

Table 1. RIA Standard Curves for DHEAS Compared on Using PEG and Charcoal Separation Methods^a

	PEG	Charcoal
Nonspecific binding (% of total)	2.1	2.1
Binding for zero standard (% of total)	20.0	20.0
Slope	0.851	0.839
ED ₅₀ , ng/L	262	271
Within-assay CV, %		
at 20% displacement	10	8
50% displacement	7	8
80% displacement	15	14

^a[³H]DHEAS was used as the tracer.

longer affected by any partial reversal of the binding equilibrium, which is a cause of assay "drift." The length of time required for the completion of charcoal separation effectively limits the run sizes that can be accommodated with that procedure.

The PEG separation requires the presence of the whole serum (or plasma) or the addition of normal gamma globulin to all tubes, to form a substantial pellet at the centrifugation stage. The method is limited to the use of analytes with a relative molecular mass of <1000 approximately, because larger molecules such as polypeptides may be partly precipitated by PEG solution at this concentration (150 g/L). However, the method is well suited to most of the analytes ordinarily labeled with tritium, such as steroids and nucleotides.

As an example, we show in Table 1 a comparison of dehydroepiandrosterone sulfate (DHEAS) standard curves obtained by using either a standard charcoal separation protocol or the PEG method. For the PEG separation the RIA tubes (700- μ L incubation volume) were treated with 2.2 mL of a 200 g/L solution of PEG-6000 (BDH, Kilsyth Vic, Australia), which had been pre-cooled to 4 °C, and 100 μ L of a 5 g/L solution of normal rabbit gamma globulin. The tubes were centrifuged (15 min, 2400 \times g, 4 °C) and the supernates were decanted and discarded. The pellets were redissolved with the addition of 1 mL of distilled water, allowed to stand for 30 min at room temperature, vortex-mixed, and added to 10 mL of Aquasure liquid scintillation cocktail (Dupont-NEN, Sydney). Data obtained with the two separation methods produced virtually superimposable standard curves with similar performance characteristics.

References

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Fingerprick Cholesterol Testing with the "Reflotron"

To the Editor:

Instant dry-chemistry analyzers for cholesterol screening in the workplace or the doctor's office are gaining in popularity, but their accuracy is largely unknown. We have investigated one such system, Boehringer's "Reflotron," in which whole blood is used. This analyzer is widely used in screening programs in the U.S.A.

When we first tested it in our Wageningen laboratory in September 1985, we found a Reflotron cholesterol value in the fingerprick blood of 69 normolipemic volunteers averaged 5.0 mmol/L (SD 1.3), 8% (SEM 1%) lower than the values for venous serum that had been obtained a few minutes earlier (n = 35) or later (n = 34) from the same subjects, and which had been analyzed with an enzymatic CHOD-PAP method of proven accuracy (bias for control sera from the Centers for Disease Control: 1%). A new batch of Reflotron strips yielded a downward bias of 12% (SEM 1%) when tested in October 1986 in a similar fashion in another 32 volunteers. Different analyzers yielded the same bias. This was to be expected, because the calibration data do not reside inside the analyzer but on the test strips.

In December 1986 we investigated in our Nijmegen laboratory 71 hyperlipidemic patients, whose cholesterol values ranged between 4.4 and 17.9 mmol/L (mean 8.8, SD 2.7) and triglycerides between 0.6 and 16.0 mmol/L (mean 2.7, SD 2.7). We now found a mean downward bias of 10% (SEM 1%) of the Reflotron method relative to values obtained in venous serum by a highly accurate reference method (bias for CDC sera, <1%). For patients with cholesterol values <10 mmol/L the bias was 8%.

In large-scale screening such a mea-

surement bias will have major consequences. For instance, 25% of white American men of ages 40 to 44 have a cholesterol over 228 mg/dL (5.9 mmol/L) (1). If values are underestimated by 9%, then the proportion of men having cholesterols >228 mg/dL drops to 10% (1); the other 15% are now dismissed as normocholesterolemic.

Better accuracy with the Reflotron was obtained in recent studies in the U.S.A. (Bachorik P., personal communication), suggesting that the manufacturer can solve the problem. Still, our data do show that the accuracy of desk-top analyzers should not be taken for granted. Suitable control materials should be available for fingerprick cholesterol analyzers, and they should be used routinely in the field to monitor accuracy.

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Reference

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Bacterial Interference with Measurement of Creatinine in Stored Plasma

To the Editor:

The creatinine concentration in plasma is commonly measured to aid in early detection of rejection in renal-transplant patients.

In our laboratory, we retain the last plasma specimen from the renal-transplant patient and re-analyze it when a new specimen is received from the same patient. We then report the original and latest result obtained for the old specimen, along with the result for the new one. This allows result interpretation to take some account of day-to-day analytical variation. All mea-