fluctuations in urine urea nitrogen reported by other Astra users with this method.

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Evaluation of a Kit to Measure HDL Cholesterol (HDL-C) in Serum

To the Editor:

A clinic laboratory got low values for serum HDL-C on using a kit supplied by Electro-Nucleonics, Inc.[1] In the kit, the precipitating reagents (phosphotungstate and $MgSO_4$) are supplied as solids in a tube, to which 0.5 mL of serum is added according to the manufacturer's package insert.

We analyzed split human serum samples in parallel with such kits and using our in-house phosphotungstate method, a procedure like that of Lopes-Virella et al. (*1*), except that we adjust the pH of the phosphotungstate reagent to 7.4 or slightly less (*2*), and add only 80 μL of phosphotungstate plus 20 μL of $MgCl_2$ reagents to 1.0 mL of serum. Values obtained with the kit were from 30 to 170 mg/L lower than results with the in-house method, differences averaging 18.2% (SD 5.4%, n = 16) for values ranging from 280 to 1120 mg/L. Such differences would cause many samples with total cholesterol/HDL-C ratios near 5.3, based on the in-house method, to have ratios near 6.5 with the kit. These higher ratios would imply an importantly greater risk of coronary artery disease (*3*).

We further evaluated the kit by titrating (*4*) three human serum samples with increasing ratios of precipitant to serum. To a series of kit tubes (each contained the prepackaged precipitant), successively decreasing volumes of serum were added at room temperature. After their contents were mixed, the tubes stood for 15 min before being centrifuged at 4 °C for 30 min. The cholesterol in each supernate was measured with use of an ABA-100 discrete analyzer and Bio-Dynamics/*bmc* enzymatic cholesterol reagents. The lipoprotein content of each supernate was monitored by agarose gel electrophoresis (*5*) to identify the "true" HDL-C value (the cholesterol concentration in the supernate from which all VLDL and HDL had been removed by precipitation without detectable loss of HDL). Results from one serum were as follows:

Tube no.	*Serum vol, mL*	*Apparent HDL-C, mg/L*
1	1.8	470
2	1.5	480
3	1.3	390
4	1.2	380
5	1.1	400
6	1.0	390
7	0.9	380
8	0.8	380
9	0.7	380
10	0.5	350

Because agarose gel electrophoresis showed no LDL or VLDL in the supernates of tubes 3–10, the "true" HDL-C concentration appears to be 390 mg/L (tubes 3–6). The HDL-C concentration measured by our in-house phosphotungstate method was 390 mg/L, while the concentration measured with 0.5 mL of serum added to the kit tube was 350 mg/L. Similarly, in the other two titrations also, the HDL-C values were low when 0.5 mL of serum was added to the kit tube. With 1.0 mL per tube, however, agreement was satisfactory.

The clinic laboratory then split a group of serum samples, measured the HDL-C in one aliquot with use of 1.0 mL of serum per kit tube, and sent us the other aliquot for analysis by our in-house method. All values measured at the clinic with the kit were 10 to 40 mg/L lower than our values, which ranged from 260 to 630 mg/L. The differences between the methods averaged 5.8% (SD 3.2%, n = 8). Although these differences were statistically significant, they were deemed acceptable because they were small and consistent. Compared with the earlier differences when 0.5 mL per tube was used, the improvement is obvious. Data from the other titrations suggest that a serum volume slightly less than 1.0 mL (e.g., 0.85 mL) would be optimal, but the gain in accuracy is apt to be offset by the difficulty in measuring such a volume.

The above findings were communicated to the kit manufacturer, Electro-Nucleonics, Inc. Their response pointed out that the package insert in the kit makes no claims about the total cholesterol/HDL-C ratio. Further, they pointed out the recognized facts that the results of HDL-C assays depend on conditions and reagents and that there were differences in both of these when our phosphotungstate reagents and method were compared with their kit and the instructions in the package insert.

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Cefoxitin Interference with Urinary 17-Hydroxycorticosteroid Determination

To the Editor:

Because of the marked minute-to-minute variation in plasma cortisol concentrations, either free cortisol in urine or total 17-hydroxycorticosteroids (17-OHCS) excretion is the best index to steroid output. The normal response to dexamethasone has been better established for urinary 17-OHCS than for free cortisol (*1*); thus, accurate determination of urinary 17-OHCS is essential.

With the large increase in the number of pharmacological agents in recent

[1] Use of products from named companies does not imply that the products are superior to similar products of comparable grade from other companies, nor does it constitute endorsement of the products by the United States Air Force.