rate collection, a problem addressed in several papers (1–5). We do not discuss these problems here but instead focus on the accuracy and precision with which the volume is measured. We use lithium as an exogenous marker, having previously demonstrated the reliability of measurements of lithium as compared with volumetric measurements (6). Here we report the results of comparing both methods of estimating urinary volumes under different conditions for a total of 833 urine specimens.

In the first part of the study, we measured the volumes of 399 urine specimens collected during 20 weeks, mainly from diabetic patients. Most of these were 24-h specimens; 347 of them were from a medical ward, the remaining 52 from a surgical ward. The nursing staff were given written and oral instructions before collection started. To each collection vessel the contents of a 5.0-mL vial containing a 288 mmol/L LiCl solution were added before the collection was started, and the label of the vial was then transferred to the collection container. At the end of the collection period the nursing staff carefully measured the volumes of collected urine, using vessels graduated every 50 mL. The senior nurse was responsible for seeing that no one receiving therapy with lithium was included in this study.

The second part of the study involved 434 urine specimens from diabetic patients who were attending an outpatient-care unit. These urine specimens were collected either during 12 h (day or night) or 24 h, brought to the laboratory, and transferred to a vessel that contained 5.0 mL of the LiCl solution, as in the first part of the study. Total volume was measured in the same type of graduated vessel as above, all by the same laboratory technician; aliquots were removed for analysis for Li⁺.

The calculated urinary volumes (lithium method, 6) correlated reasonably with the volumes measured and estimated on the wards (r = 0.90 for the 347 medical-ward samples, 0.88 for the 52 surgical-ward samples), but a pronounced scatter was apparent: The residual variation (sₑᵦ), in percent of mean volume, was 13.5 and 19.4, respectively. In contrast, when an experienced technologist measured the urine volumes in the laboratory (part two of the study), the agreement with results by the lithium method was excellent and the correlation coefficient was 1.00.

To summarize: the volumes of 23% of the urine specimens measured in the surgical ward deviated from results by the lithium method by more than ±10%, while the corresponding figures for the medical ward and laboratory were 12 and 4%, respectively; 6% of the samples from each ward deviated by more than ±20% from the lithium-based calculations. The mean percentage of deviation from the calculated volume was 9% for the surgical-ward samples, 8% for the medical-ward samples, and 4% for the laboratory samples.

We investigated possible reasons for these striking differences between the volumes as measured on the wards and by the lithium-based method. Samples with a deviation exceeding ±10% were re-analyzed for lithium. The analytical results were the same in all these cases. Accordingly, an error in analyses for Li⁺ was evidently not the cause of these differences. The associated case records were reviewed for possible explanations, and the personnel involved on the wards were interviewed. The addition of two vials of lithium per vessel had been avoided by transferring the label from the lithium chloride vial to the collection container; in any event, this type of error would have explained only the cases where the calculated volume was smaller than volumetric measurement. In only one case had lithium chloride not been added to the collection container, and this result was excluded from the study.

We suggest that Li⁺ serves as a good marker for determining or checking volumes of urine specimens. If the collection containers contain the lithium marker at the beginning of urinary sampling, further mixing is not necessary (6). The precision of volume determination by the lithium method means that only a few milliliters of sample suffices for analyses and can be withdrawn and forwarded to the laboratory at the end of urinary collection. This simplifies the sampling and handling procedures. To allow urine specimens in a nursing unit and the transportation to the laboratory. Li⁺ does not interfere with determinations of 15 common urinary analytes (6). The lithium method is, of course, not suitable for urine specimens from patients who are receiving lithium, which is excreted in urine under such conditions (7, 8).

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Gunnar Ronquist
Gunnel Andersson
Yvonne Alin
Dept. of Clin. Chem.
University Hospital
S-751 85 Uppsala, Sweden

Adenosine Deaminase Activity In Serum of Kidney-Transplant Recipients during the Early Postoperative Period

To the Editor:

Adenosine deaminase (ADA, EC 3.5.4.4), which catalyzes the deamination of adenosine to inosine (1), has been studied with increasing intensity since the discovery that its genetic absence results in severe combined immunodeficiency (2).

T lymphocytes have significantly higher ADA activity than do B or null cells (3). Renal allograft rejection has been considered to be initiated by T lymphocytes (4). Yasmin et al. (5) suggested that ADA activity in mononuclear cells isolated from peripheral blood might be useful in confirming clinical diagnosis of allograft rejection, but did not provide data.

Hitherto, ADA in the serum of renal-transplant patients has not been studied. We have investigated the role of serum ADA as a possible rejection marker in 28 kidney-transplant recipients. The activity concentration of ADA in serum was monitored in these patients during the early postoperative period by the alternative method of Giusti (6), at 37 °C, in nonhemolyzed samples stored at −70 °C until analysis no more than a month later. We found for this method a between-day CV of 8.4% for a quality-control program based on duplicates (7).

The patients were treated with aza-
thioprine in doses of 2.5 mg/kg per day and did not receive prophylactic prednisolone after surgery, this drug being given only when rejection was diagnosed.

A diagnosis of allograft rejection was made according to clinical, biochemical, and isotopic data. We discarded 10 patients from the study: three who did not show evidence of rejection and seven for whom collection of serum on the initial day of rejection (R day) was not possible. Of the remaining patients, 13 showed rejection during the first two weeks after transplantation and another two during the first four weeks after transplantation.

We evaluated serum ADA on three separate days of our investigation: R day, four days before, and four days after R day. The values for ADA did not differ significantly from each other on any of the days according to Wilcoxon's t-statistical test for paired data. We also compared the results of R day with a reference group of 60 healthy subjects, and in this case there was a significant difference (p < 0.001) according to the Mann–Whitney U statistical test. We saw no sex-related differences (Student's t-test) in values for the reference group, in agreement with other authors (8).

Table 1 summarizes our results. They show that serum ADA is not a useful indicator of rejection, but further studies are necessary to complement these findings and to explain the high values for ADA in serum during the early postoperative period after renal transplantation.

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J avatar Frey
Serv. de Bioquim.
Hosp. de Bellvitge "Principe d'Espanya"
Feixa Larga s/n
L'Hospitalet de Llobregat
Barcelona, Spain

Theophylline Concentrations in Serum, Plasma, and Whole Blood Compared

To the Editor:
Serum is the most commonly used sample for analysis of theophylline, but only plasma or whole blood may be available in certain circumstances. Accurate monitoring of the concentrations of this drug would benefit from knowledge of how they interrelate among these three matrices.

Reports in the literature (1–6) are contradictory, ranging from conclusions that theophylline in blood is wholly plasma-bound, so that the concentration in whole blood could be validly expressed as (1−hematocit) times the concentration in plasma, to findings of no difference between theophylline concentrations in serum and blood.

In the course of a study on theophylline uptake, we obtained 67 parallel serum and plasma samples from 17 adult volunteers (ages 19–58 years, 11 women and six men). We re-examined the relationship among theophylline concentrations in blood, serum, and plasma.

Once the drug had equilibrated in the patients (after two weeks), we monitored the concentrations of theophylline during the course of a day by analyzing 10 samples collected at 0.5, 1, 2, 3, 4, 5, 6, 8, 10, and 12 h after the last dose. For 12 patients, five of these samples were taken, in duplicate, into a tube without anticoagulant (for serum) and into a 5-mL "Venoject" tube containing EDTA (for plasma and whole blood). The sample for serum was promptly centrifuged and the serum transferred to another tube. After withdrawing an aliquot of whole blood we centrifuged the EDTA-treated sample and removed 1 mL of plasma. Samples were kept at 4°C until analysis, not longer than 48 h.

We measured the theophylline by "high-performance" liquid chromatography with a Model 601 liquid chromatograph equipped with a Rhodyne Model 7105 injector, a 250 × 4.6 mm (i.d.) column packed with 10-μm (particle size) Silica A, and a LC-75 variable-wavelength detector set at 275 nm (all from Perkin-Elmer Corp., Norwalk, CT 06856). The flow rate for the mobile phase hexane/chloroform/tetrahydrofuran/methanol/acetic acid (75/14/7.6/3/0.4, by vol) was 3.0 mL/min. We used a SF-4100 computing integrator (Spectra-Physics) to quantify the results. Before the chromatographic separation, we extracted the samples with chloroform/isopropanol (95/5 by vol) by the method of Jusko and Poliscekuz (7), with beta-hydroxypropyltheophylline added as the standard. Tests with theophylline-supplemented samples of serum, plasma, and whole blood showed identical analytical recoveries for all three. Other xanthine derivatives, including metabolites of theophylline and of caffeine, did not interfere.

To minimize analytical variations, we used relatively large (500 μL) samples and analyzed all samples (blood, plasma, serum) for a given patient in the same run.

The within-run CV, estimated by analyzing the same serum sample (Utak serum toxicity control, 15 μg/mL) 10 times, was 2.8%. The between-run CV of 4.3% was calculated after analyzing the same control sample 125 times during six months.

Linear regression analysis of the concentrations of theophylline in (a) serum and plasma and (b) serum and whole blood for all 67 samples gave the following results: (a) serum = 1.07 plasma + 0.062 (r = 0.994), and (b) serum = 1.24 whole blood + 0.212 (r = 0.994). Because the y-intercept is essentially zero, the concentration in one matrix can be calculated from that in

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**Table 1. Serum ADA Activity Concentration, μkat/L**

| Patient's | Four days before rejection | Four days after rejection | Day rejection diagnosed | | Signif. |
|-----------|----------------------------|--------------------------|------------------------| | |
| Mean      | 0.36                       | 0.37                     | 0.35                   | | NS |
| Range     | 0.18-0.60                  | 0.16-0.68                | 0.16-0.68              | | NS |

NS, not significant. *Same mean (and SD) values for men and women.