Comparative Evaluation of the Measurement of Coagulation and Fibrinolitic System between the Fully Automated Blood Coagulation Analyzer CS-5100 and the STA-R Evolution Coagulation Analyzer

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Information about the hemostasis system is clinically important because it is useful in the diagnosis of bleeding and thrombotic diseases, and in monitoring the effectiveness of therapy. The new fully automated coagulation analyzer CS-5100 (Sysmex Corporation) has high measurement throughput and an effective reagent cooling system, which enable short turnaround times. We compared the performance of the CS-5100 and the STA-R Evolution analyzers and found that the CS-5100 had better measurement performance and onboard stability of reagents than the STA-R Evolution. We also found that the CS-5100 was very useful for making hemostasis in emergency cases.

Key Words CS-5100, Turn Around Time (TAT), Usability

INTRODUCTION

Fully automated analysis systems wherein the clotting assays, chromogenic assays and immunologic assays are simultaneously incorporated in one analyzer are widely used for hemostasis testing. Advances in automation and multi-functionality of the analyzers have resulted in great improvements in accuracy, speed of measurement, and user-friendliness, which are major contribution to the clinical field.

We have been using the fully automated coagulation analyzer STA-R Evolution (Roche Diagnostics; hereinafter STA-R Evo) for hemostasis testing at our hospital. However, we have been facing problems like delayed reporting of results in busy time slots, such as at the time of morning blood sampling in the wards and when large number of outpatient blood samples are processed, and instability of the onboard reagents due to the effect of temperature of the reagent compartment in the analyzer.

The fully automated blood coagulation analyzer CS-5100 (Sysmex Corporation; hereinafter CS-5100) examined in the present study has a higher throughput compared with the CS-2x00i series models, and the capability of simultaneous analysis even with combinations of samples in stoppered blood collection tubes, samples in unstoppered blood collection tubes and small volume samples. Furthermore, better cooling of the onboard reagents is expected to improve their stability, enabling rapid and efficient testing. We examined the basic performance of the CS-5100 and compared it with the STA-R Evo which is currently used in our hospital laboratory, and report the results here.

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TEST SPECIMENS AND METHODS

1. Specimens

198 plasma samples (collected 3.13% sodium citrate, cooled and centrifuged at 3,000 rpm for 10 min) from patients who had been sent to our hospital for routine hemostasis testing were used in the study. Written consent for use of the study samples was obtained from each patient.

2. Analyzers

A CS-5100 and STA-R Evo were used for making measurements. The specifications of these automated analyzers are given in ***Table 1***.

3. Reagents

The parameters analyzed and reagents used are listed in ***Table 2***. Prothrombin time (PT), activated partial thromboplastin time (APTT), fibrinogen (Fbg) and thrombotest (TTO) and Normotest (NT) values were determined using clot detection; antithrombin III (AT III) and plasminogen (PLG) were determined by the chromogenic detection; and D-dimer (DD), firbrinogen/fibrin degradation products (FDP), and fibrin monomer (FM) were determined by immunologic detection.

Table 1 *The Specification of the two automated analyzers*

		CS-5100	STA-R Evo
Measurement Principle	Clotting Assay	Transmitted light detection	Electromechanical viscosity
	Chromogenic Assay	Colorimetry	Colorimetry
	Immunologic Assay	Turbidimetry	Turbidimetry
	Aggregation Assay	Transmitted light detection	-
Throughput		Maximum 400 tests/h (PT or simultaneous PT & APTT)	Maximum 300 tests/h (PT alone)
Reagent cooling		10°C±2°C 24 h cooling function	15-19°C
Characteristic features		• Multi-wavelength detection (20 channels) • Samples can be added to the STAT table any time • Responses to queries by the clinical team possible, as the estimated end time of measurement is displayed • Cap piercing function and small volume sample analysis available (combinations of stoppered and unstoppered sample tubes and sample cups can be used) • Can be connected to Laboratory Automation System (LAS)	• Not affected by hemolysis or lipemia, as a mechanical method is used • Can analyze citrated whole blood samples (complex factors) • Calibration curve can be imported by reading the reagent barcode (for PT, Fbg, NT, and DD) • Equipped with cap piercing function

4. Methods

1) *Within-run Reproducibility*
Ten consecutive measurements were made with each of Coagutrol IX and IIX (COAG IX and IIX; Sysmex), Coagu QAP control IX and IIX (QAP IX and IIX; Sysmex), FDP control (Sekisui Medical) and LIA FM control (Roche Diagnostics).
2) *Between-run Reproducibility*
Measurements were made with each of Coagutrol IX and IIX, Coagu QAP control IX and IIX, FDP control, and LIA FM control over a period of 6 days.
3) *Reagent Onboard Stability*
Each of the reagents used was kept on board in the unstoppered state on the racks of CS-5100 and STA-R Evo for 12h from 7 AM to 7 PM and measurements were made on Coagutrol IX and IIX and Coagu QAP control IX and IIX daily for 7 days.
4) *Method Comparison*
Correlations between the results of measurements made with CS-5100 and STA-R Evo on the 198 patient plasma samples were examined.
5) *Effects of lipemia and hemolysis*
The effects of lipemia and hemolysis on measurements of PT, APTT, Fbg, TTO and NT were examined with 10 lipemic patient plasma samples and 10 hemolyzed patient plasma samples.
6) *Throughput*
Five measurement patterns with different testing orders were set, 10 samples of pooled patient plasma were measured for each pattern, and the time taken for the analysis determined.

Table 2 *Reagents for measurement*

Analyzer	CS		STA	
Analyzed parameter	Reagent	Manufacturer	Reagent	Manufacturer
PT	Thromborel® S	Siemens Healthcare Diagnostics	Thromborel® S	Siemens Healthcare Diagnostics
APTT	Actin®	Siemens Healthcare Diagnostics	Actin®	Siemens Healthcare Diagnostics
Fbg	Thrombocheck Fib (L)	Sysmex	Thrombocheck Fib (L)	Sysmex
TTO	Complex factor-T Kokusai	Sysmex	Complex factor-T Kokusai	Sysmex
NT	Complex factor-H Kokusai	Sysmex	Complex factor-H Kokusai	Sysmex
AT III	L-System AT III	Sysmex	Test team S AT III	Sekisui Medical
Plg	L-System PLG	Sysmex	Test team S PLG	Sekisui Medical
D-Dimer	Nanopia D-dimer	Sekisui Medical	Nanopia D-dimer	Sekisui Medical
FDP	Nanopia P-FDP	Sekisui Medical	Nanopia P-FDP	Sekisui Medical
FM	Auto LIA-FM	Sysmex	Auto LIA-FM	Roche Diagnostics

RESULTS

1. Within-run Reproducibility

The coefficient of variation (CV %) of different parameters was determined. It was 0.26 - 2.37% with CS-5100 and 0.40 - 2.38% with STA-R Evo for clotting parameters measured, 0.44 - 4.08% with CS-5100 and 1.33 - 3.89% with STA-R Evo for chromogenic parameters, and 1.06 - 3.83% with CS-5100 and 2.29 - 4.01% with STA-R Evo for immunologic parameters (***Table 3***).

Table 3 *Within-run reproducibility*

COAG IX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	88.6	105.4	27.5	27.7	250.9	263.3	119.5	111.8	121.2	99.8	96.3	105.8	98.4	100.3
SD	0.66	2.50	0.11	0.18	5.27	4.16	0.58	1.32	0.92	1.14	0.59	1.87	0.90	1.34
CV	0.74	2.38	0.38	0.63	2.10	1.58	0.48	1.18	0.76	1.14	0.61	1.77	0.91	1.33

COAG IIX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	42.8	47.2	71.3	69.4	108.5	114.7	49.9	50.2	47.1	44.5	32.1	41.5	36.1	38.3
SD	0.38	0.63	0.34	0.28	1.55	1.34	0.19	0.63	0.33	0.53	1.31	1.35	0.49	1.06
CV	0.88	1.34	0.48	0.40	1.43	1.17	0.37	1.26	0.70	1.18	4.08	3.26	1.36	2.77

QAP IX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	81.2	80.6	25.9	25.7	233.1	243.1	108.3	101.7	88.4	86.0	94.7	103.6	99.9	102.2
SD	0.58	1.26	0.07	0.17	5.25	3.84	0.74	0.82	0.41	0.82	0.41	1.84	0.57	2.04
CV	0.71	1.57	0.26	0.66	2.25	1.58	0.69	0.81	0.47	0.95	0.44	1.77	0.57	2.00

QAP IIX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	41.3	38.1	57.9	55.1	104.3	108.3	35.2	38.9	31.3	37.3	38.2	40.6	41.0	41.2
SD	0.18	0.32	0.32	0.36	2.47	2.00	0.10	0.32	0.15	0.48	0.54	1.58	0.40	0.63
CV	0.45	0.83	0.56	0.65	2.37	1.85	0.29	0.81	0.50	1.30	1.42	3.89	0.98	1.54

FDP control and LIA FM control (n = 10)

	DD (µg/mL)				FDP (µg/mL)				FM (µg/mL)			
	LOW		HIGH		LOW		HIGH		LOW		HIGH	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	3.0	2.5	8.8	9.1	10.7	9.5	33.3	31.4	17.1	16.8	92.6	88.8
SD	0.12	0.09	0.13	0.26	0.27	0.38	0.40	1.03	0.18	0.42	1.03	2.03
CV	3.83	3.72	1.52	2.89	2.49	4.01	1.20	3.28	1.06	2.53	1.12	2.29

2. Between-run Reproducibility

The CV % of clotting parameters was 0.80 - 4.29% with CS-5100 and 1.17 - 4.31% with STA-R Evo. It was 0.49 - 3.07% with CS-5100 and 1.02 - 4.24% with STA-R Evo for chromogenic parameters, and 1.19 - 4.68% with CS-5100 and 1.69 - 4.52% with STA-R Evo for immunologic parameters (***Table 4***).

Table 4 *Between-run reproducibility*

COAG IX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	98.2	100.2	28.2	28.3	251.0	263.5	117.6	104.0	104.1	100.0	98.2	105.0	98.1	99.0
SD	1.28	4.31	0.38	0.37	6.32	3.83	4.84	3.41	3.90	1.90	0.48	3.22	1.34	1.55
CV	1.30	4.30	1.34	1.29	2.52	1.46	4.12	3.27	3.74	1.90	0.49	3.07	1.36	1.56

COAG IIX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	46.4	44.3	74.7	70.7	103.0	113.8	49.8	47.0	45.8	45.0	33.4	38.7	35.0	37.0
SD	1.09	1.63	1.68	1.18	1.55	2.32	1.76	1.78	0.37	0.89	0.52	1.21	1.07	1.10
CV	2.36	3.68	2.25	1.68	1.50	2.04	3.53	3.80	0.80	1.99	1.56	3.13	3.07	2.96

QAP IX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	80.1	83.0	26.1	26.3	227.9	248.2	116.9	117.6	90.8	89.5	93.8	99.0	100.1	97.9
SD	1.08	3.58	0.25	0.27	6.84	6.08	5.01	4.84	2.13	1.05	1.23	2.28	1.65	1.00
CV	1.35	4.31	0.97	1.04	3.00	2.45	4.29	4.12	2.35	1.17	1.31	2.30	1.65	1.02

QAP IIX (n = 10)

	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)		AT III (%)		PLG (%)	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	41.4	37.8	59.7	55.0	103.2	106.7	37.1	37.7	36.8	39.0	36.4	41.5	40.0	40.8
SD	1.17	1.47	0.86	0.76	4.12	1.75	1.47	1.37	0.89	1.55	0.86	1.76	0.59	1.72
CV	2.83	3.89	1.45	1.37	4.00	1.64	3.96	3.63	2.43	3.97	2.36	4.24	1.46	4.22

FDP control and LIA FM control (n = 10)

	DD (μg/mL)				FDP (μg/mL)				FM (μg/mL)			
	LOW		HIGH		LOW		HIGH		LOW		HIGH	
	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
Mean	3.5	3.2	9.3	9.7	12.0	9.8	35.4	34.0	16.0	15.2	90.7	91.0
SD	0.12	0.15	0.44	0.34	0.48	0.33	1.16	1.11	0.26	0.64	1.08	1.54
CV	3.43	4.52	4.68	3.51	3.99	3.37	3.27	3.25	1.62	4.21	1.19	1.69

3. Onboard Reagent Stability

Onboard stability of reagents on CS-5100 and STA-R Evo is shown in ***Fig. 1***. For CS-5100, only TTO of Coagutrol showed a decrease in the measured value from day 4, and all other parameters gave stable results. On the other hand, with STA-R Evo, the measured values of TTO, NT, and AT III decreased from day 2 and of PT from day 4.

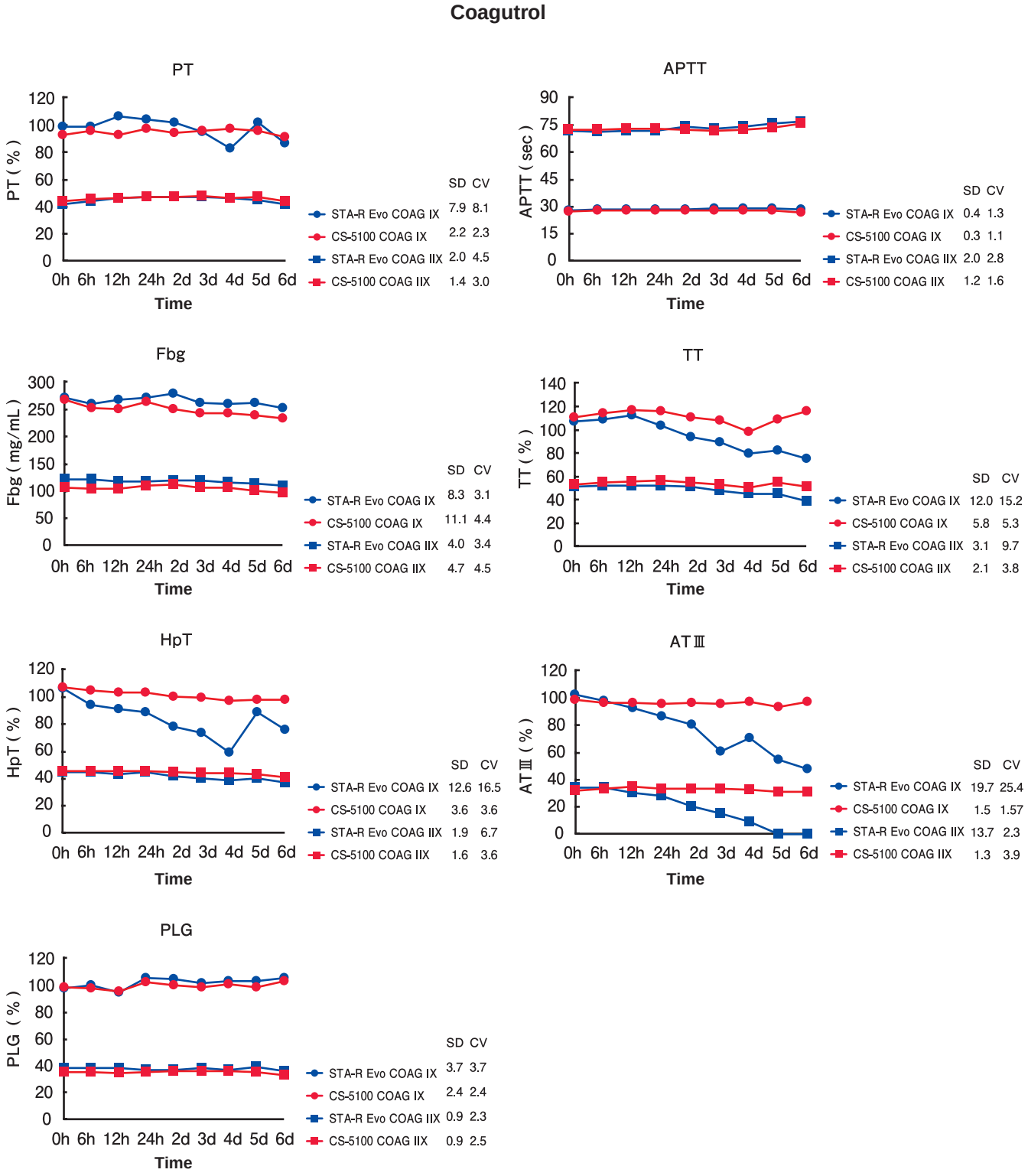
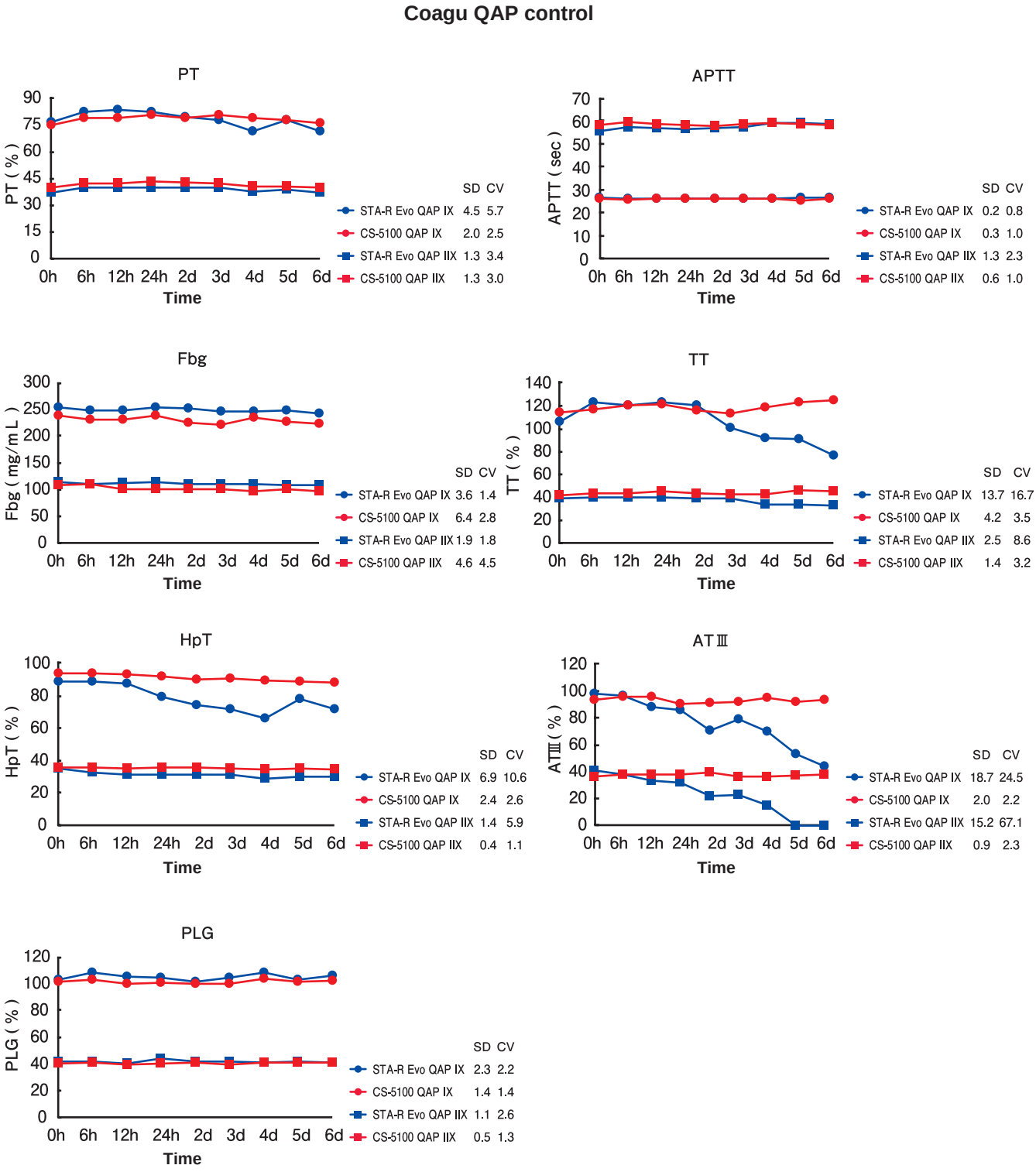


Fig. 1 *Stability of reagents on board the automated analyzer*



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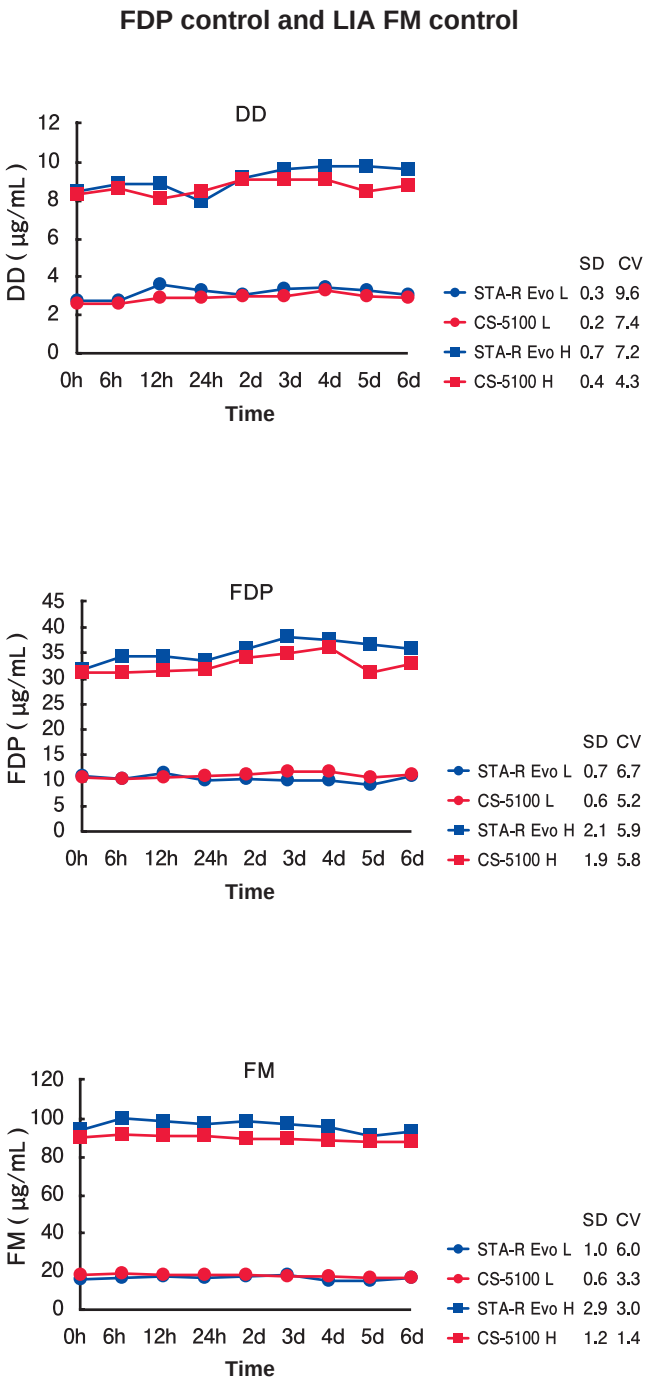

Fig. 1 *Stability of reagents on the board with automated analyzer*

4. Method Comparison

Fig. 2 shows the correlation between the analyzed values obtained using CS-5100 and STA-R Evo. The correlation was good, the correlation coefficient® being 0.941 - 0.990. However, in general, the measured values of APTT and FM were higher with CS-5100 than with STA-R Evo.

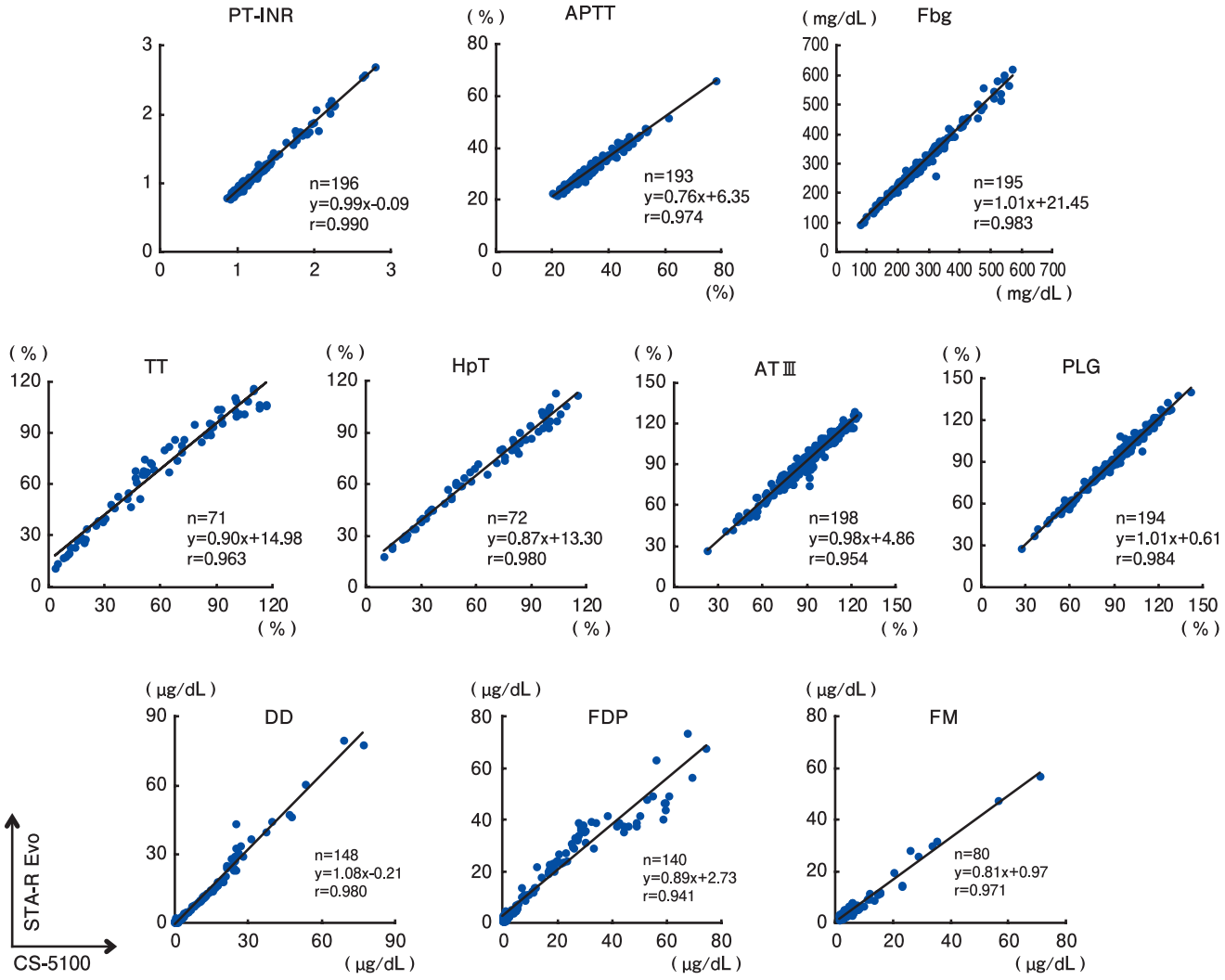

Fig. 2 *Correlation between CS-5100 and STA-R Evo*

5. Effect of Lipemia and Hemolysis

With both the CS-5100 and STA-R Evo, lipemia did not have a clear-cut effect at different concentrations of triglycerides (TG) (***Table 5***). Similarly, hemolysis of the samples also did not affect the measurement results (results not shown).

6. Throughput

Differences in the time needed for analysis between the CS-5100 and STA-R Evo for different order patterns are shown in ***Table 3***. The difference was 13 minutes and 53 seconds with Pattern (1) (9 parameters comprising PT, APTT, Fbg, TTO, NT, AT III, PLG, DD, and FDP), and 15 minutes and 15 seconds with Pattern (2) (7 parameters comprising PT, APTT, Fbg, TTO, NT, AT III, and PLG). Thus there was a difference of more than 10 minutes when a large number of parameters was ordered. Besides this, the CS-5100 completed the analysis faster than the STA-R Evo for all the patterns tested.

	Parameters analyzed (10 samples for each pattern)	Turnaround time (TAT)		Difference in TAT
		STA-R Evo	CS-5100	
Pattern (1)	PT, APTT, Fbg, TT, HPT, AT3, PLG, DD, FDP	48 min 32 sec	34 min 39 sec	13 min 53 sec
Pattern (2)	PT, APTT, Fbg, TT, HPT, AT3, PLG	37 min 54 sec	22 min 39 sec	15 min 15 sec
Pattern (3)	DD, FDP	18 min 53 sec	17 min 6 sec	1 min 47 sec
Pattern (4)	PT, APTT	11 min 8 sec	10 min 57 sec	11 sec
Pattern (5)	PT, APTT, Fbg, DD, FDP	25 min 50 sec	23 min 51 sec	1 min 59 sec

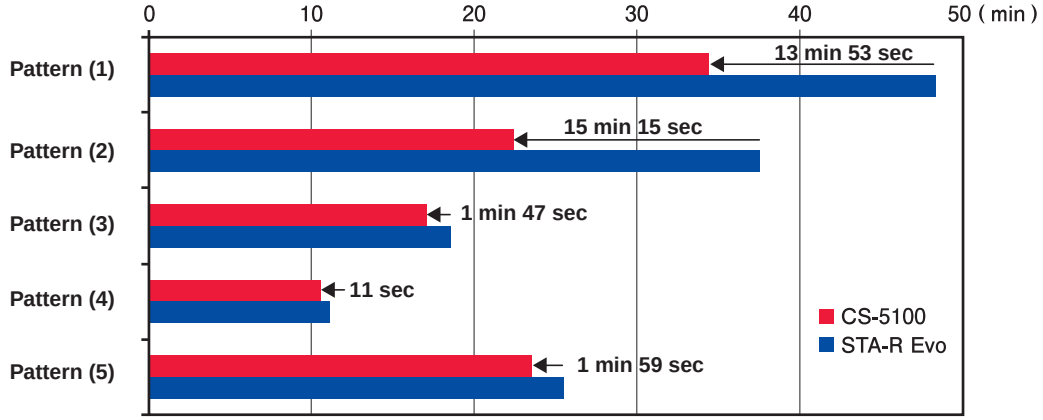

Fig. 3 *Test Patterns for Throughput Study*

Table 5 *Effect of the chylemia on the values mesured with CS-5100 and STA-R Evo*

Sample No.	TG (mg/dL)	PT (%)		APTT (sec)		Fbg (mg/dL)		TT (%)		HpT (%)	
		CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo	CS-5100	STA-R Evo
1	598	36	36	35.6	32.6	288	290	15	23	34	39
2	434	90	104	35.6	31	323	344	107	103	120	103
3	377	62	64	48.4	40.9	227	233	48	53	78	69
4	110	76	79	34.3	33.9	432	473	75	65	101	93
5	318	77	84	29	28.1	238	250	55	64	78	75
6	343	83	86	40.1	34.1	246	259	71	66	100	93
7	158	80	82	33.1	31.2	299	310	71	73	90	88
8	433	120	141	18.5	19.4	295	306	115	134	121	125
9	1217	93	102	46.3	39.2	288	320	81	95	153	150
10	180	125	145	25.5	25.3	294	306	157	150	119	119

DISCUSSION

Hemostasis testing is positioned as emergency testing, as such tests are very useful in understanding the causes and pathophysiology of bleeding and thrombotic diseases and in monitoring their treatment. The hemostasis system comprises a network in which various factors are interconnected. Therefore the results of testing need to be interpreted comprehensively. The major issue is how rapidly analysis of the parameters can be completed and comprehensive results reported[1]. At our hospital we have been using the STA-R Evo for hemostasis testing, and facing certain problems with the system, such as delays in reporting of results in busy time slots that have large numbers of samples requiring multi-parameter analysis and instability of reagents due to the effect of temperature of the reagent compartment of the analyzer. Against this background we verified the basic performance of the CS-5100, which has features such as high throughput, and an improved reagent cooling function and compared it with the STA-R Evo.

The within-run reproducibility, in terms of CV, was 0.26 - 4.08% for the CS-5100 and 0.40 - 4.01% for the STA-R Evo, and their between-run reproducibility was respectively 0.80 - 4.68% and 1.02 - 4.52%. Thus the CS-5100 was found to have about the same level of accuracy current coagulation analyzer[2-4]. We also examined the effect of lipemia and hemolysis in samples measured by the CS-5100, as there have been reports that no interference of this type occurred with the fully automated coagulation analyzer CS-2100i (Sysmex), which uses the same detection principle[5,6]. We found no major interference in measurements made by the CS-5100 compared with the STA-R Evo which uses a mechanical method of measurement. The correlation between the CS-5100 and the STA-R Evo was good, the correlation coefficient® being ≥ 0.941. However, the measured values of APTT and FM were generally higher with the CS-5100 than with STA-R Evo. One reason for the difference in the APTT results could be the difference between the analyzers in the end point detection method. In fact, the present authors and their colleagues had reported similar results in method comparison study[4] of the analyzers STA-R Evo, CS-2100i, the fully automated blood coagulation analyzer CA-7000 (Sysmex), and Coapresta 2000 (Sekisui Medical).

Investigations by other researchers have also revealed differences between analyzer models[5]. The reason for the higher values of FM measured by the CS-5100 could be differences in incubation time and conditions of detection between the analyzers. It is hoped that such differences between the two analyzers could be eliminated in the future through the adoption of suitable corrective measures.

With regard to reagent stability onboard, the TTO values measured by the CS-5100 started to show decrease from day 4. With the STA-R Evo, TTO, NT and AT III started to show a decrease from day 2, and PT from day 4. Although the inherent instability of the reagent could partly be the cause for this in the case of PT, TTO, and NT, all the analyzed parameters gave more stable results with the CS-5100. There has been a report that the AT III, DD and FDP measurements by the conventional the CS-2100i remained stable for 2 days[6]. Compared with that, we can say that the stability provided by the onboard reagent compartment temperature environment of the CS-5100 is excellent, and has sufficient promise to assure data quality and a reduction in the number of calibrations required, through the prevention of reagent degradation, during use in normal working hours at night and on holidays.

As for the throughput, analysis was completed faster with the CS-5100 than with the STA-R Evo for all the order patterns tested. There was a difference of about 15 minutes in multi-parameter analysis. This time we had tested the throughput for each pattern with 10 samples. CS-5100 has a larger detection area than the earlier CS series analyzers and uses four different measurement principles and multi-wavelength detection. This makes the CS-5100 have much less variability depending on the order pattern. There would obviously be much greater difference in the measurement throughput between the two analyzers when a larger number of samples are used. We can expect that the use of the CS-5100 would resolve the problem of delay in obtaining test reports during working hours when there is a rush of samples.

CONCLUSION

Apart from its satisfactory basic performance, the CS-5100 analyzer had a good temperature environment in the reagent compartment and excellent turnaround time, a valuable feature at the peak time of samples. CS-5100 thus provided better stability of onboard reagents during working hours and nights and holidays, and it could also speed up the comprehensive reporting of results to the clinical personnel. Therefore, it was assessed to be an analyzer with high clinical usefulness.

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