containing 3 μg/L (obtained over 11 months with multiple operators and calibration curves). Despite a CV of 30.9%, acceptable results ranged from 1.1 to 4.5 μg/L when limits of 3 SD were used (outside which we routinely and immediately recalibrate the assay). Our additional experience with the second-generation assay over 28 months and >9500 samples is that results ≥2.0 μg/L have been obtained on known tacrolimus-free blood samples on only three occasions. This concentration (2 μg/L) is used as our minimum quantifiable concentration.

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Protein Zone Electrophoresis of Pleural Effusion: The Diagnostic Separation of Transudes and Exudates

To the Editor:
The cause of a pleural effusion is not always easily determined. Invasive procedures such as pleural biopsy are indicated only in patients with exudative pleural effusions. Therefore, a frequent early step in the evaluation of pleural effusions is to classify them as transudates or exudates.

The diagnostic criteria developed by Light et al. (1) characterize pleural