umbelliferone. The rate of signal development is directly proportional to the amount of prolactin in the specimen. Analysis of as many as 24 samples is completed within 37 min, and the prolactin concentration is calculated from a stored calibration curve. Prolactin calibrators and controls are assigned values relative to the World Health Organization 2nd International Standard for prolactin (83/562).

The dynamic range of the assay is 0–200 μg/L, with a detection limit of 0.6 μg/L (the lowest concentration exceeding the 95% confidence limit for the zero calibrator). No detectable cross-reactivity was observed with placental lactogen (100 mg/L), lutropin (5000 int. units/L), follitropin (5000 int. units/L), thyrotropin (2 int. units/L), or chorionic gonadotropin (1 × 10^8 int. units/L). Cross-reactivity with growth hormone (somatotropin; 1.0 mg/L) was 0.039%. There was no interference by high concentrations of bilirubin (0.5 g/L), hemoglobin (7.5 g/L), or triglycerides (9.9 g/L). Analytical recovery of prolactin in human serum ranged from 91% to 109%, with a mean of 99%. No substantial difference in recovery was observed between serum, EDTA plasma, or heparin plasma specimens. Intra-assay, interassay, and total CVs for 40 assays performed with each of 10 IMX instruments averaged 5.0%, 2.7%, and 5.7%, respectively, over the range of 8.1–41.1 μg/L. The mean prolactin value determined for 48 healthy males was 3.2 μg/L (range 0.1–6.0 μg/L); for 46 healthy females, the mean prolactin value was 5.6 μg/L (range 0.5–18.1 μg/L).

Determination of prolactin in patients’ specimens with use of the IMX assay correlated well with that of both the Tandem®-R Prolactin Immunoassay Radiometric assay (Hybritech Inc., San Diego, CA) and the Prolactin MAIAClone® assay (Serono Diagnostics Ltd., Surrey, U.K.). Determination of prolactin values for 250 patients’ specimens with both the IMX (y) and Hybritech (x) assays yielded r = 0.996, slope = 0.996, y-intercept = 2.44, and standard error of the y-estimate = 5.22. A similar analysis of 100 patients’ specimens with the Serono assay (x) yielded r = 0.986, slope = 1.037, y-intercept = 1.23, and standard error of the y-estimate = 5.85.

Reference

Consistency of the Algorithm for \( p_{O_2} \) Correction in Blood-Gas Analysis, B. Gouret, R. Andriamahatra, Y. Gourmelin, and A. Truchaud (Lab. Biochim., Centre Hospitalier Général, rue Saint Fiacre, BP 218, 77 108 Meaux, France)

A recurrent problem in blood-gas instrumentation is the accuracy of determining \( p_{O_2} \) (1). The directly measured \( p_{O_2} \) values are corrected by built-in adjustments in all models of blood-gas analyzers. The accuracy of \( p_{O_2} \) displayed by the instrument depends on the consistency of the algorithm for corrections. No unequivocal method exists for experimentally determining with absolute accuracy the partial pressures of blood gases. Tonometry of whole blood is acknowledged as the Reference Method for assessing \( p_{O_2} \) accuracy (2). To determine whether the correction algorithm for \( p_{O_2} \) measured with the ABL 500 (Radiometer, Copenhagen, Denmark) is valid, we used film (Laue bulb; Eschweiler, Kiel, F.R.G.) and bubble (Corning Model 184; Ciba-Corning, Medfield, MA) tonometers and fresh human blood. We also compared results obtained by aspiration and injection modes.

Data for linearity (range: 0–600 mmHg) followed two patterns. In the range of 0–150 mmHg, the assigned and the measured values were identical. Above 150 mmHg, \( p_{O_2} \) was slightly overestimated, by 2.2% and 3.4% at 600 mmHg with film and bubble tonometry, respectively, for samples measured in the injection mode. With film-tonometered samples, \( p_{O_2} \) values displayed by the instrument were not identical for the injection and aspiration modes, especially for high values of \( p_{O_2} \) (Table 1). Values obtained with the ABL 500 (n = 100), compared with those obtained with the Corning 184, showed the same trend for overestimation at values >150 mmHg (allometry line: \( y = 1.03x - 1.94 \)). These discrepancies could not be explained by other features of this instrument, which overall exhibited a high degree of precision (within-run precision, n = 20: CV ≤0.37% at \( p_{O_2} \) of 40 and 160 mmHg). The differences observed between the two types of tonometers may be accounted for by the bubble effect, i.e., increasing the \( p_{O_2} \) by 7.6 mmHg at 600 mmHg. This effect is determined by the bubble diameter, the hydrostatic pressure in the tonometer vessel, and the surface tension of the liquid surrounding the bubble (3).

The differences observed with the two modes (aspiration and injection) are more likely explained by the nature of the algorithm itself, which was established by Radiometer with use of capillary tubes and the aspiration mode (4). Under these conditions, the sample volume and the degree of contamination are known precisely. The sample volume is larger and more variable and the amount of contamination is not identical when syringes and the injection mode are used. The algorithm also includes factors that compensate for the gas/liquid ratio and systematic deviations such as sample contamination in the tubing and measuring chamber. These may not be identical for both modes of sample introduction.

The differences observed with patients’ samples determined with both the Corning 184 and the ABL 500 argue also for the partial inadequacy of the empirically determined algorithm, which may be accurate for aspiration but is not appropriate for injection of the samples. Theoretically, however, this algorithm may be modified to obtain the true \( p_{O_2} \) value for any set of operating conditions or modes of sample introduction.

Note added in proof: In light of these findings of inaccuracy for injected samples with high \( p_{O_2} \) Radiometer has decided to modify the software of the ABL 500.

| Table 1. Mean (SD) \( p_{O_2} \) Values (mmHg) by Bubble and Film Tonometry with the ABL 500 (n = 23) |
|---------------------------------|------------------|-------------------|
| **Assigned values** | **Bubble tonometry:** | **Film tonometry:** |
| | Injection | Aspiration | Injection |
| 149.5 | 151.0 (0.3) | — | 150.5 (0.4) |
| 179.9 | 183.0 (0.3) | — | 181.2 (0.3) |
| 212.0 | 216.2 (0.5) | 213.2 (0.5) | 214.0 (0.6) |
| 284.0 | 291.2 (0.3) | 286.2 (0.7) | 288.0 (0.6) |
| 355.0 | 364.6 (0.4) | 358.6 (0.5) | 361.7 (0.7) |
| 500.0 | 516.5 (0.7) | 501.8 (0.9) | 510.0 (0.7) |
| 600.0 | 620.6 (0.5) | 602.6 (0.8) | 613.0 (0.6) |

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References


The final step in steroid radioimmunoassay consists in separating the free (F) from the immunologically bound (B) radioactive steroid. This separation is usually carried out with a charcoal–dextran reagent.

Although widely used, this charcoal–dextran separation step is very time-consuming. This is why the introduction of scintillation proximity assay (SPA) reagent (1–3), which eliminates the need for both charcoal–dextran reagent and scintillation liquid, may represent considerable progress in the daily practice of steroid radioimmunoassay.

We compared the results of the radioimmunoassay of three steroids—androstenedione (A), dehydroepiandrosterone (DHEA), and 11β-hydroxyandrostenedione (11βOHA)—by a technique used for several years in our laboratory (4), which includes a dextran–charcoal separation step, with the results obtained by using the newly introduced SPA reagent.

The SPA reagent consists of fluorescer-impregnated microspheres coated with anti-rabbit antisera; it was obtained from Amersham France. The working principle is as follows: During radioimmunoassay with \(^{3}H\)-labeled steroids, bound labeled ligand (B) is in close proximity to the microspheres, allowing the emitted energy to activate the fluorescer and produce light. Because the assay is carried out in 0.4 mL of buffer, most of the free labeled steroid molecules (F) are too far from the microspheres to promote the transfer of energy.

Briefly, simultaneous radioimmunoassay of A–DHEA–11βOHA was carried out after extraction, followed by Celite partition chromatography (4). We used androstenedione-ß-hemisuccinate/bovine serum albumin (BSA), DHEA-ß-carboxymethylxoline/BSA, and 11βOHA-ß-carboxymethylxoline/BSA antibodies (4) along with the corresponding tritiated steroids.

The classical technique (technique 1) involves overnight incubation at 4°C, followed by charcoal–dextran separation (4). In the new procedure with SPA reagent (technique 2), a chromatographic eluate (0.1 mL), specific antibodies (0.1 mL), tritiated steroids (0.1 mL), and SPA reagent (0.1 mL) are mixed together in minivials overnight at ambient temperature before counting the bound radioactivity.

Sensitivities of the standard curves of the two techniques were evaluated as the quantities of cold steroid displacing 50% of the radioactive ligands. With technique 1, 50% ligand displacement was obtained with 75 pg of A, 130 pg of DHEA, and 100 pg of 11βOHA. With technique 2, this required 93 pg of A, 180 pg of DHEA, and 85 pg of 11βOHA.

Reproducibility of results for standard and patients’ sample duplicates was better with SPA reagent (CV = 1.6–6%) than with technique 1 (CV = 2.6–8%).

Comparison of the results of 130 patients’ plasma concentrations obtained with the two techniques yielded the following regression equations (\( x = \text{technique } 1, y = \text{technique } 2 \): \( y = 0.92x + 0.19 \) (A); \( y = 0.97x + 0.17 \) (DHEA); \( y = 0.97x - 0.05 \) (11βOHA).

The CVs obtained for the same plasma control sample measured by the two techniques were very similar: 8.9% (A), 5.9% (DHEA), and 7.8% (11βOHA) for technique 1 and 9.0%, 6.7%, and 4.7%, respectively, for technique 2.

Overall, the analytical performances of the assays for the three steroids carried out with the SPA reagent appear very similar to those obtained by using the classical technique involving a charcoal–dextran reagent. However, substantial time was saved by using the SPA reagent technique, because filling vials with scintillation liquid, adding charcoal–dextran reagent to each assay tube, waiting before centrifugation, and decantation into the vials were all eliminated.

References

Effect of Hyperglycemia on Plasma Sodium and Potassium Concentration Revisited, Robert A. Shalowitz, Ronald L. Gingerich, Janet B. McGill, and Jay M. McDonald (Depts. of 1 Pediatrics, 2 Med., and 3 Pathol., Washington Univ. School of Med., St. Louis, MO 63110)

During hyperglycemia, intracellular water shifts to the intravascular space, which results in a decrease in the concentration of plasma sodium. This phenomenon was basically observed by Seldin and Tarail (1). Katz (2) analyzed the specifics of the relationship, showing that for each 1000 mg/L (5.6 mmol/L) increment in serum glucose, there is an expected decrease of 1.6 mmol/L in serum sodium. However, a recent study by Strand et al. (3) found a poor correlation (\( r = -0.556 \)) between simultaneous glucose and sodium concentrations in a cross section of patients with spontaneous hyperglycemia. These results were in contrast to those of McNair et al. (4), who found a consistent decrease in sodium (\( r = -0.61 \)) and an increase in potassium (\( r = 0.37 \)) with hyperglycemia in patients with diabetes mellitus. Therefore, we undertook this study to analyze the consistency of the effects of hyperglycemia on plasma sodium and potassium under controlled condi-