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## Analyzing the Quality of Dialysis Machines Input Water in Hospitals of Kashan City, in 2019

Samaneh Mehraban Navaz Kohan <sup>1</sup>, Gholam Reza Mostafaii <sup>2</sup>, Hosein Akbari <sup>2</sup>, Mohammad Bagher Miranzadeh <sup>1</sup>\*

- <sup>1</sup> Department of Environmental Health Engineering, Faculty of Health, Kashan University of Medical Sciences, Kashan, Iran.
- <sup>2</sup> Department of Biostatistics and Public Health, Faculty of Health, Kashan University of Medical Sciences, Kashan, Iran.

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#### \*Corresponding Author:

Mohammad Bagher Miranzadeh

Email:

miranzadehm@ymail.com

Tel:

+989131612852

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#### **ABSTRACT**

*Introduction:* According to high volumes of water used in hemodialysis, quality of water entering the dialysis machine is very important. The current study aims to analyze microbial and chemical quality of water used for hemodialysis in hospitals of Kashan city in 2019.

*Materials and Methods:* This descriptive cross-sectional study was performed on 54 water samples used in dialysis machines in hospitals of Kashan city during 3 months of the fall season in 2019. Microbial tests of the samples were done, and also heavy metals were assessed using inductively coupled plasma optical emission spectrometry. Statistical tests, sample t-test, and ANOVA were used to compare the mean results with standards.

**Results:** Based on the results, the mean concentrations of magnesium (Mg) (2.7  $\pm$  2.22 mg/L), sulfate (13.09  $\pm$  21.06 mg/L), sodium (Na) (17.27  $\pm$  24.47 mg/L), and potassium (K) (0.09  $\pm$  0.17 mg/L) in all samples were based on the standard levels. However, the mean concentrations of nitrate (3.22  $\pm$  1.21 mg/L), aluminum (Al) (0.26  $\pm$  0.16 mg/L), silver (Ag) (0.52  $\pm$  0.85 mg/L), lead (Pb) (0.08  $\pm$  0.13 mg/L), and zinc (Zn) (0.91  $\pm$  0.71 mg/L) were above standard levels in all the samples. Thallium (Tl) ion was reported to be zero. Moreover, heterotrophic bacteria were not observed in any of the samples.

**Conclusion:** Given the high concentration of chemicals and heavy metals in dialysis machines water input, it is necessary to plan for periodic monitoring of water treatment systems and heavy metals and regular replacement of reverse osmosis filters.

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#### Introduction

Dialysis is one of the most common methods used to treat patients with acute kidney failure <sup>1</sup>. This method is applied directly (hemodialysis) or indirectly (peritoneal dialysis) using a semipermeable membrane <sup>2,3</sup> and plays an effective role in controlling blood pressure and maintaining electrolytes balance in the body by removing toxins, salts, and excessive fluids from the body <sup>4, 5</sup>. According to statistics, at the end of 2010, about 1 million people have received

dialysis worldwide, 60% of them were from the United States of America, Japan, Germany, Brazil, and Italy <sup>6</sup>. During dialysis process, a significant amount of water is used for the preparation of dialysis fluid, so that each patient is exposed to 400 to 600 liters of water per week <sup>7-9</sup>, which increases to 580-860 liters in patients under the 24-hour treatment 10. Patients that receive hemodialysis usually from cardiovascular hypertension, diseases, diabetes, making them vulnerable

1734

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environmental conditions and infectants<sup>11, 12</sup>. The presence of microbial and chemical pollutants, such as monochloramine, copper (Cu), and zinc (Zn) in dialysis water can cause anemia, fever, cardiovascular changes, nausea, vomiting, and hypotension<sup>9, 13-15</sup>. Therefore, many of dialysis centers use reverse osmosis for the preparation of contaminant-free water<sup>14</sup>. Chemical and microbial quality of dialysis water is, therefore, important to prevent additional risks to patients and to ensure the safety of dialysis<sup>7, 16</sup>. The association for the advancement of medical instrumentation (AAMI) has developed standards to control the water quality of dialysis centers<sup>17, 18</sup>. However, several studies have reported the contamination of water used in dialysis centers across the world 19-22. Pisani et al. 23 and Heidarieh et al. 24 showed that the amount of viable heterotrophic bacteria in the studied samples was always more than recommended levels. Furthermore, the study by Okunola and Olaitan on 10 dialysis centers in Nigeria revealed that numbers of viable bacterial colonies in the analyzed samples were more than the recommended levels by AAMI <sup>6</sup>. Suzuki et al., in their study, reported the presence of pollutants, such as Cu, nitrate, and aluminum (Al) at higher concentrations<sup>25</sup>. However, Ibrahim et al. in their study on 5 dialysis centers did not observe different results. They showed that the level of chemical compounds in dialysis water of many of hospitals was within the standard range and bacterial contamination was observed in 40% of cases<sup>4</sup>. In addition, a study on 5 dialysis centers in Isfahan, central Iran, revealed that magnesium (Mg), cadmium (Cd), and chromium (Cr) levels in some centers were above standard levels and no bacterial contamination was observed 7. However, in similar studies, the use of mixed methods, including reverse osmosis electrodialysis has been less common. Therefore, given the important role of water treatment in dialysis centers and also the effect of microbial and chemical quality of water used in dialysis fluid on health and increasing the life expectancy of patients and using large volumes of water for dialysis per week, the present study aims to

investigate the microbial and chemical quality of water entering the dialysis machines of Kashan Hospitals in 2019.

#### **Materials and Methods**

#### Materials and reagents

All reagents used in water sample tests were from commercial sources, such as MERK (USA) and Sigma-Aldrich (Germany). The reagents used in the tests included ammonia buffer, EDTA 0.01 M, Eriochrome black T, morxide, sodium hydroxide solution 1N, hydrochloric acid 1N, sulfanilamide, N-(1-Naphthyl)ethylenediamine dihydrochloride, SPANDS solution, zirconium acid, barium chloride, and R2A agar.

#### Equipment

In this study, RC Meter-24P digital colorimeter was used for measuring residual chlorine and PH. WTW conductivity meter 730 was also used for measuring TDS, and **HACH-Lange** spectrophotometer (model DR2800) was utilized for measuring nitrate, nitrite, sulfate, and fluoride. Flame photometer (modelG620, Iran) was used for measuring sodium (Na) and potassium (K). Moreover, the study utilized induction plasma spectrometer (model ICP-OES; Perkin Elmer; Optima 2100 DV model, USA) for different purposes. They include measuring heavy metals with the power of the device to generate a radio frequency of 1300 watts, plasma gas flow rate of 15 liters per minute, auxiliary gas flow rate of 0.2 liters per minute, and gas spray speed of 0.8 liters per minute with nebuliser.

#### Sampling method

This cross-sectional descriptive study was conducted on water treatment devices of hospitals with dialysis centers in Kashan city, including 4 hospitals and 6 water treatment devices. Sampling was done with 3 replicates in 3 months of the fall season in 2019 by simple random sampling at the beginning, middle, and end of each month when the reverse osmosis device was operating. A total of 54 samples were examined. The water source of hospitals number 1 and 2 was from wells and hospitals number 3 and 4 was from the urban distribution network. From each location, two

samples were taken. A sample was collected for bacteriological tests in sterile glass containers with sanding cap with a volume of 300 ml inside the cold box and another sample was taken for chemical tests in 2 liter plastic containers. The samples were then transferred to the laboratory of Kashan University of Medical Sciences. In order to store the chemical samples for measuring heavy metals, 1.5 cc of concentrated nitric acid was added to each liter of sample and the pH value was reduced to less than 2.

### Microbial test of heterotrophic plate count bacteria by pour plate method

R2A agar medium was used to measure heterotrophic bacteria colonies. After melting this culture medium close to the flame, 1 cc of the sample was poured into the plate using a sterile pipette and the melted culture medium was added to it and mixed with a circular motion. The plates were placed upside down in the incubator 35 for 48 h. At the end of the culture time, the colonies on the plate were counted using colony count and reported as CFU in one ml of the sample.

#### Chemical tests

## Residual chlorine and pH measurement using RC Meter-24P digital colorimeter

The water was first allowed to flow for 1 to 2 min. Then, the device chamber was placed under water to fill. When water was spilling, the power key and the start key were then pressed to indicate a fixed number. The residual chlorine was then recorded and the pH was measured at the same time.

## Measurement of total dissolved solids using WTW conductivity meter

First, the sample was shaken well to make a uniform solution, then, the device was turned on and the device was calibrated using 0.01 M potassium chloride solution. The electrode of the device was then inserted into the sample and allowed to display the total dissolved solids (TDS) in milligrams per liter on the device monitor.

Total hardness test: 50 cc of the sample was poured into an Erlenmeyer flask and 2-5 ml of

ammonia buffer was poured under the hood to reach pH 10, then, 0.2 to 0.1 g of Eriochrome black T reagent was added. The red color was titrated by 0.01 M EDTA and continued until the appearance of blue color (end point).

The titration was performed within 5 min from the time of increasing the buffer. The total hardness was calculated from the volume of EDTA according to the following formula.

Total hardness ml/L of calcium carbonate =

 $(V \times F \times 1000)/(Ml \text{ sample})$ 

V: EDTA volume

F: EDTA factor

Calcium (Ca) hardness test: 50 ml of the sample or diluted portion to 50 ml was poured into a 250 ml Erlenmeyer flask. The pH was raised to about 12-13 by 3 ml of normal sodium hydroxide and 0.1-0.2 g of moroxide reagent was added using a spatula. After adding the reagent to the sample, a pink color was created then it was titrated with EDTA until the appearance of purple color (end point) and the volume of EDTA was recorded.

Ca hardness mg/L of Caco3 =  $(A \times B \times 1000)$ / (Ml sample)

A = EDTA factor

B = EDTA volume

Ca ion content = Ca hardness  $\times$  0.4

Mg hardness: Total hardness is the sum of Ca hardness and Mg hardness, so by having total Ca and calcium hardness, Mg hardness is calculated as follows:

Mg hardness = total hardness - Ca hardness Mg ion = Mg hardness  $\times$  44.2

Nitrate test: 50 ml of the sample was poured into the Erlenmeyer flask and 1 ml of 1 N hydrochloric acid was added. The amount of nitrate ion was read at a wavelength of 220 nm.

Nitrite test: 50 cc of the sample was poured into the Erlenmeyer flask and 2 ml of sulfanilamide reagent and 1 ml of N-(1-Naphthyl) ethylenediamine dihydrochloride reagent were added. After 10 min, the amount of nitrite ion was read at a wavelength of 543 nm using a spectrophotometer.

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1736

Fluoride test: 50 cc of the sample was poured into the Erlenmeyer and 5 ml of SPANDS solution and 5 ml of zirconium acid reagent were added. The amount of fluoride ion was read at a wavelength of 570 nm using a spectrophotometer.

Sulfate test: 1 ml of the sample was pour into 250 ml Erlenmeyer flask and 20 ml of buffer + barium chloride solution in about 1 g was added and stirred for one minute using an electric stirrer. The absorbance of the samples was recorded in 5 minutes (± 0.5 minutes) by spectrophotometer at 40 wavelengths.

Na test: First, the device was turned on and a blue flame was created by adjusting the air pressure and gas flow. Then, distilled water (control) was injected into the device and it was set to zero. Na standard solution (50 and 25 ppm) was injected into the device and the range of the device was adjusted according to the standard concentration. Then, the sample was injected into the device and Na concentration was read.

K test: First, the device was turned on and a blue flame was created by adjusting the air pressure and gas flow. The device filter was set on K and the distilled water (control) was injected into the device and it was set to zero. K standard solution (5 and 10 ppm) was injected into the device and the range of the device was adjusted according to the standard concentration. The sample was then injected and K concentration was read.

Chloride determination: 25 cc of the sample was poured into the Erlenmeyer flask and 2 ml of potassium chromate was added and titrated with silver nitrate. The titration was continued until the appearance of red brick color, which is the end point of the reaction. The amount of chlorine was calculated from the following formula.

Chlorine ion  $mg/l = ((A-B) \times N \times 35450)/(Ml \text{ sample})$ 

A = Silver nitrate used for the sample

B = Silver nitrate used for the control

N = Silver nitrate normality

Heavy metals measurement

First, all glass tools were washed with 10% nitric acid.

Acidic digestion of the samples: 100 cc of the sample was poured into the Erlenmeyer flask and 5 ml of the concentrated nitric acid was added. The Erlenmeyer flask was covered by a watch glass and placed on the heater until the sample volume reached 10-20 cc. The sample was removed from heater and after cooling, distilled water was added until reaching a volume of 100 cc. The sample was ready for measurement.

The recovery test was performed to check the accuracy. After that, for all eight elements, stock solutions were prepared. A 3-point calibration curve was drawn for each element to perform the quantification of ether concentration in the device. At 3 concentrations of 300, 700, and 1000 μg L<sup>-1</sup>, concentration-absorption curve of each reported. element was The elements concentrations in the samples were determined according to the standard curve and the amount of adsorption related to each element in the unknown sample. Then, in order to reanalyze the samples, all analytical steps were performed. Spike recoveries for the element were from 92.9% to 99.2% based on the recovery test results <sup>26</sup>.

#### Statistical analysis

All information collected from the samples and measurements were extracted using SPSS16 software and the results were plotted and analyzed in graphs and tables. Moreover, water parameters were compared with standard values using one-sample t-test. The results were also compared with standard values in different hospitals using ANOVA and Kruskal-Wallis statistical tests.

#### **Ethical issue**

This article was extracted from a plan approved by the Department of Environmental Health Engineering, School of Public Health, Kashan University of Medical Sciences (Approval code: 98, 096) and the ethical code of the Ethics Committee is IR.KAUMS.NUHEPM.REC.1398. 031

#### **Results**

In this study, microbial and chemical quality of 54 water samples used in dialysis machines in four hospitals of Kashan city were analyzed and compared with AAMI standards. Chemical and microbial properties of water samples are shown in Table 1. Based on the results, water samples used in hospital number 4 (22.48 mg/L CaCO<sub>3</sub>) and hospital number 2 (11.99 mg/L CaCO<sub>3</sub>) had the highest and lowest total water hardness, respectively. Generally, there was no significant difference between these hospitals in terms of water hardness (P-value = 0.13). Also, water sample collected from hospital number 3 showed that the highest Ca hardness and concentrations were 9.26 mg/L CaCO<sub>3</sub> and 3.67 mg/L CaCO<sub>3</sub>, respectively. Hospital number 2 had the lowest Ca hardness (4.52 mg/L CaCO<sub>3</sub>) and Ca concentration was 1.80 mg/L CaCO<sub>3</sub>. Mean Ca hardness and Ca concentration in all the studied hospitals were 7.97 mg/L CaCO<sub>3</sub> and 3.16 mg/L CaCO<sub>3</sub> and there was no significant difference between them (P-value = 0.195). However, a significant difference (P < 0.05) was observed between Ca concentration in water used by hospital number 4 and standard levels. Mean concentration of Ca in 25% of hospitals was within the recommended levels. The highest mean concentration of Mg was observed in hospital number 4 (3.19 mg/L CaCO<sub>3</sub>) and total mean concentration of Mg in all the studied hospitals was 2.76 mg/L CaCO<sub>3</sub>. Comparison of Mg concentration in water used by these hospitals and its standard levels suggested a significant difference except in the case of hospital number 3 (P-value = 0.28). The samples collected from hospital number 2 showed the lowest Mg concentration (1.8 mg/L CaCO<sub>3</sub>). Furthermore, Mg, Na, K, and sulfate concentrations in all the samples were in good agreement with standard levels.

Analysis of collected samples showed that mean concentrations of Na, K, chloride, fluoride, sulfate, nitrate, and nitrite were higher in the samples collected from hospital number 1 compared to others. Nitrate amount in the samples of all hospitals was more than standard levels (Figure 1) .Total nitrite concentration in all the samples was 0.01 mg/L and there was no significant difference between them (P-value = 0.144). In 75% of the hospitals, fluoride concentration was in agreement with standard levels. According to the results, hospitals number 1 and 3 showed the highest (457.11 mg/L) and lowest (34.12 mg/L) amounts of dissolved solids. The total mean of dissolved solids in all the hospitals was 11.47 mg/L and there was a significant difference (P < 0.001) between them. The lowest (pH = 6.87) and highest (pH = 6.83) acidity were observed in samples collected from hospital number 1 and hospital number 3, respectively. Water pH levels in all the samples were very close to each other. The remained chloride amount was equal to zero.

Concentrations of heavy metals in dialysis water samples collected from different hospitals are shown in Table 2. The results revealed that concentrations of lead (Pb), Al (Figure 2), Zn, and silver (Ag) were more than standard levels. Mean concentrations of Cd (0.018 24 mg/L), Al (0.34 24 mg/L), Cu (0.2224 mg/L), Pb (0.224 mg/L), and Zn (0.24 mg/L) in the samples collected from hospital number 2 were higher than others. Cu and Cr in 25% of the hospitals and Cd in 50% of the hospitals were within standard levels. Thallium (Tl) concentration was zero. It should be mentioned that heterotroph bacteria were not observed in none of the water samples.

1/3/

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**Table 1:** Mean and standard deviations of chemical and microbial parameters of dialysis water used in the hospitals compared to AAMI standards

Hospital	<b>T</b> 7 4.	Hospital 1		Hospital 2		Hospital 3		Hospital 4		Total		AAMI
Parameter	Unit	$\overline{x} \pm SD$	P-value									
Total hardness	mg/lcaco3	16.360 ± 7.696	-	$11.997 \pm 7.378$	-	21.124 ± 13.733	-	22.488 ± 13.651	-	19.491 ± 12.380	-	-
Ca hardness	mg/lcaco3	$7.00 \pm 4.546$	-	$4.524 \pm 4.228$	-	$9.268 \pm 5.961$	-	$8.964 \pm 6.256$	-	$7.949 \pm 5.775$	-	-
Calcium	mg/l	$2.801 \pm 1.818$	0.223	$1.807 \pm 1.690$	0.742	$3.678 \pm 2.401$	0.069	$3.574 \pm 2.504$	0.003	$3.168 \pm 2.311$	0.001	2
Magnesium	mg/l	$2.277 \pm 1.650$	0.014	$1.802 \pm 1.201$	0.001	$2.903 \pm 2.842$	0.28	$3.196 \pm 2.393$	0.009	$2.762 \pm 2.225$	0.001	4
Sodium	mg/l	$69.333 \pm 12.093$	0.873	$14.111 \pm 4.935$	0.001	$4.744 \pm 2.971$	0.001	$5.155 \pm 4.117$	0.001	$17.275 \pm 24.47$	0.001	70
Potassium	mg/l	$0.177 \pm 0.120$	0.001	$0.144 \pm 0.292$	0.001	$0.007 \pm 0.009$	0.001	$0.085 \pm 0.150$	0.001	$0.098 \pm 0.170$	0.001	8
chloride	mg/l	$95.222 \pm 51.014$	-	$21.922 \pm 14.590$	-	$23.421 \pm 14.654$	_	$36.804 \pm 51.895$	-	$41.829 \pm 48.993$	-	-
Fluoride	mg/l	$0.204 \pm 0.122$	0.916	$0.195 \pm 0.122$	0.916	$0.165 \pm 0.153$	0.521	$0.161 \pm 0.166$	0.244	$0.175 \pm 0.148$	0.224	0.2
Nitrate	mg/l	$4.088 \pm 1.538$	0.04	$3.333 \pm 1.208$	0.011	$3.411 \pm 0.862$	0.01	$2.811 \pm 1.062$	0.01	$3.211 \pm 1.210$	0.001	2
Nitrite	mg/l	$0.019 \pm 0.006$	-	$0.013 \pm 0.003$	-	$0.010 \pm 0.003$	-	$0.014 \pm 0.010$	-	$19.491 \pm 12.380$	-	-
Sulfate	mg/l	$48.056 \pm 22.337$	00.01	$4.102 \pm 2.713$	0.001	$12.768 \pm 26.104$	0.001	$4.544 \pm 3.616$	0.001	$7.949 \pm 5.775$	-	100
TDS	mg/l	$457.11 \pm 119.96$	-	$75 \pm 11.302$	-	$34.122 \pm 33.582$	-	$40.666 \pm 27.755$	-	$3.168 \pm 2.311$	0.001	-
pH	-	$6.87 \pm 0.13$	-	$6.86 \pm 0.14$	-	$6.83 \pm 0.12$	-	$6.85 \pm 0.13$	-	$2.762 \pm 2.225$	0.001	-
Heterotroph bacteria	Cfu/ml	0	0.001	•	0.001	•	0.001	•	0.001	$17.275 \pm 24.47$	0.001	200

Table 2: Mean and standard deviations of heavy metals concentrations in dialysis water used in the hospitals compared to AAMI standards

Hospital		Hospital 1		Hospital 2		Hospital 3		Hospital 4		Total		AAMI
Parameter	Unit	$\overline{x} \pm SD$	P- value	standard								
Aluminum	mg/l	$0.244 \pm 0.134$	0.01	$0.343 \pm 0.175$	0.001	$0.308 \pm 0.107$	0.001	$0.238 \pm 0.187$	0.001	$0.268 \pm 0.167$	0.001	0.01
Chromium	mg/l	$0.891 \pm 2.665$	0.352	$0.006 \pm 0.002$	0.001	$0.005 \pm 0.002$	0.001	$0.005 \pm 0.003$	0.001	$0.153 \pm 1.087$	0.352	0.014
Cadmium	mg/l	$0.001 \pm 0.002$	0.813	$0.018 \pm 0.034$	0.181	$0.003 \pm 0.007$	0.305	$0 \pm 0.002$	0.726	$0.004 \pm 0.015$	0.126	0.026
Copper	mg/l	$0.115 \pm 0.035$	0.225	$0.227 \pm 0.038$	0.001	$0.142 \pm 0.036$	0.09	$0.097 \pm 0.017$	0.404	$0.129 \pm 0.54$	0.001	0. 1
Silver	mg/l	$0.539 \pm 0.439$	0.006	$0.208 \pm 0.096$	0.001	$1.315 \pm 1.643$	0.044	$0.369 \pm 0.566$	0.003	$0.528 \pm 0.855$	0.001	0.005
Lead	mg/l	$0.053 \pm 0.019$	0.001	$0.204 \pm 0.318$	0.096	$0.060 \pm 0.045$	0.006	$0.057 \pm 0.025$	0.001	$0.081 \pm 0.138$	0.001	0.005
Zinc	mg/l	$0.046 \pm 0.059$	0.106	$0.249 \pm 0.282$	0.035	$0.167 \pm 0.213$	0.058	$0.243 \pm 0.997$	0.236	$0.198 \pm 0.716$	0.058	0. 1
Thallium	mg/l	0	0.001	0	0.001	0	0.001	0	0.001	0	0.001	0.002

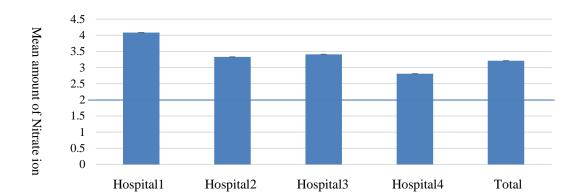


Figure 1: Mean concentration of nitrate ion in water samples collected from the hospitals

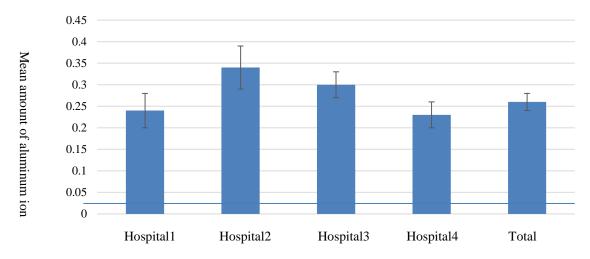


Figure 2: Mean concentration of Al ion in water samples collected from the hospitals

#### Discussion

Today, chronic renal failure (CRF) is an important problem in public health across the world<sup>23, 27</sup>, which is increasing constantly<sup>28</sup>. Each patient needs a large volume of water during each treatment session<sup>29</sup> and usually three dialysis sessions per week is necessary. Therefore, control of water quality is crucial to prevent the risks of side effects on patients<sup>30-32</sup>. Non-compliance of water quality with local and international standards can lead to infectious diseases, pyogenic reactions, and mortality in these patients<sup>33</sup>. Therefore, the current study was carried out to analyze the chemical and microbial quality of dialysis water in 4 hospitals in Kashan city. So far, many studies have been conducted worldwide to evaluate the quality of water used in dialysis. The results of many of these studies indicate water contamination used in hemodialysis centers<sup>19, 22, 34</sup>. The results of the present study showed that the amount of Ca ions in water samples of 75% of the studied hospitals was more than the allowable limit. Ca is an element that is not inherently toxic, but its excessive amounts for kidney patients can cause complications, such as sudden hyperkalemia (hardwater syndrome) with high blood pressure, headache, vomiting, and lethargy<sup>35-37</sup>. A study conducted in West Germany showed that about 17.8% of the tested hemodialysis water samples lacked the necessary restrictions and standards<sup>38</sup>.

Shahryari et al. conducted a study on 30 samples of dialysis machines input water of 5 hospitals in Isfahan province. They concluded that the concentration of chemicals (Cu, Zn, sulfate, fluoride, chloramine, and free chlorine) did not exceed the concentration recommended by the

1740

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AAMI. Moreover, Pb, nitrate, Al, and Ca were not detected in the samples<sup>7</sup>. The reason for the high Ca in the present study is that the water entering the equipment was groundwater. However, in similar studies, which mostly lacked Ca or had the least amount of Ca, the RO device input water was treated and was from surface sources undergoing a treatment process. On the other hand, among the four studied hospitals, hospitals number 1 and 2 used groundwater and hospitals 3 and 4 used urban water distribution network. Ca ion concentration was higher in water samples from hospitals 1 and 2 compared to 3 and 4. It could be due to differences in hospital water supply wells and water distribution network, in terms of geology, the weakness in the municipal water treatment system or Ca deposition in the distribution network. The results also showed that nitrate concentration in the studied water samples was more than the allowable limit. Abbaszadeh et al. also studied the microbial and chemical quality of dialysis water in East Azerbaijan hospitals. They concluded that except for Ca, Mg, fluoride, and nitrate, concentrations of other cations and anions in 100% of the samples were evaluated below the European Pharmacopoeia (Eu.Ph) standard<sup>39</sup>. Rall et al. studied distilled and dialysis water samples collected from dialysis centers. They reported that the concentrations of fluoride, nitrate, and sulfate in the sample were within allowable limits 40 ,which could be related to the defects in the water treatment system of the studied hospitals. Based on several reports, presence of large amounts of nitrate can cause methemoglobinemia<sup>41, 43</sup>. The amount of Na ion detected in the studied samples was within the allowable limit. In a study conducted by Braimoh et al. in Nigeria, concentrations of chemical pollutants, such as Al, Cu, Zn, Mg, Ca, Na, K, fluoride, sulfate, nitrate, chloramine, and free chloride were significantly more than AAMI standards<sup>44</sup>. The results of these two studies were not in line with the study by Shahryari et al.

In addition, the presence of heavy metals in dialysis water can cause irreversible consequences for hemodialysis patients. The results of the present study showed that the concentrations of Pb, Al, Zn, and Ag in all the samples were more than standard levels. Furthermore, based on findings of the current study, mean concentrations of Cd, Cu, Pb, and Zn in the samples collected from hospital number 2 were more than other hospitals and standard levels. It could be attributed to the deficient water treatment system, inadequate washing of dialysis systems after disinfection or newly installed dialysis systems. Suzuki et al. indicated that Al concentration was more than allowable limits<sup>25</sup>. A study conducted in Iraq by Humudat et al. showed that the concentration of Al in 75% of the samples was higher than the standard, which is consistent with the results of the present study<sup>45</sup>. The presence of Al in drinking water is mainly due to the use of coagulant salts during the water treatment process. Excessive amounts of Al in dialysis solutions lead to the accumulation of this element in the body, which gradually results in dementia, bone weakness, and anemia<sup>36</sup>. Abualhasan et al. studied the quality of dialysis water in Palestine. They showed that the Pb concentration in dialysis water in all samples were more than standard levels, raising concerns for the researchers<sup>46</sup>, which is in line with the results of the present study. Worn out hospital water pipes can be a possible reason for this issue. Exposure to high amounts of metals, such as Al, Cd, Cu, Pb, and Zn may cause side effects, such as anemia, nausea, vomiting, neurological disorders, 14, 47 pain<sup>9,</sup> bone Microbiological and contamination can be considered as the main cause of complications and mortality in hemodialysis patients<sup>48, 49</sup>. In the 54 samples collected from 4 hospitals, no microbial contamination with heterotrophic bacteria was observed. A study by Hilinski et al. on the quality of dialysis water in dialysis centers of São Paulo state showed that the bacterial contamination was 54.8% 50. Studies in Morocco, Nigeria, and Iraq showed large microbial contaminations<sup>2, 19, 31, 51</sup>. However, Totaro et al., in their study, showed that 78% of dialysis water samples collected from 9 hospitals in Italy (2015-2016) did not show any microbial contamination<sup>48</sup>. Moreover, in dialysis water samples collected from

5 dialysis centers in Iran no bacterial contamination was observed<sup>7</sup>, which is consistent with the results of the present study and pH neutrality can be considered as a possible reason for this observation<sup>39</sup>.

One of the most important advantages of the present study was the ability to measure the differences of the monitored parameters in relation to the quality of dialysis water in hospitals with international standards. Heavy metals measurement in dialysis water was evaluated for the first time in the study area. This study draws the attention of hospital managers to monitor the quality control of water treatment system performance and the need to comply with international standards and the importance of continuous heavy metal measurement. On the other hand, the lack of sufficient budget and time to continue the present study, prevented the continuous review of the study. Therefore, planning for continuous monitoring is required.

#### **Conclusion**

Given the importance of dialysis water quality, the results of the study indicated high concentration of heavy metals in the dialysis machines input water, the complications of which can affect the health of dialysis patients. Therefore, periodic monitoring of water treatment systems and measurement of heavy metals at regular intervals and regular replacement of reverse osmosis filters are essential.

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#### **Conflict of interests**

The authors declare that they have no conflict of interest.

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