Measurement accuracy of total cell volume by automated dialyzer reprocessing: A prospective cohort study

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HIGHLIGHTS

- Over the past decade, dialyzer reprocessing machines have replaced human labor and time spent in preparing re-usable dialyzers.
- It also made the process of total cell volume (TCV) measurement become faster.
- Volumetric evaluation was considered as the standard to compare with the TCV values from the reprocessing machine.
- Nevertheless, there has been a lack of data on efficacy of weight evaluation on TCV by machine compared to volume evaluation by the conventional method.
- The aim of study was to evaluate the efficacy of TCV measurement performed by the reprocessing machine compared to that of the conventional method.

ARTICLE INFO

ABSTRACT

Introduction: Dialyzer reprocessing machines have replaced human labor in preparing re-usable dialyzers. It also made the process of total cell volume (TCV) measurement become faster. Nevertheless, there has been a lack of data on efficacy of weight evaluation on TCV by machine compared to volume evaluation by the conventional method. The aim of this study was to evaluate the efficacy of TCV measurement performed by Kidney-Kleen® reprocessing machine, produced by MEDITOP Company in Thailand, compared to that of the conventional method.
Methods: This prospective cohort study was performed during September 2014 to December 2015.The low-flux (N = 101) and high-flux dialyzers (N = 100) were included for TCV evaluation. Reused times were up to 5 in the low-flux and 20 in the high-flux dialyzers. The Bland Altman analysis was used to evaluate value measured by different methods.
Results: The values measured by weight evaluation (by machine) were higher than those obtained by volumetric evaluation of the conventional method in the low-flux (0.81 ± 0.20%) and high-flux (1.32 ± 0.39%) dialyzers. The correlation of TCV values of the two methods were r = 0.98, *p < 0.001* and r = 0.71, p < 0.001 for the low- and high-flux dialyzers. Moreover, there was robust association and agreement between the two methods, confirmed by the Bland-Altman Analysis, which suggested that the values acquired by machine were within the limits of agreement, indicating acceptable accuracy of equipment.
Conclusion: The approach of measurement differed from that of the conventional method (weight evaluation was used instead of volumetric evaluation), the reprocessing machine could offer accurate results.

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1. Introduction

A hemodialyzer is an instrument that has been used universally to purify fluid and waste metabolites from the blood of renal failure patients. Different types of dialysis membrane (flux) were categorized by the clearance of β2 microglobulin across membrane during hemodialysis. The dialyzers with β2 microglobulin clearance less than 20 ml/min are called 'low-flux dialyzer', usually used for small uremic toxin removal in acute kidney injury. Meanwhile, the dialyzers with β2 microglobulin clearance more than 20 ml/min are called 'high-flux dialyzer', usually used for middle molecular size removal such as in setting of chronic hemodialysis for end-stage renal disease patients.

Reprocessing dialyzer machines have been used worldwide for economic advantage [1–4], improvement in blood-dialyzer membrane biocompatibility, and benefits of preventing the first-use syndrome which is an anaphylactoid reaction to the dialysis membrane causing wide-range of symptoms including cardiac arrest [5–7]. The machines have helped shorten the period of cleaning, leak testing, and sterilant filling. However, there have been still some concerns about the use of machines such as infection. The Centers of Disease Control and Prevention (CDC) have recommendations against dialyzer reuse in patients with active bacterial and hepatitis B infection [8–11]. Decline in dialyzer performance after reuse has also been of concern. Performance indices can be measured by two approaches, namely total cell volume (TCV) measurement and urea clearance evaluation. The KDOQI guidelines [1] have suggested that a dialyzer is suitable for reuse only when a TCV value is at least at 80% of the baseline or the urea clearance of the dialyzer is at least at 90% of the original value [12–15].

TCV, one of the parameters indicating dialyzer performance mentioned above, refers to the volume of the blood compartment of a dialyzer. A TCV value is determined by measurement of volume of water being filled in a blood compartment of a dialyzer either with the conventional method or with automated reprocessing machines. With the conventional approach, a dialysis nurse fills reverse-osmosis (RO) water into the blood compartment of dialyzer and later measures the volume of water flowing out of the compartment equipped with an air pump. With the development of the reprocessing machines, several hemodialysis centers have replaced the conventional TCV evaluation with an automated method in addition to the cleaning of dialyzer. Evaluation of TCV relies on the principle of fluid mechanics by volumetric evaluation. There has been an attempt to discover the best indirect approach to measure TCV in order to substitute volumetric evaluation performed by human such as weight measurement, hydrostatic pressure measurement, and ultrasonic detection [16–20].

Kidney-Kleen® employs weight measurement, one of the most popular techniques, to determine TCV. Weight measurement is an indirect approach to measure and translate weight into volume, based on an assumption that 1 mg of water is equal to 1 mL of water. However, several factors may have affected on the weight measurement of TCV by the reprocessing machine such as space-occupying air bubbles, weight of debris particles in patient's blood, incomplete collection of fluid from dialyzers' membrane. By using the automated approach, the reprocessing and the TCV measurement are done simultaneously, and the healthcare provider may benefit from reduced human workload and shortening of overall process time. Nevertheless, the efficacy of TCV measurement by weight has not been widely studied since the main purpose of the reprocessing machines was to clean the dialyzer, not to measure the TCV. Therefore, this study is the first to compare the efficacy of conventional measurement and automated approach. Our hypothesis is that TCV measurement from volumetric measurement (manual method) can be cost-beneficially replaced by the weight measurement (automated machine).

2. Materials and methods

2.1. Clinical data collection

This prospective cohort study was performed at the hemodialysis unit within HRH Princess Maha Chakri Sirindhorn Medical Center, Srinakharinwirot University, Thailand. Our study was conducted during September of 2014 to December of 2015. All dialysis patients receiving either high or low flux dialysis during this period were informed of study, and all gave full consent to participation.

In this study, the low-flux dialyzer, was equipped with synthetic polysulfone membrane with 1.5 m^2 effective surface area and 90 mL of TCV (Diacap Polysulfone® LO PS 15 Dialyzer) and the high-flux dialyzer was synthetic polynephron membrane dialyzer with effective surface area of 1.9 m^2 and 115 mL of TCV (Elisio-190HR®). The protocol and patient's participation were approved by the Human Research Ethics Board of Srinakharinwirot University (Issue #SWUEC-X-037/2557).

The reused times were up to 5 times in the low-flux and 20 times in the high-flux with acute kidney injury (AKI) and end stage kidney disease (ESRD), respectively. Patients with HIV, hepatitis B, hepatitis C infection and suspected sepsis or bacteremia were excluded. All patients were dialyzed for 4 h per a dialysis session which was maintained by an initial loading of intravenous heparin 3000 IU, followed by hourly bolus of heparin 1000 IU intravenously. Each dialyzer was reprocessed with formaldehyde 4% and reused again for the same patient only when TCV was ≥80% of the original value.

2.2. TCV evaluation

All the dialyzers were cleaned by the reprocessing machine (Kidney-Kleen®) before the measurement of TCV was performed. TCV was measured for the evaluation of quality of each dialyzer values were calculated as a percentage ratio compared to the baseline value.

2.3. The conventional TCV evaluation

After the cleaning process by machine, a TCV was first measured by machine. A dialyzer was then removed and underwent 2 separate TCV conventional evaluations by 2 blinded dialysis nurses who have at least 5-year experience on cleaning processes of dialyzer. Both the dialysis nurses were blinded to the patient's clinical presentation, any value from the machine, and the value of TCV obtained from each other. The blood and dialysate compartment of the machine were filled with reverse osmosis (RO) water, and TCV was subsequently measured by evacuating water from the blood compartment with an air pump. Two manual TCV values from the same dialyzer were averaged and used as its reference value for quality evaluation and onward comparison with the value obtained from the machine. Any dialyzer with referenced TCV of less than 80% of its original value was discarded (<72 mL and <92 mL for low-flux and high-flux machines, respectively).

2.4. The reprocessing machine TCV evaluation

Similar to the conventional TCV measurement, RO water was filled into the blood compartment vertically from bottom to top of the dialyzer. Later, water was evacuated from the blood compartment into a measure tank and the weight of water was measured by a load cell sensor as shown in Fig. 1.
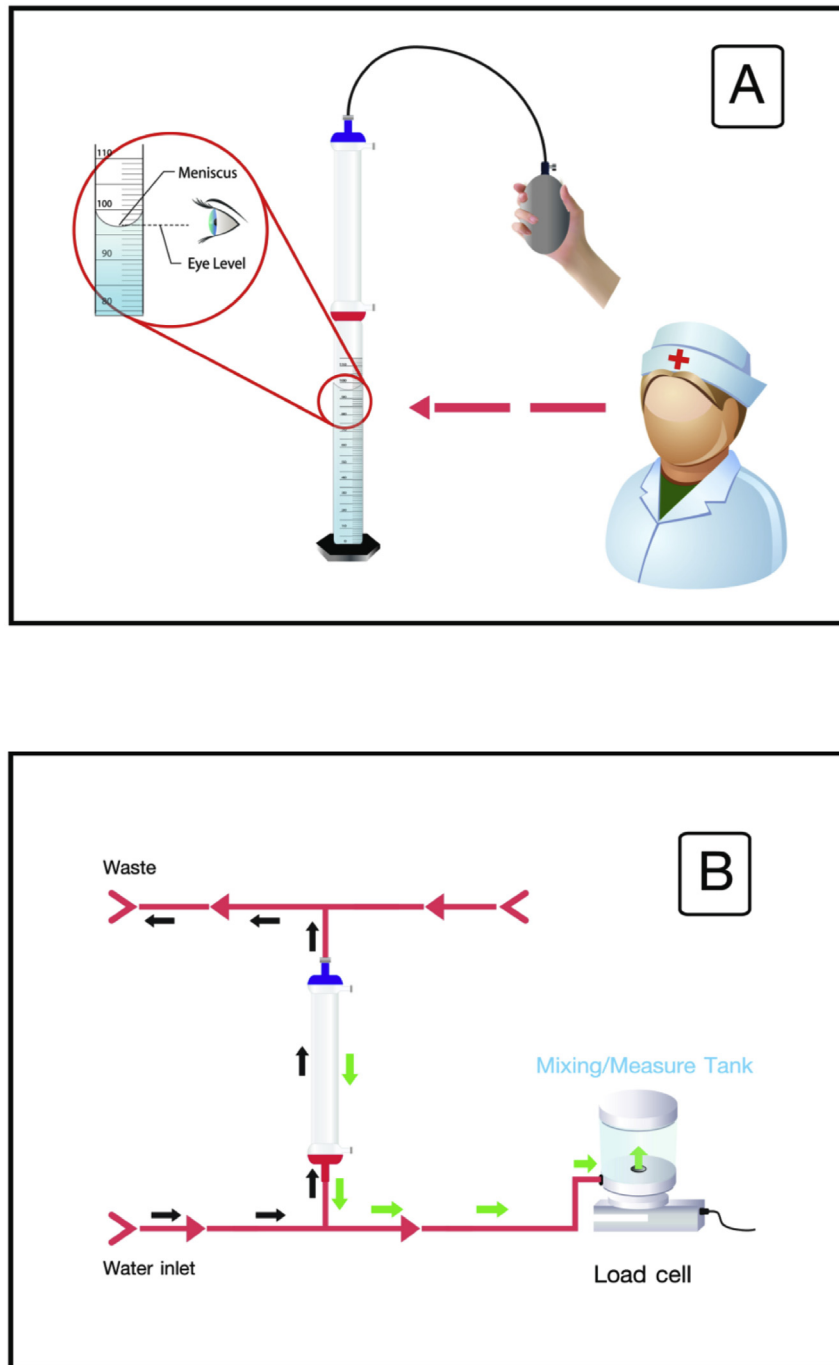
Fig. 1. A; Conventional method (volumetric evaluation): a dialysis nurse filled up RO water into blood and dialysate compartments to expel air within a dialyzer. Then an air pump pressured water from the blood compartment into a cylinder. The nurse assessed a volume at the lowest point of fluid curve (lower meniscus).The volume was measured independently by two nurses and the values were pooled together to find average TCV values. **B**; Automated machine method (weight evaluation): RO water flew through a water inlet and went into a blood compartment (indicated by black arrows). Both water and air were then removed out of the dialyzer through the waste line. Later, the machine evacuated water in the blood compartment by generating negative pressure, making water flow out of the dialyzer and flow into a measure tank (depicted by green arrows). The measure tank then measured water weight and translated it into volume.

2.5. Statistical analysis

Data analyses were performed by using R software (version 3.2.1).Continuous variables were presented as means ± standard deviations; categorical variables were presented as percentages. Differences between groups of patients were compared with the Pearson chi square test for categorical variables and with the 2-tailed student's T -test for continuous variables. The correlation of selection algorithms between continuous variables and ordinal variables were tested with the Spearman correlations coefficient. The Bland Altman plot was employed to evaluate the agreement of the two measurement techniques.The 95% limits of agreement of Bland Altman plot ($\bar{d}$) was calculated by $\bar{d} \pm 1.96$ = mean difference ±(1.96 × standard deviation of the difference) [21] and p-values≤ 0.05 were considered statistically significant.

The risk of committing a type II error was accepted only less

than 5%. Hence, the result of the conventional TCV evaluation method would be discarded if the values between 2 dialysis nurses were different >5%. The sample size was analyzed for the paired sample T test. $N = 4(SD^2/\overline{d}^2)$ with 95% confidence [22], when N = sample size, SD = standard deviation and $\overline{d}$ = mean of difference. The previous observational data from the center showed SD = 1.6% with $\overline{d}$ = 4.2% and SD = 2.0% with $\overline{d}$ = 5.7% for the low-flux and high-flux dialyzers. Subsequently, the minimal estimate sample size was calculated, yielding requirements of 56 and 66 of TCV evaluation for the low-flux and high-flux dialyzers, respectively. We finally decided to include the 100 samples of each item to the accuracy of our study.

3. Results

3.1. Patient demographic data

We evaluated TCV from total 201 reused dialyzers of two different techniques of evaluation (conventional vs. machine) from 51 patients. Forty one patients (male 54.9% with age average (mean ± standard deviation) 60.9 ± 18.9 years) were diagnosed with acute kidney injury (AKI) or AKI on top of advanced chronic kidney disease (CKD stage 3–4), receiving acute dialysis with a low-flux dialyzer (N = 101) and 10 patients (male 60.0% with age average 60.5 ± 8.0 years) were diagnosed end-stage renal disease (ESRD; CKD stage 5) receiving chronic dialysis with a high-flux dialyzer (N = 100). Causes of AKI/ESRD as well as indication of dialysis were shown in Table 1. The average (mean ± standard deviation) reused times were 3.2 ± 1.7 times/dialyzer and 12.0 ± 5.0 times/dialyzer for the low-flux and high-flux dialyzer machines, with the total reused times up to 5 times and 20 times, respectively.

3.2. The agreement analysis

The purpose of this study was to compare the efficacy of TCV measurement by weight evaluation by using the automated reprocessing machines compared to that of volumetric evaluation by using conventional technique. Each dialyzer needed to be measured TCV twice by the two methods. To avoid interference with quality of dialysis membrane, we measured TCV by machine before measuring it by the conventional method for comparison.

Agreement bias might occur when the same object (a dialyzer) is measured by different methods. Though the different methods yield results in the same direction, it is hard to evaluate true efficacy of each method. Hence, the Bland Altman analysis was used to eliminate such bias in this study where the performance of the same dialyzer was measured by different methods.

We used the TCV values from conventional method as a reference. The different values between the two methods were calculated for each dialyzer for each time of reuse. The results showed that all values of the mean TCV evaluated the weights obtained by machine, both in the low-flux and high-flux dialyzers, were higher than the volumes measured by the conventional method, causing different TCV values to become negative. We then calculated the mean of different values of TCV in the same time of reused which were called **"the mean difference"**. We obtained 5 mean difference TCV values of the low-flux dialyzers (N = 101) and 20 mean difference TCV values of the high-flux dialyzers (N = 100) from 5 to 20 reused time, respectively. Consequently, the mean difference TCV values were plotted and analyzed by the Bland Altman analysis to figure out an agreement interval as shown in Figs. 2–4.

3.3. The TCV agreement in low-flux dialyzers

The mean different values and percentages of mean different of the reused TCV low-flux dialyzers measured by the 2 methods were illustrated in Table 2. There was a significant correlation between conventional and machine evaluations in term of the values measured (r = 0.98, p < 0.001). The Bland-Altman plot, shown in Fig. 2, indicated good distribution of values and the limits of agreement was valid. The plot also demonstrated the average of mean difference value which was −0.71, with a lower limit of −1.05 and an upper limit of −3.63. All of the mean difference values were in the limits of agreement. The average of percentage error (percent of the difference of mean) after reuse times up to 5 times was 0.81± 0.20% in case of the low-flux dialyzers (Table 2).

3.4. TCV agreement in high-flux dialyzers

Similar to the agreement in the low-flux dialyzers, the Bland-Altman plot also showed an average mean difference value of −1.67 with the limit of agreement possessing a lower limit

Table 1
Patient demographic data.

	Number of Patient	Number of Dialyzer
Acute dialysis with low-flux dialyzers (number)	41	101
	Male 54.9%	
	Age (mean ± SD) 60.9 ± 18.9 years	
Cause of AKI (%)		
Acute Tubular Necrosis (including toxin, cardiogenic shock, etc.)	90.2	
Acute glomerulonephritis	4.9	
Post renal (obstructive) AKI	4.9	
Indication of acute dialysis (%)		
Volume overload	56.1	
Uremia	39.0	
Hyperkalemia (K > 5.5)	2.4	
Severe metabolic acidosis (serum bicarbonate <15 mEq/L)	2.4	
Chronic dialysis with high-flux dialyzers (number)	10	100
	Male 60.0%	
	Age (mean ± SD)	
	60.5 ± 8.0 years	
Cause of ESRD (%)		
Diabetic nephropathy	80	
Chronic glomerulonephropathy	10	
Unknown cause of CKD	10	

Abbreviations; AKI = acute kidney injury, ESRD = end stage renal disease, CKD = chronic kidney disease, SD = standard deviation, K = potassium, mEq/L = milliequivalent per litre.
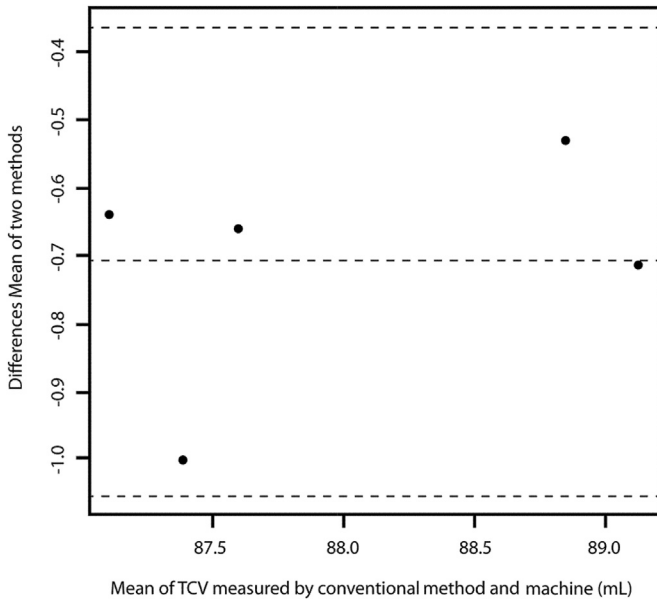
Fig. 2. The Bland Altman plots show the mean difference of TCV (each black ball) of low-flux dialyzers between volumetric and weighing evaluation. The average of mean difference was −0.71 (lower limits as −1.05 and upper limit as −3.63). All values of the mean difference (each black ball) were placed in the limits of agreement, which indicated positive correlation between methods to evaluate TCV in low-flux dialyzers.
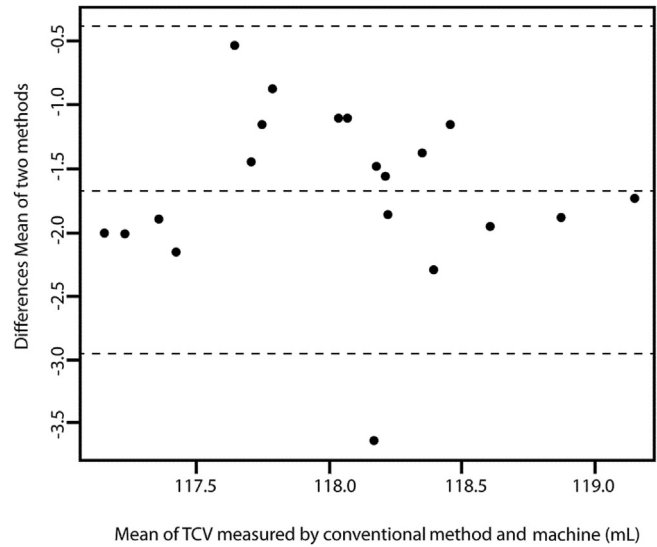

Fig. 3. The Bland Altman plots show the mean difference of TCV (each black ball) of high-flux dialyzers between volumetric and weighing evaluation. The average of mean difference was −1.67 (lower limit as −2.95, and upper limit as −0.38). All values of the mean difference (each black ball) were placed in the limits of agreement which indicated good correlation between the methods to evaluate TCV in high-flux dialyzers.

of −2.95 and an upper limit of −0.38 (Fig. 3). There was a good correlation of TCV between two methods (r = 0.71, p < 0.001), and the average of percentage error (difference mean of percent) after reuse times up to 20 times was 1.32 ± 0.39% in case of the high-flux dialyzers (Table 3 and Fig. 3).

3.5. The subsequent period agreement for TCV evaluation in the high-flux dialyzers

Reuse of the high-flux dialyzers up to 20 times could decrease membrane performance, affecting correlation in the Bland-Altman Analysis. Thus, we divided the period of use into 1–5, 6–10, 11–15, and 16–20 reused times and made a further analysis to figure out agreement among all periods in order to minimize confounding effects of the decline in membrane performance after reuse. Results were reported in Table 4 and Fig. 4A–D.

The subsequent agreement analysis showed that all the mean different values were in the limit of agreement as −1.95 (limit as −2.36 to −1.53), −0.96 (limit as −2.93 to 1.00), −0.96 (limit

as −1.49 to −0.44) and −1.41 (limit as −1.72 to −1.10) for 1–5, 6–10, 11–15, 16–20 reused time, respectively. In addition, the difference mean of percent (Table 4) demonstrated the values of error would be similar as whole analysis as higher than that of the conventional method by 1.64 ± 0.18%, 0.82± 0.85%, 0.82 ± 0.23%, and 1.20 ± 0.14% for 1–5, 6–10, 11–15, and 16–20 reused time analysis, respectively.

4. Discussion

Reprocessing dialyzer machines have been used widely for several advantages. The accuracy of TCV measurement by machine using weight evaluation compared to that of the conventional method using volume evaluation was confirmed by the results of present study. Though the TCV values of the automated machine were higher than those of the conventional method, the debris or particles in the patient's blood may have interfered with the load cell sensor, increasing the weight of fluid measured. The TCV values obtained by the two approaches showed positive correlation and significant agreement when analyzed by the Bland-Altman

Table 2
The different TCV values of low-flux dialyzers between reprocessing machine and conventional method.

Reused time	Conventional TCV (mL)	Machine TCV (mL)	Mean of TCV (mL)	Mean difference of TCV[d] (mL)	Percent of the difference of mean (%)
0	92				
1	88.77	89.48	89.13	−0.71	−0.80
2	88.58	89.11	88.85	−0.53	−0.60
3	87.27	87.93	87.60	−0.66	−0.75
4	86.89	87.89	87.39	−1.00	−1.14
5	86.79	87.43	87.11	−0.64	−0.73
$\bar{d}$				−0.71	−0.81
SD				0.18	0.20
The limits of agreement				−1.05 to −3.63	
Pearson correlation				R = 0.98*	

Symbols: $\bar{d}$ = mean of difference, * = statistically significant (p < 0.001), d = direction of difference (The minus values suggest that the values measured by machine were higher than those obtained by the conventional method.).
Abbreviations; TCV = total cell volume, SD = standard deviation, mL = milliliter.

Table 3
The different TCV values of high-flux dialyzers between reprocessing machine and conventional method.

Reused time	Conventional TCV (mL)	Machine TCV (mL)	Mean of TCV (mL)	Mean difference of TCV[d] (mL)	Percent of the difference of mean (%)
0	124				
1	118.28	120.02	119.15	−1.74	−1.46
2	117.94	119.82	118.88	−1.88	−1.58
3	117.25	119.55	118.40	−2.30	−1.94
4	117.63	119.59	118.61	−1.96	−1.65
5	117.29	119.15	118.22	−1.86	−1.57
6	116.35	118.51	117.43	−2.16	−1.84
7	116.41	118.31	117.36	−1.90	−1.62
8	116.15	118.16	117.16	−2.01	−1.72
9	116.23	118.24	117.24	−2.01	−1.71
10	116.35	117.99	117.17	−1.64	−1.40
11	117.38	117.91	117.65	−0.53	−0.45
12	117.35	118.23	117.79	−0.88	−0.75
13	117.48	118.60	118.04	−1.12	−0.95
14	117.17	118.34	117.76	−1.17	−0.99
15	117.50	118.62	118.06	−1.12	−0.95
16	117.43	118.93	118.18	−1.50	−1.27
17	117.73	119.00	118.37	−1.27	−1.07
18	117.67	119.04	118.36	−1.37	−1.16
19	117.88	119.04	118.46	−1.16	−0.98
20	116.98	118.44	117.71	−1.46	−1.24
$\bar{d}$				−1.67	−1.32
SD				0.66	0.39
The limits of agreement				−2.95 to −0.38	
Pearson correlation				R = 0.71*	

Symbols: $\bar{d}$ = mean of difference, * = statistically significant (p < 0.001), d = direction of difference (The minus values suggest that the values measured by machine were higher than those obtained by the conventional method.).
Abbreviations; TCV = total cell volume, SD = standard deviation, mL = milliliter.

Analysis.

It is widely accepted that reuse of the same dialyzer could cause alteration of membrane pore sizes and also lead to twist of hollow fibers, resulting in decline in membrane performance [23,24]. Multiple use of the same dialyzer may have also interfered with the results by increasing variation of the mean ± SD and acted as a confounder. We used mean ± SD in determination of the upper and lower limits of agreement, so any error in mean ± SD could affect the results of statistical analysis. The plots out of the limits of agreement could be an error resulting from the variation caused by repetitive use of the same dialyzer. Subsequent analyses on each period of reuse aiming to reduce effects of such confounder showed the same results in both the conventional and automated machine groups. The results of the subsequent analysis indicated that the decline in membrane performance did not affect agreement on TCV evaluation by machine when compared to the conventional method. However, for the last reuse time of high-flux, the average TCV value was approximately 94% when measured by the conventional method (data not shown). The decline in membrane

performance may not have been significant enough to be detected as a confounder. Nevertheless, it is our conviction that repetitive use of the same dialyzer for more than 20 times or the reuse until a TCV value was less than 85%–90% could lead to error which may have affected the results of statistical analysis and agreement between the two methods. In addition, the limitation of the study is the order effect, measuring the TCV by machine before by the conventional method, might show the different results causing from more filling fluid volume of reprocessing methods by machine. For this reason, there should be further studies on effects of such repetitive use.

Due to the lack of data on the reliability of performance when the reuse times are more than 5 and 20 times in low-flux and high-flux dialyzers, it is necessary that TCV evaluation should be measured by the conventional method to figure out a performance index before reuse. In addition, if a TCV value is less than 85% when measured by machine, we strongly recommend TCV measurement by the conventional approach. It is because when an actual TCV was less than 80%, the membrane performance may have not been able

Table 4
Summary of the mean differences of reused time for each period of high-flux dialyzers.

Mean of differences[d]	SD	The percent of mean different (% ± SD)[d]	Lower limit	Upper limit
Total (1−20 reuses)				
−1.67	0.66	−1.41 ± 0.56	−2.95	−0.38
Reused 1−5				
−1.95	0.21	−1.64 ± 0.18	−2.36	−1.53
Reused 6−10				
−0.96	1.01	−0.82 ± 0.85	−2.93	1.00
Reused 11−15				
−0.96	0.27	−0.82 ± 0.23	−1.49	−0.44
Reused 16−20				
−1.41	0.16	−1.20 ± 0.14	−1.72	−1.10

Symbols: d = Direction of difference (The minus values suggest that the values measured by machine were higher than those obtained by the conventional method.).
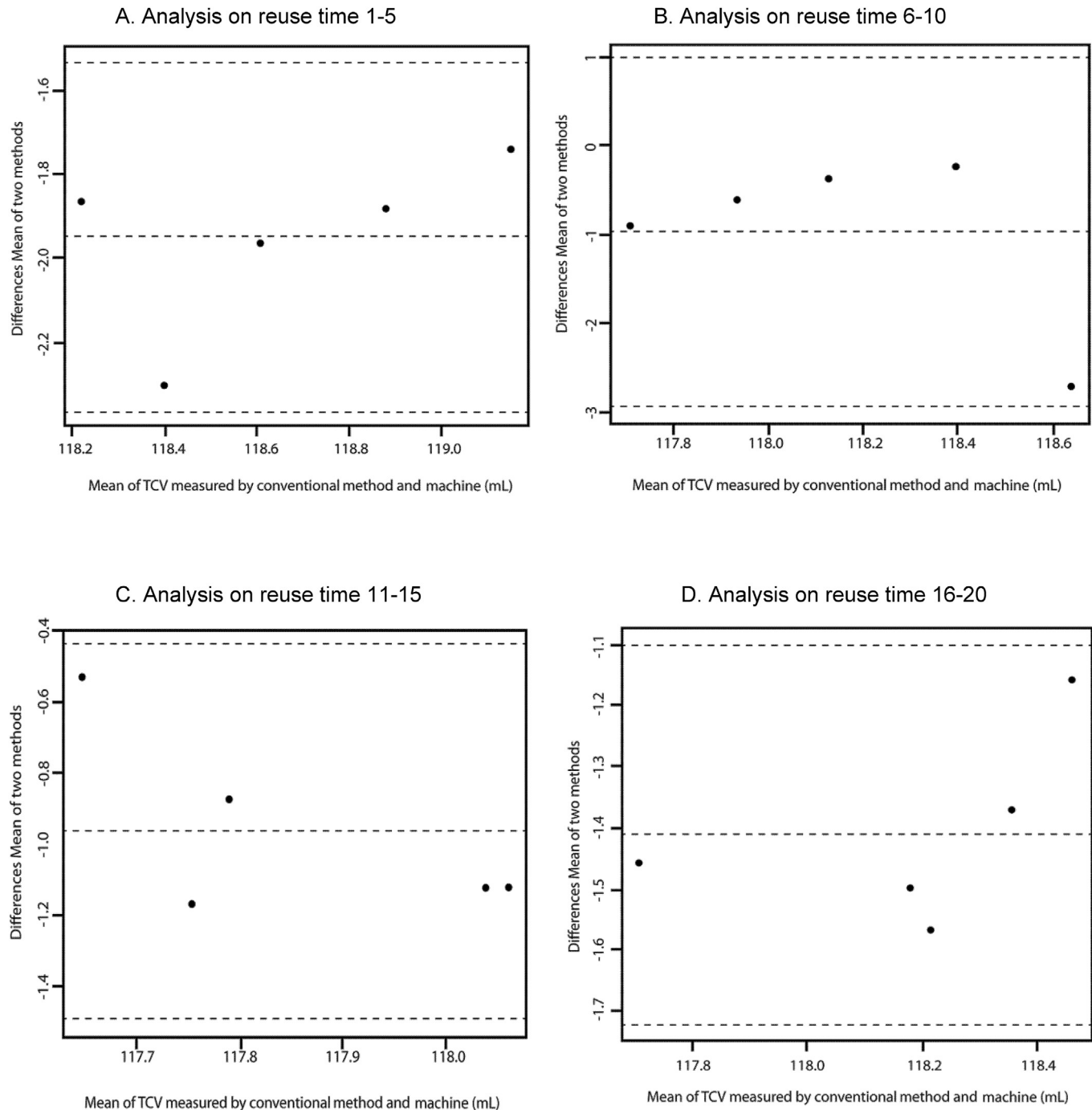Abbreviations; SD = standard deviation.

A. Analysis on reuse time 1-5

B. Analysis on reuse time 6-10

C. Analysis on reuse time 11-15

D. Analysis on reuse time 16-20

Fig. 4. The Bland Altman plots show results in each period of reused time of high-flux dialyzers. Black balls represent the mean difference of TCV in each time of reuse. This subsequent Bland Altman analysis has been done for A) 1—5, B) 6—10, C) 11—15, and D) 16—20 reused times. The values of mean difference (each black ball) were placed in the limits of agreement which indicate good correlation between the methods in high-flux dialyzers.

to remove waste products from patients.

Our study is the first to compare the efficacy of TCV measurement of weight evaluation by machine compared to that of volumetric evaluation by the conventional method. The results from the two methods were comparable and showed positive correlation. The development of any automated machine evaluating TCV should use the range of acceptable error of not more than 1% in case of low-flux dialyzers, and not more than 2% in case of high-flux dialyzers. It is because our results demonstrated the average of percentage error of $0.81 \pm 0.20\%$ and $1.32 \pm 0.39\%$ in the two types of dialyzers, respectively and our results were into the limits of agreement. In

conclusion, the accuracy of TCV measurement by machine using weight evaluation compared to that of the conventional method using volume evaluation was confirmed by the results of present study. Hence, TCV measurement by machine could be used with confidence to replace the conventional TCV evaluation in order to reduce workload and time, as well as to provide better care for patients undergoing hemodialysis.

Ethical approval

The protocol and patient's participation were approved by the

Author contribution

Chatchai Kreepala;- study design, data collections, data analysis, writing manuscript.
Aroonchai Sangpanich-data collections.
Phirudee Boonchoo-data collections.
Warit Rungsrithananon-writing manuscript.

Conflicts of interest

The authors have no competing financial interests.

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