to 2.0 mL of serum resulted in the desired concentrations with minimal dilution effects. There were no significant differences between standard curves obtained with two different lots of material. Nonspecific binding was routinely less than 8% of the total counts. Three serum pools with different concentrations of aldosterone were used in the analysis of variation. Between-assay means (and SD) for low-, medium-, and high-concentration pools (n = 5 each) were (ng/L): 103.2 (11.0), 338.7 (30.5), and 625.2 (53.1), with respective CV's of 10.7, 9.0, and 8.5%. Within-assay results for the same pools (n = 15 each) were (ng/L): 101.3 (8.1), 330.5 (24.1), and 620.5 (42.8), with respective CV's of 8.0, 7.3, and 6.5%.

Analytical-recovery studies were performed by adding various concentrations of aldosterone (between 75 and 600 ng/L) to a serum pool with a mean endogenous aldosterone concentration of 103 ng/L. The mean recovery of aldosterone added to this pool at four concentrations was 98% (range 95–101%). The lower limit of sensitivity, determined as twice the SD of the zero standard, was 43 ng/L.

Studies were performed to determine whether other serum constituents would interfere with the analytical recovery of aldosterone. Hemolysis up to 6.0 g of hemoglobin per liter, and icterus up to 100 mg of bilirubin per liter had no effect on the recovery of aldosterone at 300 ng/L. Lipemia (triglycerides up to 6.25 g/L) had no effect on recovery, whereas at triglyceride concentrations of 12.5 and 25 g/L, recovery of 300 ng of added aldosterone per liter was 112 and 116%, respectively.

Because albumin is the chief aldosterone-binding protein in serum, we performed another series of recovery experiments, using hyper- and hypoprotenemic specimens with various albumin/globulin ratios. The results indicate that the assay of aldosterone is unaffected by albumin in various concentrations; albumin, from 13 to 58 g/L, and albumin/globulin ratios ranging from 0.32 to 1.36 were associated with 96 to 103% analytical recovery of aldosterone at 300 ng/L.

Plasma aldosterone was evaluated in 50 control subjects, each judged to be free of disease, who were on a diet restricted in sodium, and whose results for a clinical chemistry profile were normal. The mean (and SD) was 200 (82) ng/L, which compares well with values reported by others: 155 (66) ng/L (1), 239 (123) ng/L (2), and 149 (63) ng/L (3). I also compared results obtained with this kit with those obtained according to the method of Abraham et al. (9) (Table 1). The method includes an extraction step with dichloromethane, followed by incubation with [3H]aldosterone and antibody, and separation of the antigen-antibody complex with dextran-coated charcoal.

The 125I labelled kit was compared to the [3H]aldosterone extraction method in a 25 patient split-sample correlation study. A linear least square regression analysis showed a correlation coefficient of 0.997 with a slope of 1.01, and a y-intercept of 7.22.

The two methods demonstrated comparable results over the range of 60 to 954 ng of aldosterone per liter. Statistical analysis of the correlation data by Student’s t-test for paired data indicated that the correlation between the two sets of values was highly significant (t-test, p <0.001) and that there was no significant difference between individual means.

I conclude that this radioimmunoassay kit gives rapid, accurate, and reproducible results. The ease and simplicity of the procedure, requiring no extraction and presenting no problems related to blank values, is such that many samples can be assayed with minimal manipulation.

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Two-Site Immunoradiometric Assay for Serum Ferritin Altered by a Change in Solid-Phase Support Medium

To the Editor:

A two-site immunoradiometric assay for measurement of serum ferritin was first described by Miles et al. (1). Several similar methods, including analogous enzyme-linked immunosorbent assays, have since been published not only for serum ferritin (2) but also for assay of other proteins (3, 4). These assays all involve a plastic surface, usually the inside of a counting tube, coated with a specific antisemur, which results in a solid-phase antibody.

Serum ferritin is assayed in this laboratory by a two-site immunoradiometric assay based on that of Miles et al. (1). We have used polystyrene tubes for
this assay and for the assay of serum vitamin B₁₂ by the method of Tibbling (5), which involves a centrifugation step. Because these tubes occasionally have small cracks, which make them unsuitable for centrifugation, the manufacturer changed the type of plastic used from polystyrene to styrene-acrylonitrile. We found this affects the serum ferritin assay results.

Polystyrene and styrene-acrylonitrile tubes were incubated with 500 µL of a 10 000-fold dilution of anti-liver ferritin antiseraum in sodium bicarbonate buffer (20 mmol/L, pH 9.6) at room temperature. The tubes were then washed twice with 2-ML portions of 50 mmol/L (pH 8.6) Tris containing 1 g of albumin per liter and twice with distilled water, drained, and stored inverted at 4 °C until used at the same time in parallel assays.

Purified human liver ferritin was used as a standard. Sera were obtained from human serum pools that were used for the routine quality control of this assay. Little difference was seen between the standard curves obtained with each type of tube. However, there was a marked difference in the case of quality-control sera, count rates being some 35% lower in the styrene-acrylonitrile tubes.

Both sets of tubes gave similar results for the standard ferritin solutions, so evidently the same amount of antibody was bound to each type of tube. The observed differences indicate a qualitative difference in the bound antibody, the nature of which is unknown to us at present but may be related to the serum inhibition phenomenon noted by Miles et al. (6).

Whatever the cause, it is clear that users of in-house assays of this type should be wary of changes in the composition of tubes used in their assay. Clearly, further studies are needed on the nature of the bonding between antibodies and various plastic surfaces, because this method is being applied to a wide variety of assays.

References

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Interference by Liposyn® with Common Laboratory Tests

To the Editor:

We evaluated the interference caused by Liposyn® (Abbott Hospital Supply) with the following tests: total protein, albumin, calcium, phosphorus, cholesterol, glucose, uric acid, total bilirubin, alkaline phosphatase, lactate dehydrogenase, aspartate aminotransferase, sodium, potassium, chloride, carbon dioxide, urea nitrogen, and creatinine. All these analytes were measured with use of a Technicon SMA (1), a Technicon SMA 18-channel analyzer (2) with a Vickers computer, and (except electrolytes) a Dupont aca (3). In addition, glucose, urea nitrogen, creatinine, sodium, potassium, chloride, and CO₂ were measured with an Instrumentation Laboratory seven-channel (4) analyzer. Assays were performed as described by the manufacturers except for the SMA 18 glucose (glucose oxidase) and cholesterol (enzymic).

Liposyn, a nutrient solution of safflower oil, egg phosphatides, and glycerine, adjusted to pH 8.0 with NaOH, is intravenously administered to patients, 500 mL during 6 h. Many of these patients are critically ill and urgent laboratory tests frequently are ordered during the period of administration. We found that such samples caused different amounts of interference when used with the different instruments. To evaluate this interference we diluted Liposyn 10-, 20-, 30-, and 100-fold with serum and analyzed these mixtures with the various instruments.

Liposyn assayed alone contained the following apparent concentrations, per liter:

1) SMA: total protein 14 mg, cholesterol 1980 mg, uric acid 2 mg, Na 0.5 mg, and creatinine 2 mg.
2) aca: Ca 66 mg, phosphorus 65 mg, cholesterol 650 mg, glucose 830 mg, uric acid 5 mg, and total bilirubin 190 mg.
3) Liposyn had no apparent concentration of any analyte measured with the SMA-Vickers or the IL instrument.

Liposyn was diluted 10-fold with serum; samples with similar composition might be expected if blood were drawn during administration of Liposyn. At this concentration Liposyn did not interfere with any assays on the SMA. It interfered with bilirubin (+500%) and LDH (−49%) on the SMA-Vickers. It interfered with total protein (−26%), phosphorus (+42%), glucose (+25%), and bilirubin (+24%) on the Dupont aca. It did not interfere with any of the tests on the IL seven-channel instrument.

At a Liposyn/serum proportion of 1/99 by volume, apparent concentrations with the aca were: total protein −4.4%, phosphorus +6.6%, glucose +5%, and total bilirubin −4.2%; with the SMA-Vickers they were: total bilirubin +9.3% and LDH −6% of the values before Liposyn was added.

Although we did not evaluate other hyperalimentation fluids, laboratories should be aware that such fluids can cause various problems with laboratory tests. For example, we would expect the same type of interference with Intralipid (Cutter Diagnostics, Berkeley, CA 94710), because the major difference is the use of soybean oil in place of safflower oil.

References
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Photographic Enhancement of Oligoclonal Bands in Cerebrospinal Fluid after Cellulose Acetate Electrophoresis

To the Editor:

We confirm the findings of van der Helm et al. (1), who suggest that electrophoresis on cellulose acetate can be used to identify oligoclonal bands in cerebrospinal fluid (CSF). We also agree that the conditions for electrophoresis and staining are critical, but we have managed to overcome the effect of these variables by use of photographic enhancement, a modification of a tech-