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# Comparison of Three Blood Collection Tubes for 35 Biochemical Analytes: The Becton Dickinson Barricor Tube, Serum Separating Tube, and Plasma Separating Tube

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The Barricor tube (Becton Dickinson [BD], Sunnyvale, CA, USA) was recently developed to mechanically separate plasma by increasing the centrifugation rate. We compared the Barricor tube with existing serum- and plasma-based tubes based on 35 biochemical analytes and preanalytical turnaround time (TAT). Blood samples were collected from 30 healthy volunteers in a Barricor tube, serum separating tube (SST, Vacutainer SST II Tube 8.5 mL, #368972; BD), or plasma separating tube (PST, Vacutainer PST Tube 8.0 mL, #367964; BD) in random order. Next, 27 chemistry analytes, six immunochemistry analytes, and two cardiac markers were compared using Passing-Bablok regression and the Bland-Altman method. Preanalytical TAT was measured for each tube.

The Barricor tube exhibited bias exceeding the desirable limit for nine and four analytes compared with the SST and PST, respectively. The Barricor tube lactate dehydrogenase value showed a bias of -10.29% and -9.86% compared with that of the SST and PST, respectively. The preanalytical TAT of Barricor tube was 8.8 minutes, which was the shortest among the three tubes. The clinical performance of the Barricor tube was equivalent to that of the SST and PST for most analytes, with an apparent advantage in preanalytical TAT. When using the Barricor tube, the reference range needs to be changed for some analytes that exceed the desirable bias limit.

**Key Words:** Barricor tube, Bias, Turnaround time, Analytes, Serum separating tube, Plasma separating tube

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The BD Vacutainer Barricor plasma blood collection tube (Becton Dickinson [BD], Sunnyvale, CA, USA) is a recently introduced plasma separating tube (PST) that utilizes a mechanical separator which allows a higher centrifugation speed. We measured a total of 35 analytes using the Barricor tube (Vacutainer Barricor Tube 5.5 mL, #365057; BD), serum separating tube (SST; Vacutainer SST II Tube 8.5 mL, #368972; BD), and PST (Vacutainer PST Tube 8.0 mL, #367964; BD), for performance evaluation of Barricor tubes. Previous studies on Barricor tube were either performed only with PST [1-4] or without considering immunochemistry analytes when compared with SST [5]. Other than comparison studies on Barricor tube, centrifugation conditions and sample quality have also been studied [6]; furthermore, hemolytic index and processing under vacuum have also been explored [7]. This study provides comparison data for the Barricor tube, SST, and PST for 27 chemistry analytes and two cardiac markers and for the Barricor tube and SST for six immunochemistry analytes, thus covering more analytes than

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before with SST [4, 5]. The preanalytical turnaround time (TAT) of the Barricor tube, SST, and PST was compared, and the current status of the SST sample quality in our routine chemistry laboratory was also assessed. A total of 30 healthy volunteers were recruited prospectively. All volunteers were over 19 years of age and none were pregnant or breastfeeding; 22 mL of blood was collected by venipuncture from all volunteers, after obtaining informed consent. This study was conducted at Samsung Medical Center, Seoul, Korea, in March 2018. The study was reviewed and approved by the Samsung Medical Center Institutional Review Board (IRB no. 2018-01-101).

For TAT assessment, each time point was recorded manually at the start of blood collection, before centrifugation, before measuring the analytes with sample aliquots, and at the final reporting time of the analyzed results from selected equipment. The TAT preanalytical phase, calculated as the sum of the coagulation time, centrifugation time, time for visual inspection, aliquoting time, and time spent in sending samples to the equipment, was determined for each tube. Samples in SSTs were allowed to clot for 30 minutes immediately after blood collection and were then centrifuged at 25°C and 2,000  $\times g$  for 10 minutes, while those in Barricor tubes and PSTs were centrifuged immediately after blood collection at  $1,300 \times g$  for 10 minutes and  $3,500 \times g$ for 5 minutes, respectively, at 25°C. Although the optimal centrifugation condition for the Barricor tube provided by the manufacturer is  $4,000-5,000 \times g$  for 3 minutes, different conditions were used owing to the equipment currently installed in the clinical chemistry laboratory at the Samsung Medical Center.

A total of 27 routine chemistry analytes were measured with a Roche Cobas c702 chemistry analyzer (Roche Diagnostics, Basel, Switzerland), using aliquots from the Barricor tubes, SSTs, and PSTs (Table 1). Six immunochemistry analytes were measured with an ADVIA Centaur XP Immunoassay System (Siemens Healthineers, Erlangen, Germany), using aliquots from the Barricor tubes and SSTs (Table 2). Cardiac markers, cardiac troponin I (cTnI) and creatine kinase-myocardial band (CK-MB), were measured with an ADVIA Centaur XP Immunoassay System (Siemens), using aliquots from the Barricor tubes, SSTs, and PSTs. The performance of the Barricor tube was compared with that of the SST and PST using Passing-Bablok regression. The slope and intercept of the regression line were calculated with 95% confidence interval (CI). Percent biases at medical decision points were calculated for analytes with either a Passing-Bablok slope of one outside the 95% CI or an intercept of zero outside the 95% CI (Table 3). Statistically significant differences between the Barricor tube and SST were calculated using paired t-test, and P < 0.05 was considered statistically significant. Clinical significance was judged based on the desirable biological variation database or total allowable error according to the Clinical Laboratory Improvement Amendments criteria, whichever was lower [8, 9].

Nine analytes from the Barricor tube exceeded the desirable bias limit compared with those from the SST. Compared with that for the SST, the lactate dehydrogenase (LDH) value for the Barricor tube showed a percent bias of -10.29%, which exceeded the desirable bias limit. The biases for albumin (ALB), aspartate aminotransferase (AST), calcium (Ca), glucose, potassium, phosphorus, sodium, and total protein also exceeded the desirable bias limit, ranging from -6.75% to 4.78%.

When the Barricor tube was compared with the PST, all analytes, except AST, Ca, carbon dioxide (CO<sub>2</sub>), and LDH, were within the desirable bias limit, and values exceeding the desirable bias limit ranged from -1.16% to -9.86%. The Ca bias was higher than the desirable bias limit compared with both SST and PST, although the value for both was <1.5%. Of the immunochemistry analytes, the folate and free thyroxine (FT4) values for the Barricor tube exceeded the desirable bias limit compared with those for the SST (Table 2). Regression for cTnI was not calculated, as most of the measured data were identical for the Barricor tubes, SST, and PST at values of 0.006  $\mu$ g/L, which is the limit of detection of the test.

SST sample quality was also evaluated in our routine chemistry laboratory. Throughout the observational period, from January 11-16, 2019, 3,057 SSTs in total were visually inspected, after centrifugation, for quality review. Employees counted the number of samples containing artifacts such as gel globules or fibrin; of the SSTs, 229 (7.5%) required manual removal of either gel globules or fibrin after the centrifugation process. This remediation rate was consistent with the results of a global survey, which reported a remediation incidence of up to 6.0% for gel globule or fibrin [10]. Barricor tubes with less gel globules or fibrin could thus reduce manual labor and preanalytical TAT. Issues related to fibrin interference are known, and some clinical laboratories prefer plasma to serum [11]. The Barricor tubes presented the shortest preanalytical TAT, with an average preanalytical TAT of 8.8 minutes, owing to the short centrifugation step and higher centrifuge speed, whereas the SSTs exhibited the longest preanalytical TAT (average=43.6 minutes) owing to a coagulation step of 30 minutes. The average preanalytical TAT of the PSTs was 13.8 minutes.

Studies on Barricor tubes in terms of analyte stability, effect of centrifugation speed, performance comparison with SST or PST,

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	Barricor	SST	PST		Barric	or vs SST			Barı	icor vs PST		
Analyte	Mean (SD)	Mean (SD)	Mean (SD)	% Bias	P-B 95% CI slope	P-B 95% CI intercept	P(SST)	% Bias	P-B 95% CI slope	P-B 95% CI intercept	P (PST)	uesirable bias (%) <sup>‡</sup>
ALB (g/L)	46.0 (2.5)	46.9 (2.3)	46.4 (2.3)	-1.90	1.00 - 1.33	-1.60-0.10	< 0.05	-0.97	1.00-1.20	-0.96-0.0	< 0.05	1.43
ALP (µkat/L)	0.95 (0.22)	0.97 (0.22)	0.96 (0.22)	-2.34	0.96 - 1.00	-2.00-1.25	< 0.05	-0.33	0.94 - 1.00	-1.00 - 3.59	0.50	6.72
ALT (µkat/L)	0.35 (0.45)	0.36 (0.45)	0.35 (0.45)	-7.53	0.89-1.01	-1.44-0.67	< 0.05*	-1.83	0.88-1.02	-1.23 - 1.46	0.20*	11.48
AMY (µkat/L)	1.06 (0.42)	1.06 (0.42)	1.06 (0.43)	-0.53	0.99-1.03	-1.71-0.35	0.137	-0.35	0.97-1.00	-0.20-1.40	0.073	7.40
AST (µkat/L)	0.33 (0.25)	0.35 (0.25)	0.35 (0.25)	-6.75	0.95-1.33	-7.07-0.50	0.06*	-8.82	0.99-1.20	-5.430.84	< 0.05*	6.54
BUN (mmol/L)	3.85 (1.15)	3.86 (1.10)	3.87 (1.15)	-0.72	1.00-1.07	-0.74-0.0	0.54	-0.83	1.00 - 1.07	-0.76-0.00	0.18	5.60
Ca (mmol/L)	2.33 (0.05)	2.36 (0.05)	2.35 (0.05)	-1.46	0.82-1.25	-2.51 - 1.58	< 0.05	-1.16	1.00 - 1.50	-4.93-0.00	< 0.05	0.80
CO <sub>2</sub> (mmol/L)	26.31 (2.79)	26.62 (2.52)	27.12 (2.30)	-1.26	0.77-1.30	-8.27-6.74	0.55	-3.23	0.93-1.40	-10.99 - 1.92	0.074	1.60
CL (mmol/L)	101 (1.64)	101.17 (1.81)	101.01 (1.53)	-0.16	0.75-1.00	0.00-25.5	0.42	0	1.00 - 1.34	-34.71-0.00	0.68	0.50
CHOL (mmol/L)	4.38 (0.60)	4.48 (0.60)	4.42 (0.60)	-2.24	0.95-1.06	-14.87-5.00	< 0.05	-0.92	0.97-1.05	-9.61 - 4.13	< 0.05	4.10
CREAT (µmol/L)	64.55 (12.14)	64.55 (14.51)	66.32 (12.38)	0.23	0.86 - 1.10	-0.07-0.10	0.912	-2.26	0.87-1.08	-0.08-0.08	< 0.05	4.00
DBIL (µmol/L)	4.62 (1.86)	4.62 (1.86)	4.45 (2.79)	1.33	1.00 - 1.00	0.00-0.00	1.00*	4.89	1.00 - 1.00	0.00-0.00	0.52*	14.20
TBIL (µmol/L)	9.58 (4.71)	9.92 (4.71)	9.92 (4.86)	-1.90	1.00 - 1.00	0.00-0.00	0.10	-2.82	1.00 - 1.00	0.00-0.00	0.16	9.00
GGT (µkat/L)	0.35 (0.34)	0.36 (0.34)	0.36 (0.34)	-0.89	1.00-1.02	-0.62-0.00	0.58*	-2.32	0.90 - 1.01	-1.00 - 1.19	0.53*	11.10
Glucose (mmol/L)	5.35 (0.71)	5.09 (0.66)	5.32 (0.66)	4.78	0.94-1.17	-10.17-9.63	< 0.05	0.49	0.98-1.17	-14.67–2.45	0.31	2.30
HDL (mmol/L)	1.67 (0.44)	1.69 (0.44)	1.68 (0.44)	-0.97	0.98-1.00	-1.00-0.74	< 0.05	-0.44	1.00-1.02	-1.39-0.00	0.165	5.60
Fe (µmol/L)	16.31 (5.59)	16.84 (5.59)	16.59 (5.59)	-3.46	0.97-1.01	-4.150.59	< 0.05	-1.96	1.00 - 1.00	-2.001.00	< 0.05	8.80
LDL (mmol/L)	2.79 (0.48.9)	2.83 (0.49)	2.78 (0.49)	-1.36	0.96-1.11	-12.67-3.15	< 0.05	0.24	0.96-1.07	-7.04-5.00	0.596	5.46
LDH (µkat/L)	2.82 (0.71)	3.12 (0.73)	3.10 (0.72)	-10.29	0.90-1.35	-74.32-1.95	< 0.05	-9.86	0.92-1.28	-62.381.42	< 0.05	4.30
LIP (µkat/L)	0.56 (0.74)	0.57 (0.74)	0.56 (0.74)	-0.95	0.95-1.01	-0.45-0.97	0.136*	0.62	0.95-1.02	-0.49 - 1.17	0.309*	11.30
Mg (mmol/L)	0.83 (0.05)	0.84 (0.05)	0.84 (0.05)	-1.47	1.00 - 1.00	-0.05-0.00	< 0.05	-1.50	1.00 - 1.00	0.00-0.00	< 0.05	1.80
Potassium (mmol/L)	4.07 (0.23)	4.25 (0.24)	4.04 (0.24)	-4.41	0.67-1.25	-1.33 - 1.30	< 0.05	0.75	0.80-1.50	-1.90-0.77	0.43	1.80
Phosphorus (mmol/L)	1.09 (0.14)	1.15 (0.14)	1.08 (0.14)	-5.69	1.00 - 1.00	-0.200.20	< 0.05	0.31	1.00 - 1.00	0.00-0.00	0.33	3.38
Na (mmol/L)	140.07 (1.89)	140.37 (1.81)	139.93 (1.80)	-0.21	1.00 - 1.50	-71.25-0.00	0.184	0.00	1.00 - 1.00	0.00-0.00	0.46	0.20
TP (g/L)	74.00 (3.7)	72.60 (3.5)	74.90 (3.3)	1.94	0.83-1.33	-2.35-1.44	< 0.05	-1.20	0.90 - 1.50	-3.85—0.64	< 0.05	1.40
TG (mmol/L)	1.13 (0.48)	1.14 (0.50)	1.11 (0.49)	-1.12	0.93-1.00	-1.00-3.75	0.060	1.83	0.96 - 1.00	0.00 - 4.19	< 0.05	9.60
UA (mmol/L)	0.29 (0.08)	0.29 (0.08)	0.29 (0.08)	0.27	1.00 - 1.06	-0.24-0.10	0.28	0.23	1.00-1.02	-0.08-0.00	0.33	4.90
cTnl (µg/L)	0.007 (0.006)	0.007 (0.006)	0.007 (0.005)	0.27	N/A <sup>†</sup>	N/A <sup>†</sup>	1.00	$1.00^{*}$	N/A	N/A	$1.00^{*}$	16.32
CK-MB (µg/L)	0.99 (0.82)	0.97 (0.79)	0.98 (0.81)	2.00	0.95-1.14	-0.09-0.07	0.20	1.78	0.93-1.06	-0.06-0.08	0.57	14.88
Measured with Roche (Siemens Healthineers *Calculated with paired	Cobas c702 che , Erlangen, Germ 1 Wilcoxon signec	emistry analyzer any). Bold numt d-rank test. All ot	(Roche Diagnosti bers indicate perce ther values withou	cs, Basel, 5 ent bias exc it asterisks v	witzerland) ex eeding the de: vere calculated	cept for cTnl ar sirable bias limit d with paired t-te	nd CK-MB w or Passing-fest; *Regress	hich were 3ablok inter ion was no	measured wit cept of zero o t calculated; <sup>‡</sup>	h ADVIA Centaur utside the 95% C Desirable bias pro	XP Immuno I. Dvided in the	assay System desirable bio-
logical variation databa	se specifications	[8].			-			. V V V				
Abbreviations: P-B, PE urea nitrogen; Ca, calc	ssing-Bablok; U, ium; CO <sub>2</sub> , carbor	r dioxide; CL, ch	Ival; ALB, alburni. Ioride; CHOL, chc	n; ALI, alar desterol; CF	REAT, creatinin	e; DBIL, direct l	valine pnosp. bilirubin; TBi	L, total bilir	Y, amylase; Α ubin; GGT, γ-	glutamyltransfera	notransterase se; HDL, higt	density lipo-
protein; Fe, iron; LDL, MB, creatine kinase-M	low density lipop B; SST, serum se	rotein; LDH, lact. sparating tube; P.	ate dehydrogenas ST, plasma separa	se; LIP, lipas ating tube.	ie; Mg, magne	sium; Na, sodiu	ım; TP, total	protein; TG	, triglycerides;	UA, uric acid; c	Inl, cardiac tr	oponin I; CK-

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	Barricor	SST		Desirable bias			
Analyte	Mean (SD)	Mean (SD)	%Bias	Passing-Bablok 95% CI slope	Passing-Bablok 95% CI intercept	P(SST)	(%) <sup>‡</sup>
Vit. B <sub>12</sub> (pmol/L)	444.72 (186.90)	400.42 (161.28)	10.01	1.06-1.20	-45.99-18.83	< 0.05	17.70
Folate (nmol/L*)	31.77 (8.35)	20.58 (6.88)	45.13	0.98-1.17	3.26-4.86	< 0.05	19.20
Ferritin (µg/L)	96.10 (108.45)	106.3 (115.5)	-1.48	0.91-0.97	-0.57-1.69	$< 0.05^{+}$	5.20
TSH (mIU/L*)	1.54 (0.97)	1.54 (0.96)	-3.54	0.98-1.02	-0.02-0.03	0.88	7.80
FT4 (pmol/L*)	16.86 (3.33)	15.96 (3.68)	5.31	0.84-1.29	-0.27-0.26	< 0.05	3.30
FSH (IU/L*)	12.38 (21.07)	12.40 (22.09)	3.16	0.94-1.07	-0.21-0.47	$0.012^{\dagger}$	12.10

#### Table 2. Analytical data for Barricor tube and SST immunochemistry analyte values

Measured with ADVIA Centaur XP Immunoassay System (Siemens Healthineers, Erlangen, Germany). Bold numbers indicate Passing-Bablok slope of one outside the 95% CI.

\*Reagent was not validated for plasma; thus, the Barricor tube results are unreliable; <sup>†</sup>Calculated with paired Wilcoxon signed-rank test. All other values were calculated with paired t-test; <sup>†</sup>Desirable bias provided in the desirable biological variation database specifications [8].

Abbreviations: CI, confidence interval; Vit. B<sub>12</sub>, vitamin B<sub>12</sub>; TSH, thyroid stimulating hormone; FT4, free thyroxine; FSH, follicle-stimulating hormone; SST, serum separating tube.

Table 3. Percentage bias at r	medical decision poi	pints for selected analy	/tes in Barricor tube
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	Barricor tube						
Analyte	Medical decision point 1	% Bias (vs SST)	% Bias (vs PST)	Medical decision point 2	% Bias (vs SST)	% Bias (vs PST)	(%) <sup>†</sup>
ALB (g/L)	35	-2.86	-1.27	52	-1.92	-1.71	1.43
AST (µkat/L)	0.53	-2.52	-6.25	1.67	0.04	-2.00	6.54
Fe (µmol/L)	5.91	-9.10	-6.06	34.55	-1.55	-1.04	8.80
LDH (µkat/L)	2.25	-12.64	-6.55	3.57	-14.24	-6.40	5.46
Phosphorus (mmol/L)	0.81	-8.00	0.00	1.45	-4.44	0.0	3.38
Vit B12 (pmol/L)	155.72	6.44	N/A*	673.32	10.68	N/A*	17.70
Ferritin (µg/L)	10	-4.79	N/A*	291	-6.82	N/A*	5.20

Bold numbers indicate percent bias exceeding the desirable bias limit.

\*Comparison between Barricor tube and PST was not performed; <sup>†</sup>Desirable bias provided in the desirable biological variation database specifications [8]. Abbreviations: SST, serum separating tube; PST, plasma separating tube; ALB, albumin; AST, aspartate aminotransferase; LDH, lactate dehydrogenase; Fe, iron; Vit. B<sub>12</sub>, vitamin B<sub>12</sub>.

and effects on therapeutic drug monitoring have reported stable performance of Barricor tubes for most routine chemistry tests [1–7, 12]. While most analytes included in Barricor tube comparison studies have yielded clinically non-significant results, LDH has been commonly reported to have a bias compared with SST or PST [1, 3–5]. Our Barricor tube LDH values showed a bias of ~10% compared with those obtained using SST or PST. Although Arslan, *et al.* [5] reported a positive bias of 10–20% using Barricor tubes compared with SSTs and PSTs, our LDH results showed a negative bias. Previous studies have reported LDH values with a positive or negative bias <5% compared with those using PSTs [1, 3, 4]. In our study, AST using Barricor tubes showed a negative bias >5% compared with that using SST, and the value exceeded the desirable bias limit. Potassium and phosphorus using Barricor tubes showed a negative bias close to 5%, and the value

exceeded the desirable bias limit compared with those using SST.

AST, alanine aminotransferase, and LDH, along with many other analytes, are known to be affected by hemolytic interference [13], which is suspected as the cause of discrepant results. The higher centrifuge speed required for the Barricor tube might cause hemolytic interference. However, studies on the effects of centrifuge speed on the Barricor tube for measured analytes have not provided meaningful results [4, 5]. Several stability studies have suggested prolonged contact of serum/plasma and clot along with disrupted cell-membrane integrity of erythrocytes as factors contributing to the increased LDH values using serum or plasma [11, 14, 15]. LDH value is also known to increase with membrane leakage from necrotic cells [16]. Another stability study with Barricor tubes showed more stable LDH values compared with PSTs [1]. The Barricor tube mechanical sep-

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arators provide cell and plasma separation using a different mechanism from that of gel separators, and this difference could contribute to varying sample separation quality, resulting in different LDH values. In cases of hemolytic interference, the biases observed for AST and LDH should be either both positive or both negative, as both are intracellular components released during hemolysis [13]. While our study using Barricor tubes showed negative biases for both AST and LDH, Arslan, *et al.* [5] reported positive biases for both AST and LDH. The different experimental conditions used in the two studies might have led to a different hemolytic effect. However, further studies with varying centrifugation speed, temperature, and aliquot conditions are required to elucidate these discrepancies.

Potassium values are known to be elevated in serum, and our results are consistent with previous findings [17]. In addition, the potassium values using the Barricor tubes were comparable to those using PSTs, and this result is consistent with previous findings [1, 3–5]. Glucose value has been reported to be lower in serum, as the 30-minute coagulation step required for SST increases the chance of glycolysis during the process [18, 19].

Of the six immunochemistry analytes tested, folate and FT4 showed biases exceeding the desirable bias limit (45.13% and 5.31%, respectively). While the reagents used for the immunochemistry analytes vitamin B<sub>12</sub> (Vit B<sub>12</sub>) and ferritin were validated for both serum and plasma, the reagents for folate, thyroid stimulating hormone (TSH), FT4, and follicle-stimulating hormone (FSH) were only validated for serum samples. The measured bias of Vit. B<sub>12</sub> was 10.01%, which was lower than the desirable bias limit. A previous study reported that Vit. B<sub>12</sub> values using the Barricor tubes were clinically equivalent to those using SSTs and PSTs, with a bias <1% [5]. Fournier, *et al.* [1] reported a bias of 0.08% for Vit. B<sub>12</sub> in a comparative study between the Barricor tubes and PSTs; however, they did not perform a comparison with SSTs.

To date, the Barricor tubes and SSTs have not been compared with respect to the immunochemistry analytes folate, ferritin, TSH, FT4, and FSH. Of these six immunochemistry analytes, our data showed that Vit.  $B_{12}$ , ferritin, TSH, and FSH were within the desirable bias limit. Although the reagents for TSH and FSH were not validated for plasma, the Barricor tube-measured data were still within the desirable bias limit.

Of the selected analytes, the LDH bias exceeded the desirable bias limit at the medical decision points, compared with those of both SST and PST, with biases ranging from -6.40% to -14.24% (Table 3). In addition, the biases for ALB, Fe, phosphorus, and ferritin were higher than the desirable bias limit at one or more

of the medical decision points.

As this study used peripheral blood samples from healthy volunteers, we could not conduct a performance evaluation covering the entire analytical measurement range. In addition, most of the cTnl values showed limit values <0.006  $\mu$ g/L. These values were not normally distributed, which was a limiting factor of this study.

In conclusion, the clinical performance of Barricor tubes was equivalent to that of SSTs and PSTs for most of the analytes. AST, Ca, and LDH showed relatively high biases, exceeding the desirable bias limit. Vit. B<sub>12</sub> also showed a relatively high bias using the Barricor tube compared with SST; however, the value was still within the acceptable range. We showed clinically equivalent performance for the Barricor tubes at a centrifugation speed of  $3,500 \times g$ , which was slower than the optimal speed recommended by the manufacturer. When using the Barricor tube, the reference range might need to be changed for analytes exceeding the desirable bias limit. Moreover, additional studies on the Barricor tube are needed to elucidate the discrepant biases between our study and previous studies.

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#### **AUTHOR CONTRIBUTIONS**

HDP designed the study concept and reviewed data analysis and manuscript. JO reviewed the data. SS performed statistical data analysis and wrote the manuscript. All the authors have accepted responsibility for the entire content of this submitted manuscript and approved submission.

### **CONFLICTS OF INTEREST**

None declared.

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None declared.

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