Bedside Heparin Monitoring by Activated Partial Thromboplastin Time Measured with a Dry Reagent, Rikard Linder, Lars Grip, Eva Olson, and Margareta Blomh"ack (Depts. of Cardiol. and Lab. Med./Blood Coag. Res., Karolinska Hosp., S-171 76 Stockholm, Sweden; author for correspondence: fax 46-8-324597; e-mail rl@cardio.ks.se)

Activated partial thromboplastin time (APTT; also coagulation surface induced time [1]) is a routine method for monitoring heparin treatment [2-5]. This time is influenced by the type of reagent, the method of analysis, and the laboratory instrument used; thus, the result will vary from one laboratory to another [6, 7]. Variation in heparin sensitivity between batches of reagents produced from the same manufacturer has been reported [8], and attempts to standardize APTT reagents have not been entirely successful [9, 10]. Temperature and duration of sample storage until centrifugation and analysis also affect the APTT [11]. The APTT in whole blood decreases with time if left without centrifugation [12] because the heparin is neutralized by plasma proteins, the most important of which is considered to be platelet factor 4. Among the bedside methods developed for APTT measurement, some appear to be reliable alternatives to a central laboratory method [12, 13]. Here we compare a dry-reagent bedside system for APTT analysis with a central laboratory method for monitoring patients treated with heparin for unstable angina or after percutaneous transluminal coronary angioplasty (PTCA).

Seventy-five patients (ages 36-84 years) treated with heparin at the Department of Cardiology, Karolinska Hospital, Stockholm, Sweden, for either unstable angina (n = 54) or after PTCA (n = 21) were included in the study between November 1994 and April 1995. We also sampled healthy volunteers (n = 15) and patients without heparin treatment (n = 10) to compare the two APTT methods at values near the reference interval.

The procedure followed was part of the routine care and in accordance with the ethical standards of our institution. Samples from all patients were taken only once, by a well-trained nurse applying minimum stasis. Venous blood from either an intravenous catheter or from a disposable needle was drawn into two 5-mL Vacutainer Tubes (Becton Dickinson, Rutherford, NJ) containing 0.5 mL of 129 mmol/L trisodium citrate (pH 7.4). One tube was sent for APTT analysis to the central laboratory at the hospital and one tube was used for APTT analysis by the Thrombolytic Assessment System (TAS; Cardiovascular Diagnostics, Raleigh, NC). The latter analysis was performed within 15 min by one specially trained research nurse, who had performed >100 analyses with the TAS before the study.

The TAS is a portable instrument designed for bedside monitoring of APTT with a dry reagent system. The dry reagent contains phospholipids derived from a chloroform extract of dried rabbit brain, 12.5 mmol/L calcium chloride, aluminum magnesium silicate as a contact activator, buffers, stabilizers, and paramagnetic iron oxide particles (PIOP). A drop of citrated whole blood (or plasma, although we did not study it here) is placed on a disposable test card and inserted in the instrument. The blood mixes with the reagent in a reaction chamber. The PIOP move under the influence of a pulsating magnetic field. A light source shines on the PIOP, which reflect light onto a photodetector that records signals. Plasma or blood entering the reaction chamber will dissolve the dry reagent and result in a change of reflected light when the PIOP start to move. The signal then produced triggers the instrument to start recording the clotting time. Clotting results in a slowing and eventually a cessation of PIOP movement, after which a computer algorithm is used to calculate the clotting time. The time (APTT) is displayed in seconds.

For comparison, we used the routine method for APTT analysis at the hospital. This measures the coagulation time, after contact activation, in platelet-poor plasma (obtained after centrifugation at 2000g for 15 min). The reagent used (Cephotest; Nycomed, Oslo, Norway) consists of a factor XII activator, ellagic acid, and a phospholipid. Recalcification was achieved with calcium chloride, 25 mmol/L. We performed the test with a Thrombolyzer (Behnk Electronic, Norderstedt, Germany), an optical clot-detecting device. For control we used an upper-concentration range control, Scandipath, three times a day, and a lower-range control, Scandinorm, once a day (both from Stago, Asnieres Cedex, France). The CV for this method was not measured during the study but in previous studies has been 4.6% for the high control and 3.4% for the low control. The APTT reference interval is 24-35 s, and 60-89 s is the target value during heparin treatment.

APTT analysis was performed twice on two parallel TAS instruments; the results, TAS A and TAS B, each represent a mean of two measurements on the same instrument. For 5 of the 75 patients, one of the two APTT values from either of the TAS instruments was missing, and for 1 patient one of the two values was missing from both analyzers; in these cases we used the one value obtained instead of the mean. Quality control of the two TAS instruments was performed once daily with the upper-range control plasma (Scandipath; APTT ~45 s).

In the statistical analysis APTTs from the two TAS instruments and APTTs from TAS A and the Cephotest system were compared. The Cephotest APTT upper limit of analysis is 180 s, whereas the range for the TAS extends to 300 s. All values >180 s (n = 11) with either method were excluded from the regression analysis and from calculating the difference between the methods. Results for the remaining 64 patients were included in the comparison calculations. The differences in absolute terms are given in mean ± SD. We also used linear regression analysis, reporting the slopes, the intercepts, correlation coefficients, and, to express the dispersion about the regression lines, the standard error of the estimate (S_{est}). We calculated the CV for the TAS instruments at the therapeutic value to ascertain similarity to the CV for the TAS measurements of the control plasma. To calculate this CV, we used pairs of values (double measurements) in the APTT range of 60-89 s for any of the instruments (only one instrument's APTT reading had to be within this range), inserting these into Dahlberg's formula for estimation of the SD from duplicates: SD = \sqrt{(d^2/n)}, where d = difference between the values and n = number of measurements with one of the instruments.

The CV with the Scandipath control for TAS A was 7.6% and for TAS B 9.7%. The CV for duplicate measurements in the therapeutic range 60-89 s was 8.16% (TAS A) and 9.16% (TAS B).

Including all patients and volunteers, without regard to heparin treatment, the correlation between the two TAS instruments was r = 0.98 and between the TAS A instrument and the Cephotest system was r = 0.93 (Fig. 1). The differences and the correlations, by heparin treatment or not, are presented in Table 1. In general, and especially for values above the therapeutic interval for APTT, the TAS had a tendency to report higher values than the comparison system.
Decision-making regarding the infusion rate of heparin at the clinic is based on the therapeutic interval of 60–89 s for APTT. For values >89 s or <60 s, a progressive and stepwise change of the infusion rate is made. If the TAS readings had been used for control of the heparin infusion rate, 20 patients of the 75 would have received a somewhat different infusion rate (8 patients a higher rate, 12 patients a lower rate—all 20 being within a one-step difference in treatment of 2160 IU/24 h) from that implied by the comparison system measurement. In no case, however, would a TAS APTT have led to an adjustment of the heparin infusion rate that would have been contradictory to an adjustment based on the corresponding comparison method APTT (i.e., the TAS never indicated an increase in the dose when the Cephotest was indicating a decrease, or vice versa).

The correlation between the two APTT systems was good. The precision of the TAS APTTs was somewhat less than that of the comparison method (in earlier studies), but still was <10% (CV).

A bedside system for APTT analysis at the clinical ward can reduce the personnel and time-demanding procedures required for communication with the central laboratory. The use of a bedside system that enables an APTT to be analyzed within minutes may also facilitate fast decision-making regarding the heparin infusion rate and will be of special benefit for patients in cardiology care.

References

Serum Lactate and Lactate Dehydrogenase in High Concentrations Interfere in Enzymatic Assay of Ethylene Glycol, Anne F. Eder, Yvonne G. Dowdy, Jo Ann M. Gardiner, Bryan A. Wolf,* and Leslie M. Shaw (Dept. of Pathol. and Lab. Med., Univ. of Pennsylvania School of Med. and Hosp. of the Univ. of Pennsylvania, Philadelphia, PA 19104; *address for correspondence: 217 John Morgan Bldg., 3620 Hamilton Walk, Philadelphia, PA 19104-6082; fax 215-573-2266, e-mail wolfb@mail.med.upenn.edu)

The enzymatic assay to measure ethylene glycol in serum utilizes glycerol dehydrogenase in an oxidation reaction that produces NADH [1–3]. The concentration of NADH is determined spectrophotometrically by measuring absorbance at 340 nm. Although this enzymatic assay is specific for ethylene glycol, other reactions that generate NADH in patients' sera may produce false-positive results. This interference is observed in serum samples with high concentrations of l-lactate dehydrogenase (LD) and lactate.

We first documented false-positive results in the ethylene glycol enzymatic assay during the evaluation of a 23-year-old man with a 1-year history of dilated cardiomyopathy and pulmonary hypertension who presented with shortness of breath, productive cough, hemoptysis, nausea, and vomiting. Medications he was taking included captopril, digoxin, hydrosemide, and cimetidine. Physical examination found him to be