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Comparison of Five Point-of-Care Prothrombin and Activated Partial Thromboplastin Time Devices Based on Age of Blood Sample

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Abstract: Delays in processing statium (STAT) blood samples have led to the production of an increasing number of point-of-care tests. Product inserts recommend measuring blood samples immediately after procurement, suggesting that delays may invalidate the test results. We studied the effect of the age of blood samples on point-of-care (POC) prothrombin time (PT) and an activated partial thromboplastin time (aPTT) result. Informed consent was obtained from 11 patients undergoing cardiopulmonary bypass (CPB). Blood samples (40 mL) were taken from each patient. Each blood sample was used to perform five PT tests and six aPTT tests on five POC devices (Gem PCL, Hemochron 801, Hemochron Jr. Signature, Hemochron Response, Rapidpoint Coag) at three different sample ages [<60 s (fresh blood), 10 and 18 min after sample collection]. Blood samples were procured in a plastic syringe devoid of air bubbles, which was left undisturbed between tests but was gently agitated before initiating the 10- and 18-min tests. For tests requiring citrated whole blood, a fraction of each sample was anticoagulated (3.8% citrate) at each age. Statistical analysis was used for comparison of test results for fresh blood to aged samples (10 and 18 min). Test values were recorded as International Normalized Ratio (INR) and seconds for PT and aPTT, respectively. Two devices, the Hemochron 801 and Hemochron response showed statistically, although not clinically, significant variation in PT test results when the samples were aged to 10 and 18 minutes. As for aPTT results, Hemochron 801, Hemochron response, Hemochron Jr. signature, and Gem PCL showed statistically significant variation at 18 minutes. One device (Hemochron 801) reported results with 10-min aged blood that were statistically different from fresh blood. None of the aPTT tests results from any device produced results with aged blood that were clinically different from fresh blood. This study suggests that, in the tests evaluated, blood samples that have aged 10 or 18 min will produce clinically relevant aPTT and PT results, respectively. **Keywords:** point-of-care, prothrombin time, partial thromboplastin time. *JECT. 2002;34:178–181*

Prothrombin time was first reported in the literature by A. J. Quick in 1935 (1) and later clearly described as a quantitative tool in a text also authored by Quick in 1942 (2). The Partial Thromboplastin Time test was first described by Langdell et al. in 1953 (3) and was later modified by Margolis (4) in 1958 with the addition of kaolin as an activator. This "activated partial thromboplastin time" was improved upon in 1961 by Proctor and Rapaport (5), who described the test widely used today. The development of these tests was a significant contribution to the field of hematology and greatly improved clinicians' ability to diagnose coagulation defects. Despite major improvements in laboratory technology in the past four decades, these tests still have limitations that significantly inhibit their application in the perioperative cardiac surgical arena (6). Primarily, the standard laboratory PT and aPTT tests are performed on citrated plasma. This requirement necessitates a modicum of sample handling and additional equipment apart from the actual analysis device. Consequently, these tests are best performed in a centralized clinical laboratory, not in the operating room, which translates to significant delays for the open-heart surgical team attempting to manage a wet post-operative surgical field.

In response to these limitations, point-of-care tests have been developed that can provide PT and aPTT result on whole blood samples, thereby greatly reducing the sample

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handling and establishing the viability of bedside devices that can be operated by nonlaboratory staff. These tests can quickly provide test results that generally correlate well with laboratory standards (7–13). They reduce the processing time, as compared to central laboratory sample processing, and reduce errors associated with mislabeling, mishandling and/or losing the sample, incorrectly transcribing the results, or sample degradation over time. In addition, many POC tests provide test results from single microliter samples, thus decreasing the patients' blood loss related to phlebotomy (6).

Although minor, these tests also have limitations. Invariably, the product literature for these tests recommend that whole blood samples be tested without delay and citrated samples be tested within 15 minutes of procurement. These recommendations suggest that delays in processing of blood samples may invalidate the test results. Processing delays may be attributed to inadequate preparation of the technician or the equipment, machine failure following test initiation, or protocol-dependent research investigations. Therefore, to evaluate the effect of the age of the blood sample on the results of the PT and aPTT tests, we used five different POC devices to process fresh and aged blood.

MATERIALS AND METHODS

Following approval from the Institutional Review Board for the Protection of Human Subjects, informed consent was obtained from 11 adult patients undergoing elective cardiopulmonary bypass (CPB). Before sample processing, machine performance was tested against electronic quality controls. All disposable materials were stored according to manufacturers' recommendations. Before anesthetizing each patient, a radial artery catheter was placed for hemodynamic monitoring and blood sample collection. From each patient, two to four 40-mL blood samples were procured in a plastic syringe and analyzed for PT and aPTT results. Eleven different tests were investigated using five different POC devices: Gem PCL (Instrumentation Laboratory, Lexington. MA); Hemochron 801, Hemochron Jr. Signature; Hemochron Response (International Technidyne Corp., Edison, NJ); and RapidPoint Coag (Chiron Corp, Norwood, MA). Table 1 provides details regarding the devices, sample volume used, and tests investigated with each device.

Analysis of each blood sample occurred at three separate times for all devices and tests. Describing the age of the blood sample relative to the duration of time it had been in the syringe, samples were tested at less than 60 s (fresh blood), 10 min, and 18 min of age. For the test requiring citrated whole blood, a fraction of each sample was anticoagulated (3.8% citrate) at each age. The syringe containing the blood sample was maintained free of air bubbles and was left undisturbed between tests, but was gently agitated before initiating the 10 and 18 min tests to ensure the uniformity of the sample.

Repeated analysis of variance (ANOVA) measures with Student–Newman-Keuls post hoc test was used for comparison of test results for fresh blood to aged samples (10 and 18 min). Test values were recorded as international normalized ratio (INR) and seconds for PT and aPTT, respectively. Results are reported as the mean ± standard error (SE).

RESULTS

Tests that were initiated but produced error codes were necessarily removed from the data pool. Consequently, the number of samples, *N*, analyzed varied for different tests. The results of the PT tests are presented in Table 2. None of the six PT tests evaluated produced results from blood that had aged 10 and 18 min that differed clinically from the results produced from fresh blood. Two devices (Hemochron 801 and Hemochron Response) did report results that were statistically different ($p<.05$) at both 10 and 18 min when the INR was considered. However, the difference was small (<0.1 INR units) and had no clinical significance.

The results of the aPTT tests are presented in Table 3.

Table 1. Devices and tests investigated.

Device	Test Type	Test Name	Sample Volume (mL)	Sample Type
Gem PCL	PT	Pt test cartridge	7.5 μ	Fresh whole blood
	aPTT	aPTT test cartridge	10 μ	Fresh whole blood
Hemochron 801	PT	A 201 tube	2 mL	Fresh whole blood
	aPTT	A 103 tube	2 mL	Fresh whole blood
Hemochron Jr. Signature	PT	PT test cartridge	Microsample	Fresh whole blood
	aPTT	aPTT test cartridge	Microsample	Fresh whole blood
Hemochron Response	PT	A 201 tube	2 mL	Fresh whole blood
	aPTT	A 103 tube	2 mL	Fresh whole blood
RapidPoint Coag	PT	PT-NC card	35 μ	Fresh whole blood
	PT	PT-one card	35 μ	Citrated whole blood
	aPTT	aPTT card	35 μ	Citrated whole blood

This table provides information regarding the devices and the tests investigated in this study.

Table 2. PT test results (INR).

Device	Test Name	*N*	t = <60 s	t = 10 min	t = 18 min
Gem PCL	PT test cartridge	25	1.2 ± 0.04	1.1 ± 0.02	1.0 ± 0.03
Hemochron 801	A 201 tube	20	1.3 ± 0.09	1.2 ± 0.07 ¥	1.3 ± 0.09 ¥
Hemochron Signature	PT test cartridge	27	1.3 ± 0.14	1.3 ± 0.11	1.2 ± 0.11
Hemochron Response	A 201 tube	28	1.0 ± 0.01	1.0 ± 0.02 ¥	1.1 ± 0.02 ¥
RapidPoint Coag	PT-NC card	28	1.1 ± 0.02	1.1 ± 0.03	1.0 ± 0.02
	PT-One card	20	1.4 ± 0.05	1.5 ± 0.06	1.6 ± 0.09

Results for six different POC PT tests comparing test results obtained with fresh blood (<60 s following collection) to the same blood sample aged for 10 and 18 min from sample collection. Test values were recorded as the Mean ± SE. (¥ = $p < .05$) of the International Normalized Ratio (INR).

Of the five test evaluated for aPTT results in seconds, four tests did not report any statistical difference between fresh blood and blood that had aged 10 minutes. One test (RapidPoint) did not report any difference between fresh blood and blood that had aged 18 minutes. One test (Hemochron 801) reported statistically different ($p<.05$) results with blood that had aged 10 minutes. Four tests (Gem PCL, Hemochron 801, Hemochron Jr. Signature, Hemochron Response) reported statistically different results with blood that had aged 18 minutes.

DISCUSSION

Numerous researchers have studied a variety of sample-related variables and how these variables affect the results of the standard laboratory PT and aPTT tests. In an effort to reduce the patient discomfort caused by multiple veinpunture procedures necessary for daily PT and aPTT testing, no fewer than eight groups have investigated the influence of the site of sample procurement on the test results. Lindley et al. (14) determined that a peripheral IV located on the patient's forearm and maintained with a constant flow of unheparinized solution can be used for sample collection without introducing error into the results of the tests. Similarly, Arrants et al. (15) and Powers et al. (16) have established that peripheral IV lines that are flushed with saline and capped off can be used for sample procurement providing an adequate volume of waste is drawn through the line before sample collection.

Although Pinto (17) reported that PT and aPTT results from blood drawn from central venous lines were significantly prolonged, Mills et al. (18) reported that blood collected from the jugular venous catheter of the dogs in their study produced results equivalent to the results from blood collected via veinpuncture.

Haynes et al. (19) demonstrated that because of the heparinized flush solutions that continuously run through arterial lines, the results of PT and aPTT test collected from these lines may be misleading and that separate veinpuncture should be performed. However, Templin et al. (20) and Heap et al. (21) have since reported that by discarding a volume of blood greater than the dead space of the arterial catheter before collecting the sample, results were equivalent to veinpuncture.

Regarding the topic of sample volume, Peterson et al. (22) reported that underfilling, but not overfilling of the citrate tube may cause profound effects on the results of PT and aPTT. These data were supported by Adcock et al. (23) who went on to demonstrate that the newer collection tubes containing 3.2% citrate were far less affected by underfilling than the 3.8% tubes investigated by Peterson's group. In fact, the 3.2% tubes provided acceptable results when filled to only 60 and 70% for PT and aPTT tests, respectively.

In an effort to minimize the amount of blood wasted during sample procurement, Yawn et al. (24) and Brigden et al. (25) determined that, contrary to the accepted protocol of the day, accurate PT and aPTT test results could be obtained from the first tube collected via veinpuncture, and the need to discard the first tube and test the second tube collected was unnecessary.

Regarding the topic addressed in this study, age of sample, Neofotistos et al. (26) reported that citrated blood

Table 3. aPTT test results (seconds).

Device	Test Name	*N*	t = <60 s	t = 10 min	t = 18 min
Gem PCL	aPTT Test cartridge	19	44.3 ± 1.5	42.9 ± 2.2	35.7 ± 2.4 ¥
Hemochron 801	A 103 tube	29	135.7 ± 5.1	128.7 ± 5.0 ¥	123.2 ± 5.1 ¥
Hemochron Jr. Signature	aPTT Test cartridge	23	37.9 ± 1.1	37.1 ± 1.4	31.6 ± 1.3 ¥
Hemochron Response	A 103 tube	26	123.4 ± 2.1	122.6 ± 3.0	109.7 ± 2.8 ¥
RapidPoint Coag	aPTT card	25	39.7 ± 1.9	44.7 ± 3.4	39.8 ± 2.7

Results for five different POC aPTT tests comparing test results obtained with fresh blood (<60 s following collection) to the same blood sample which has aged 10 and 18 min from sample collection. Test values were recorded as the Mean ± SE. (¥ = $p < .05$) of test results in s.

samples that have been spun down to separate the plasma from the cells and maintained at room temperature will produce acceptable PT and aPTT results for at least 8 hours after collection. Brigden et al. (25) established that citrated whole blood samples could be maintained unspun and at room temperature for at least 24 hours and still produce acceptable PT and aPTT results. Iazbik et al. (27) demonstrated that citrated plasma preserved at –30°C for at least 30 days also produce clinically relevant results.

Unfortunately there are very few, if any, papers in the literature that report on how similar variables affect POC PT and aPTT tests. We can assume from the work of other researchers that, provided the technician takes appropriate precautions to draw an adequate waste sample, POC tests may be used to analyze blood drawn from the peripheral, central, or arterial IV lines of the cardiac patient perioperatively, following complete reversal of all circulating heparin with protamine.

This paper demonstrates for the first time the degree of stability of noncitrated whole blood as determined by several POC PT and aPTT tests. This information will aid the clinician in determining if unexpected delays in initiating these tests may confound the results of the test. Based on these data, when a time delay is introduced between sample collection and the initiation of the POC test, which may be the result of a patient's acute clinical condition, device malfunction, operator error, or another cause, the clinician may expect to receive clinically relevant results provided the sample has not aged more than 10 or 18 minutes for the aPTT and PT tests investigated, respectively.

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