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ORIGINAL ARTICLE

Heavy Metals Contamination and Distribution in Drinking Water from Urban Area of Mashhad City in Northeast Iran: Implications for Water Quality Assessment

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KEYWORDS

Heavy metals;
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ABSTRACT: The fast industrialization and urbanization in the world have led to increasing heavy metal pollution in the water supplies. Here, we examined the spatial and temporal distribution and contamination of heavy metals and physicochemical parameters in the drinking water sources of Mashhad in Iran. In this survey, 432 samples of drinking water were collected from 5 zones and 36 stations from August 2017 to May 2018. The results of heavy metal measurements showed that the average concentrations of arsenic (As), mercury (Hg), chrome (Cr), nickel (Ni) and Pb were 0.198±0.11, 0.018±0.04, 5.80±7.87, 1.695±2.16 and 0.574± 0.22 µg L^{-1} for arsenic (As), mercury (Hg), chrome (Cr), nickel (Ni) and Pb, respectively. This result showed that the concentration of heavy metals and physicochemical parameters were comparatively lower than the threshold values throughout the study period; however, some stations showed metrics values above the acceptable limit. Thus, there is still potential contamination in drinking water due to potential heavy metal interactions and long-term exposure. Results of this study showed the current pollution status of drinking water in Mashhad needs remediation efforts to protect human health in urban regions, which highlighted a basis for decision-making in the future to take the main action on contamination control.

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INTRODUCTION

Rapid industrialization and urbanization have resulted in the enhancing discharge of chemical contaminants into water sources [1, 2]. Drinking-water supply is essential and important for human life and its quality is under constant pressure [3, 4]. More than 2 billion people live in countries with limited water resource, and about 4 billion people experience water scarcity during at least one month per year [5, 6]. The use of contaminated water seriously threats human health [7, 8]. In 2015, around 2.1 billion people have used unsafe drinking water. Annually, 842000 people die in the world due to the use of contaminated water[9]. Water quality can be compromised by the presence of radiological hazards, toxic chemicals, and infectious agents. One of the main pollutants of drinking water is heavy metals, which are the common pollution on seven continents including Asia [10, 11]. Epidemiological studies suggest that exposure to heavy metals is a serious concern due to the nature of their non-degradability [12]. Heavy metals pollution poses a serious a threat to human health due to their bioaccumulation, carcinogenicity, toxicity, and stability in the environmental media such as water, sediment, and soil [13, 14]. In recent decades, human activities such as agricultural and industrial activities have led to an increase in heavy metals in the environment and in water[15]. Using geographic information system (GIS) for geographic analysis can better understand large-scale pollution paths, and has a higher ability to investigate the spatial distribution and division of pollutants to assess the source [16]. Although some heavy metals such as iron, copper, and nickel are required for biological systems, at high concentrations, they can be toxic to living organisms. Some other heavy metals such as cadmium, arsenic, mercury, and lead are toxic to humans and living organisms [17] and cause various health effects, such as various disorders of the body, cancer, and death [12, 18].

Arsenic has been classified as one of the carcinogenic compounds by the International Agency for Research on Cancer [19, 20]. The widespread toxicity caused by the consumption of arsenic-contaminated water in Argentina, Chile, and Bangladesh has been reported [21]. Mercury is one of the highly toxic pollutants in the environment [22] with high bio-magnification [23]. This element enters the environment through the mining of mercury, paper mills, plastics, the use of fungicides, and industrial wastewater [22, 24]. The mercury affects the central nervous system, brain, liver, kidneys, and kidney and cause death [21, 23, 25, 26]. Chromium is a very toxic and mutagenic element. In the classification of the International Agency for Research on Cancer, it has been classified as a carcinogenic group [23, 27]. This element can damage the kidneys, skin, neurons, liver, respiratory and cardiovascular systems [26]. High levels of nickel are toxic to the human body and can cause respiratory cancer, osteoporosis, cardiovascular, kidney and thyroid diseases [25, 28]. Lead belongs to the carcinogenic group B2 of the International Agency for Research on Cancer [29, 30]. This element can cause renal failure, seizure, nervous system disorders, high blood pressure, and damage to the reproductive, cardiovascular and nervous system. Also, lead is one of the primary threats for the children health and reduces their intelligence quotient (IQ), behavioral disorder, anemia, and nervous stresses [21, 25].

In Iran, artificial reservoirs, well, rivers, and lakes are the main sources to supply drinking water. In recent years, there has been accidental leakage and illegal emission of wastewater containing chemical pollutants into water supply source. Likewise, developing countries, like as Iran, are at risk of contamination with heavy metals due to the lack of proper environmental standards in the disposal of industrial and urban waste [12, 31, 32]. Therefore, assessing the quality of water sources is necessary because it is closely associated to the human health. Mashhad is the second-largest city in Iran. [33]. Mashhad has experienced rapid industrial and commercial development, which has led to increase in population, resulting deterioration and contamination of freshwater-supply quality. Deterioration of water quality in this city drew national attention [31, 32]. Heavy metals contaminations were frequently reported in water source of Mashhad. However, their concentrations were region specific. Therefore, specific focus need given to the pollution associated with heavy metals in drinking water source. Heavy metals pollutions are region specific, and the study on sources of water must be conducted on a case-by-case basis. Therefore, regular monitoring of these elements in drinking water in this city is very important to support human health [32, 34]. Furthermore, the current information on the concentration and distribution of heavy metals in the drinking water of Mashhad is inadequate. Since, no updated background available on contamination of toxic heavy metals in drinking water in Mashhad, this study is the first study on heavy metal pollution, in which we provide comprehensive information about heavy metals' distribution and contamination pattern in this region. Likewise, given that Mashhad is one of the metropolitan city in Iran and there are great industrial and tourism centers in this city [27], more attention should be taken to assess water quality sources and municipal water distribution system. The purpose of this study was to evaluate the spatial and temporal distribution and contamination of heavy metals (As, Hg, Cr, Ni, and Pb) and physicochemical parameters (electrical conductivity, pH, cholera, calcium, and Magnesium) in drinking water of Mashhad. Results of this study can establish as data base for decision-making in future to conduct essential action on controlling water pollution to maintain the safety and human health.

MATERIALS AND METHODS

Sampling

According to the location and land use, the city of Mashhad was divided into 5 zones (north, south, east, west, and center zones) and 36 stations for sampling drinking water (Figure 1, Table 1). In this survey, 432 samples of drinking water were collected from different areas from August 2017 to May 2018. We used the factorial method to measure the sample size based on 4 seasons (industrial and traditional), 36 stations, and 3 repetitions. The cluster sampling methods were used to divide the whole city into 5 clusters (based on municipal zoning). Then, 7 stations were randomly selected from each region, and drinking water samples were collected from each station in 3 repetitions to study the concentration of heavy metals.

Each point was sampled two times in the dry season (August and September 2017) and wet season (March and April 2017) to examine Seasonal differences. Several studies reported that seasonal differences are attributed to the concentration of pollution-sensitive groups.

The electrical conductivity (EC) and pH of all samples were locally measured l by EC meter (WTW 3310) and portable pH meter (WTW 3310), respectively. All plastic bottles were washed twice with distilled water and nitric acid (HNO_3, 20%). We used GF/F Whatman (0.45 µm Millipore size) to filter all samples and 3 ml of nitric acid (HNO_3, 69%) was also added to all samples to prevent the crystallization and adsorption of metals prior to laboratory analysis. The water samples were then transferred to the laboratory by cold and dark containers and stored in a refrigerator at a temperature of 4ºC [35].

Table 1. Temporal variation of the physicochemical parameters and heavy metals in drinking water during sampling periods

Seasons	EC (µS cm^{-1})	Mg (mg L^{-1})	Ca (mg L^{-1})	Cl (mg L^{-1})	As (µg L^{-1})	Hg (µg L^{-1})	Cr (µg L^{-1})	Pb (µg L^{-1})	Ni (µg L^{-1})
Spring	930.39	32.3	63.22	78.79	0.183	0.022	3.383	0.685	2.172
Summer	911.2	29.37	59.17	81.5	0.214	0.003	7.733	0.462	1.217
Average	924	30.84	61.2	80.12	0.198	0.018	5.808	0.574	1.695
Standard Division	227.08	8.57	11.34	34.86	0.11	0.04	7.87	0.22	2.16
*** Asym Sig**	0.514	0.16	0.128	0.973	0.982	0.032	0.082	0.063	0.031

The mean and standard deviation (±) concentrations; * Asymptotic significant at $p < 0.05$ based on Kruskal–Wallis test and testing significant change on the temporal variation of physicochemical parameters and heavy metal in drinking water from 35 stations

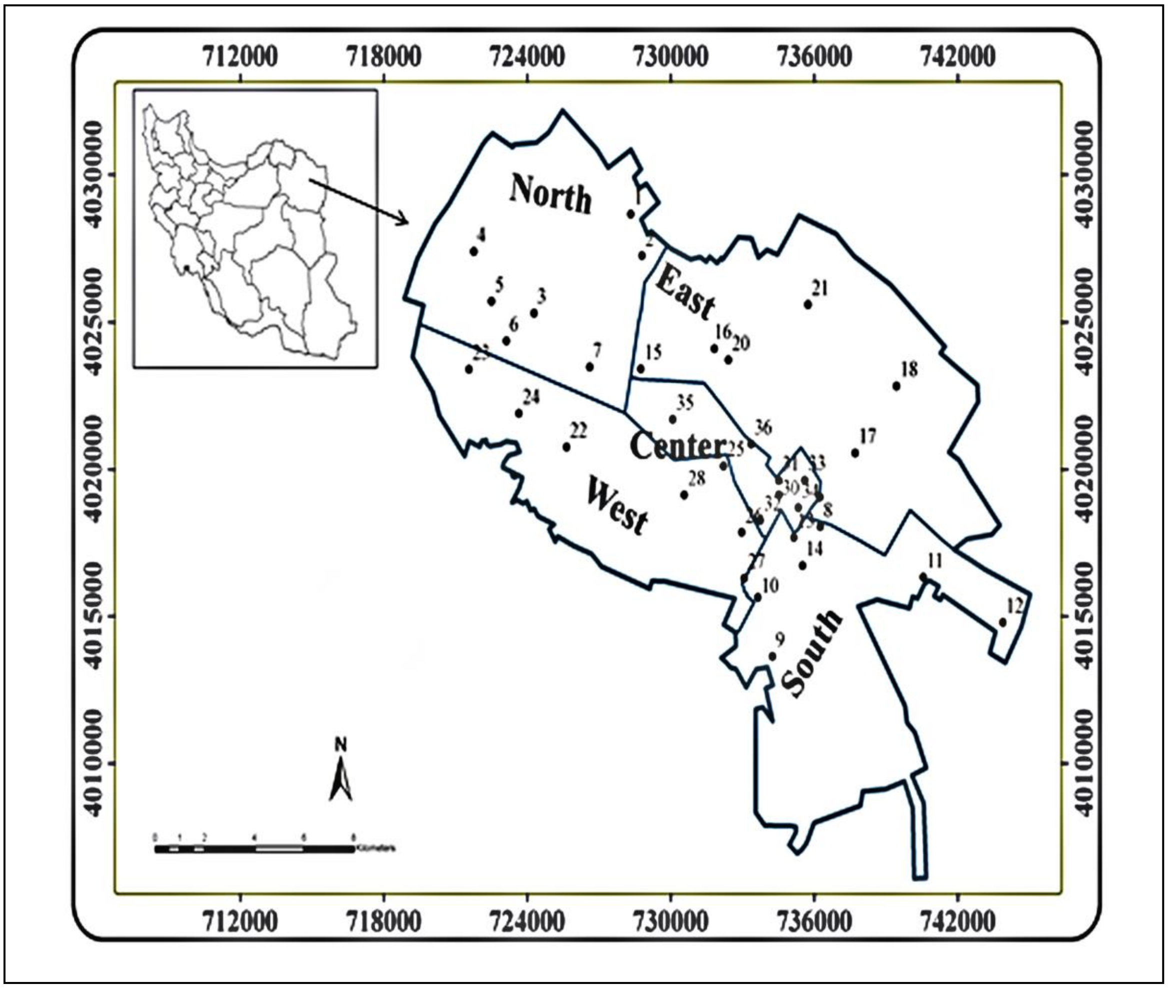

Figure 1. Location of Sampling Stations and Zones in Mashhad, Iran

Chemical analyzes

All the standard solutions, reagents, and acids were purchased from Merck (Darmstadt, Germany). Mercury, As, Cr, Pb and, Ni were measured according to the EPA methods (6020) by the Plasma Mass Spectrometer (ICP-MS, 7700 series). Standard solutions were provided based on concentrations of metal ions that were reported in previous studies and the limit of detection (LOD) of the device. The correlation coefficient of the calibration curves for all elements was more than 99%.

Quality control and assurance

In this study, we acid washed all containers and bottles in diluted HNO_3 for 24 h and rinsed with deionized water. The quality control of heavy metal analysis was performed using a metal solution of a specified concentration, blank and three sample replicates. Considering the high level of confidence (95%), for interpretation, the average concentration of each sample was used. The LOD was 0.04, 0.038, 0.09, 0.11, and 0.22 µg L^{-1} for As, Cr, Hg, Pb, and Ni, respectively. The heavy metals concentration was reported in micrograms per liter (µg L^{-1}) and physical physicochemical parameters were reported in milligram per liter (mg L^{-1}). Standard reference solutions (spiked solation) and certified reference materials (CRMs) were used as control samples to check the quality and accuracy of the results for the measurements of heavy metals. Recovery ratios for each element were in an acceptable range (85.7-115%), which is acceptable range based on EPA.

Statistical analysis

Statistical software including SPSS 17 (SPSS, Chicago, IL) and Excel 2007 (Micro-soft Office) were used to calculate descriptive statistics (frequency, mean, and standard deviation). Likewise, Kruskal-Wallis Test was used for the analysis of variables in different regions. Bivariate tests including analysis of variance (comparison between parameter changes in time and spatial interval) and Pearson correlation coefficient were used to measure the significant variation in different groups. To clarify the level of contamination, the concentration of heavy metals in all samples was compared to the national standard of Iran, the World Health Organization (WHO), and the United State Environmental Protection Agency (US EPA). Distribution maps for each element were also plotted by Arc-GIS v.10.2.3.

RESULTS

Temporal and spatial distribution

The average concentrations of heavy metals in drinking water in the different stations and seasons have been summarized in Tables 1 and 2. Our results showed that changes in As, Cr and Pb are more than 0.05. Consequently, there is no significant difference between the mean of these variables in the temporal scales (spring and summer seasons). However, there are significant differences in average Hg and Ni in spring and summer. In addition, there is no significant difference in the average change of physicochemical parameters on the time scale (Table 2). The average concentration of As was 0.198 µg L^{-1} and it was ranged from 0.089 to 0.728 µg L^{-1}. The distribution map of As showed that the highest concentration of arsenic was observed at stations 4, 6, 18 and 23, while the lowest concentration was at stations 1-3, 9, 15, 17, and 35 (Figure 1). The average concentration of Hg was 0.018 µg L^{-1} and it was ranged from 0 to 0.15 µg L^{-1}. According to the distribution map, the highest concentration of Hg has been observed at stations 5, 6, 8, 22, 32, and 34. The average Cr concentration was 5.808 µg L^{-1} and it ranged from 0.315 to 38.3 µg L^{-1}. The highest concentration of Cr was observed in stations 7, 17, and 34, and the lowest concentration was in stations 1-3, 5, 9 15, 18, 20, 22, 25, and 32 (Figure 1). The average concentration of Ni was 1.695 µg L^{-1} and it ranged from 0.185 to 13.145 µg L^{-1}. The highest Ni concentration has been observed at stations 23 and 3. The average concentration of Pb was 0.574 µg l^{-1} and it varied from 0.213 to 21.23 µg L^{-1}. The distribution map showed that the highest concentration of Pb was observed at stations 1, 24, 23, 33, and 34, and the lowest concentrations were observed at stations 1, 7, 11, 12, 13, 15, 17, and 24 (Figure 2).

In this study, pH ranged from 7 to 8.16, and the average of EC was 924±227.08 micrsiemens per centimeter (µS cm^{-1}) and it ranged from 544 to 1500 µS cm^{-1}. The average concentration of Ca, Mg, and Cl were 61.2±11.34, 30.84 ±8.57, and 80.12±34.86 mg L^{-1}, respectively. The distribution of EC, Ca, Mg, and Cl exhibited a homogeneous pattern that decreased from the south to north direction (Figure 3).

Table 2. Spatial variation of the physicochemical parameters and heavy metals in drinking water during sampling periods

Zones	Stations	EC (µS cm^{-1})	Cl (mg L^{-1})	Ca (mg L^{-1})	Mg (mg L^{-1})	AS (µg L^{-1})	Hg (µg L^{-1})	Cr (µg L^{-1})	Ni (µg L^{-1})	Pb (µg L^{-1})
North	1	554	30.18	41	28.2	0.15	Nd*	0.42	1.22	0.83
	2	544	23.08	42	27	0.14	Nd	0.57	0.90	0.42
	3	634	23.08	56	27	0.12	Nd	0.97	3.20	0.71
	4	604	42.6	46	16.2	0.27	Nd	6.03	1.23	1.07
	5	638	30.18	52	31.2	0.15	Nd	2.73	1.56	0.49
	6	625	31.95	63	18	0.25	Nd	3.65	1.89	0.71
	7	608	37.28	60	16.2	0.20	Nd	17/67	0.45	0.37
South	8	1026	90.53	59	35.4	0.17	Nd	0.36	0.80	0.36
	9	1088	134.9	63	41.4	0.09	Nd	0.54	0.40	0.53
	10	802	83.43	52	22.2	0.20	Nd	3.83	1.09	0.49
	11	1161	117.15	56	35.4	0.21	Nd	8.25	0.19	0.39
	12	1020	97.63	58	36	0.18	Nd	8.27	1.27	0.31
	13	1500	122.48	75	42	0.14	Nd	6.41	1.31	0.59
	14	1018	111.83	69	37.8	0.18	Nd	10.21	0.36	0.40
East	15	836	74.55	50	31.8	0.10	Nd	0.68	0.28	0.33
	16	1174	122.48	71	36	0.19	Nd	11.17	0.23	0.52
	17	1017	99.4	56	40.8	0.12	Nd	23.25	1.72	0.38
	18	1006	101.18	49	40.8	0.28	Nd	0.47	1.82	0.63
	19	1107	110.05	63	32.4	0.21	Nd	0.57	0.35	0.50
	20	1089	110.05	77	34.8	0.15	Nd	0.59	0.58	0.56
	21	887	74.55	74	24	0.16	Nd	4.16	0.86	0.75
West	22	1065	180.28	64	38.4	0.16	0.2	2.42	1.72	0.54
	23	631	30.18	61	21	0.39	Nd	6.34	13.15	1.33
	24	630	30.18	62	23.4	0.18	Nd	7.51	0.40	0.31
	25	938	85.2	63	25.2	0.18	Nd	0.32	3.05	0.63
	26	866	74.55	57	27.6	0.23	Nd	1.19	2.96	0.53
	27	808	65.68	54	25.8	0.24	Nd	4.46	1.41	0.64
	28	946	51.48	66	31.2	0.20	Nd	5.56	2.47	0.75
Center	29	1059	92.3	73	29.4	0.23	Nd	4.08	1.49	0.51
	30	923	79.88	65	30.6	0.20	Nd	2.53	1.15	0.54
	31	1068	108.28	66	34.2	0.14	Nd	1.59	1.52	0.56
	32	1075	104.73	68	36.6	0.16	0.2	7.88	0.95	0.58
	33	1070	108.28	72	33	0.13	Nd	1.44	1.59	0.45
	34	1065	101.18	66	39	0.20	0.1	19.50	3.63	0.81
	35	933	83.43	64	26.4	0.14	Nd	0.40	1.19	0.64
	36	1037	92.3	70	33.6	0.19	Nd	1.85	2.69	0.70
Average		921	80.12	61.2	30.83	0.198	0.018	5.808	1.695	0.574
Standard Division		222.69	23.35	9.06	7.11	0.11	0.04	7.87	2.16	0.22
Asym Sig		<0.001	<0.001	0.002	0.001	0.01	<0.001	0.02	<0.001	0.03
National Standard		1500-2000	250-400	300	30	10	6	50	70	10
EPA Standard		-	250	200	30	10	2	100	-	15
WHO Guideline		-	200-250	-	-	10	6	50	70	10

Nd: not detectable; * Asymptotic significant at $p < 0.05$ based on Kruskal–Wallis test

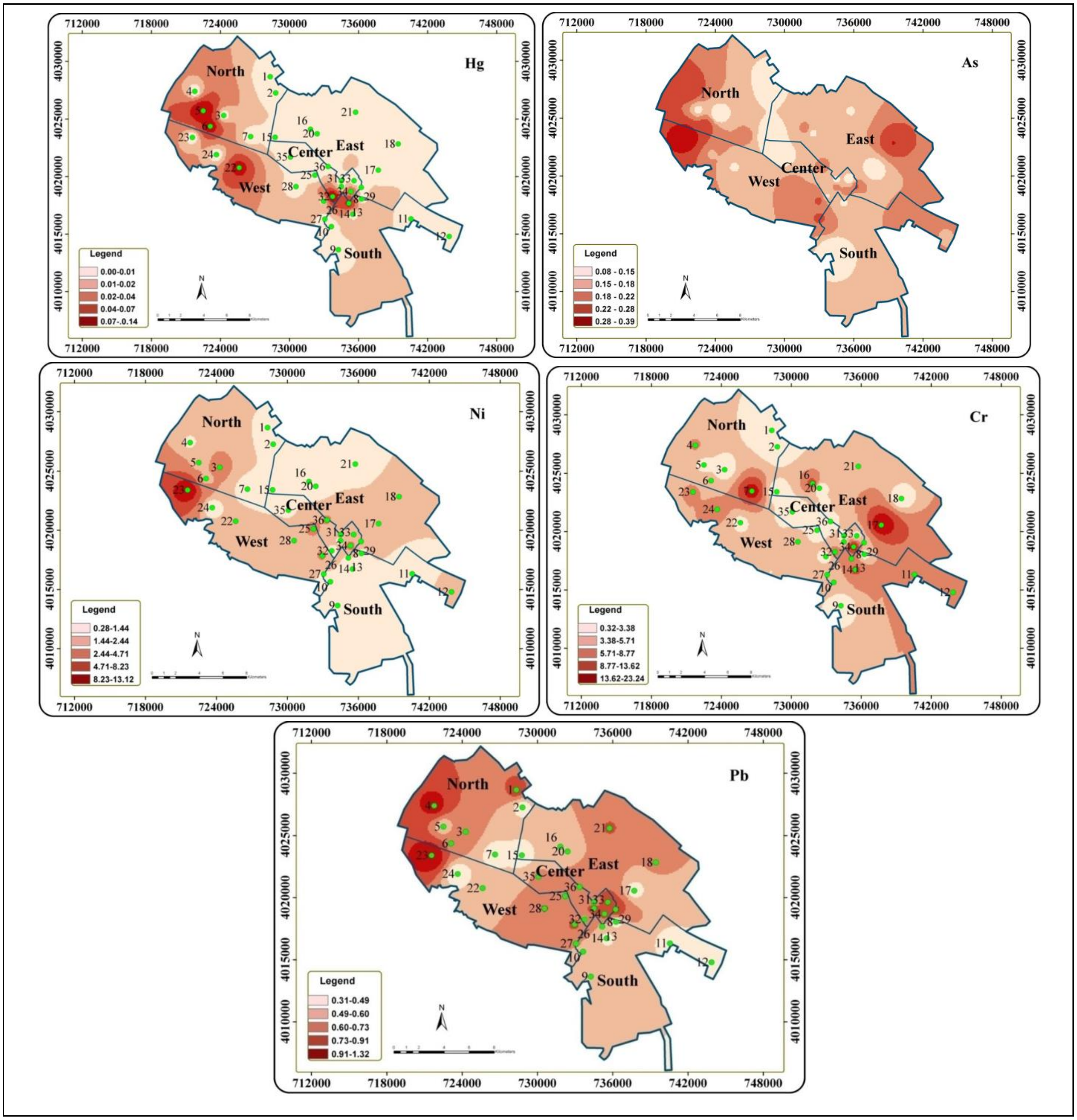

Figure 2. Spatial distribution of heavy metals (µg L^{-1}) in Drinking water of Mashhad, Mashhad

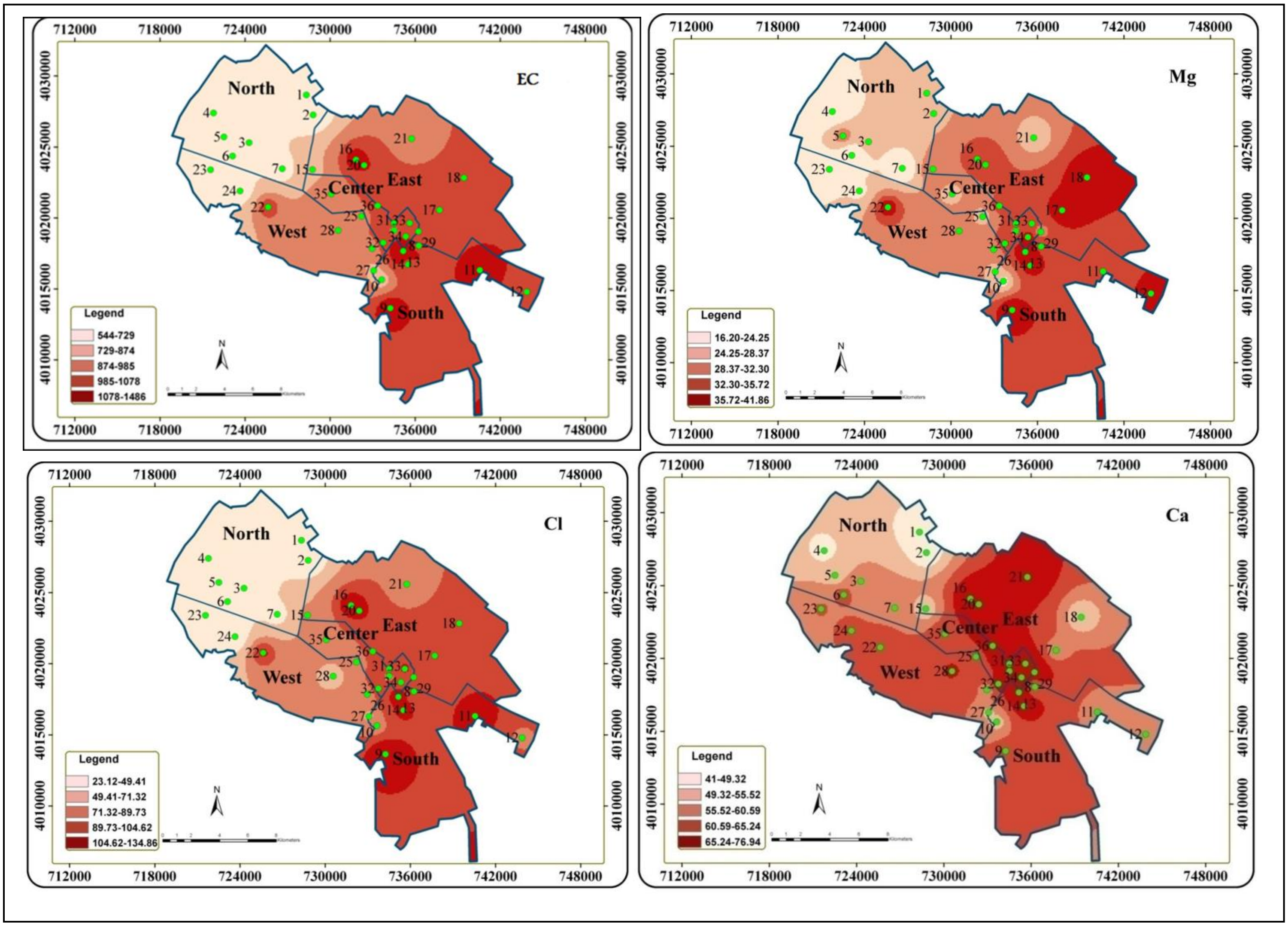

Figure 3. Spatial distribution of physicochemical parameters in drinking water of Mashhad, Mashhad

Contamination assessment

The average concentration of As, Hg, Cr, Ni, and Pb in all sampling stations were lower than the thresholds of guidelines such as WHO, USEPA, and Iranian national standard (Figure 4, 5). The EC level was lower than the Maximum Acceptable Concentration (MAC) in all stations during sampling periods (except stations 13 and 14), while the concentration of Mg was higher than MAC level in most stations (Figure 6). The enrichment value of Ca and Cl were also lower than MAC level in all stations during sampling periods (Figure 6).

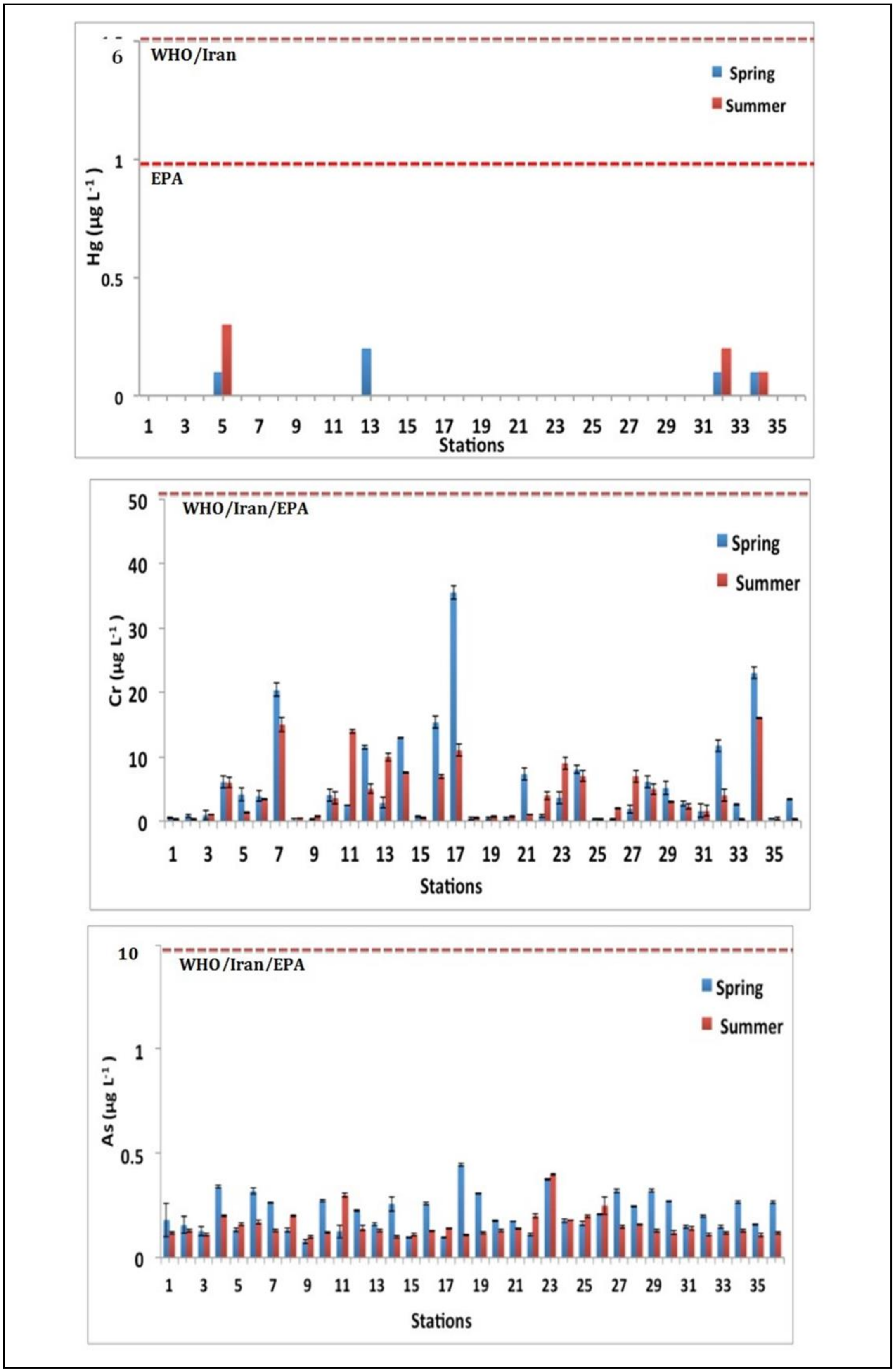

Figure 4. Comparison of heavy metals concentrations in drinking water of Mashhad with thresholds limit

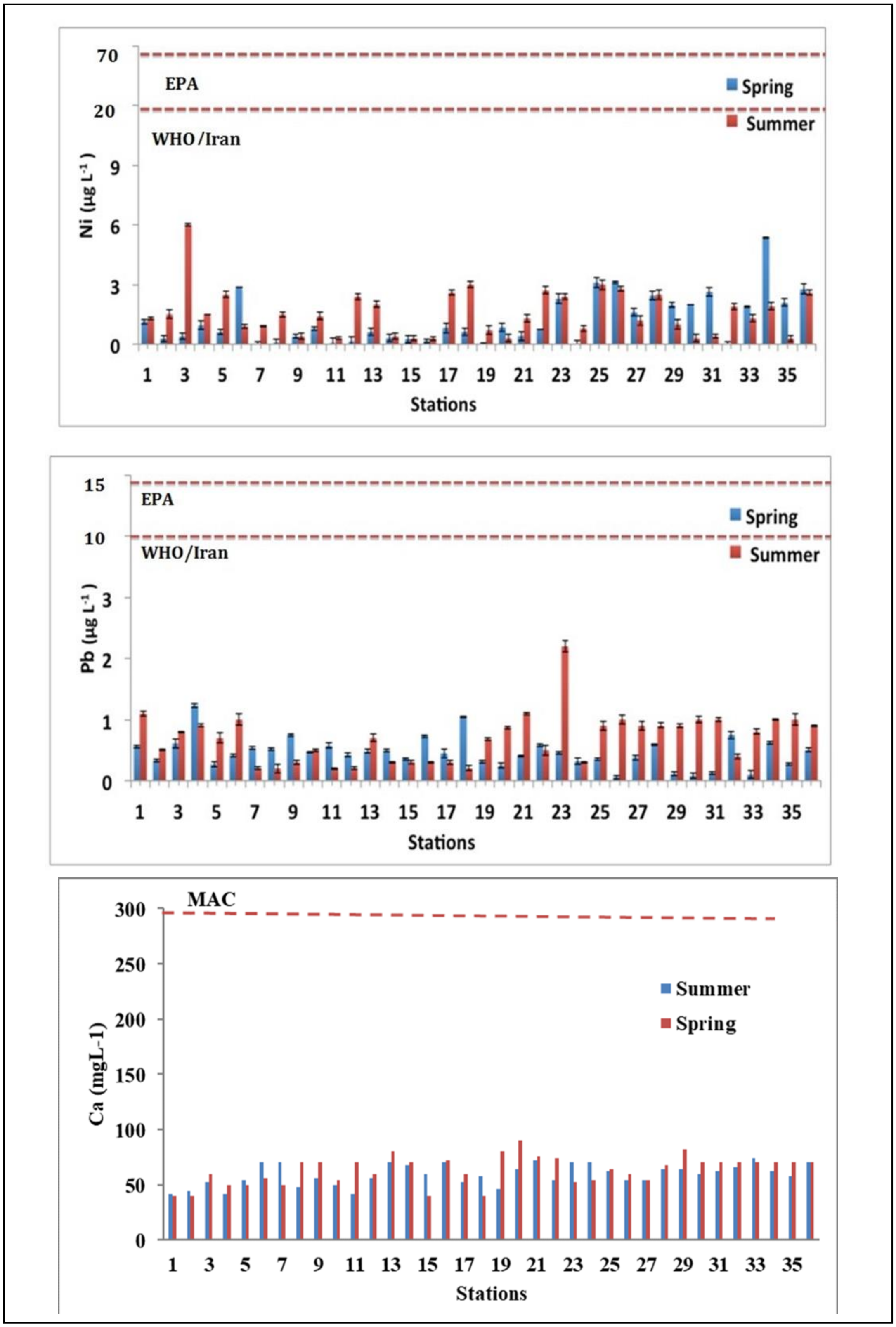

Figure 5. Comparison of heavy metals and physicochemical parameters in drinking water of Mashhad with standard Maximum Acceptable Concentration (MAC); Maximum Permissible Concentration (MPC)

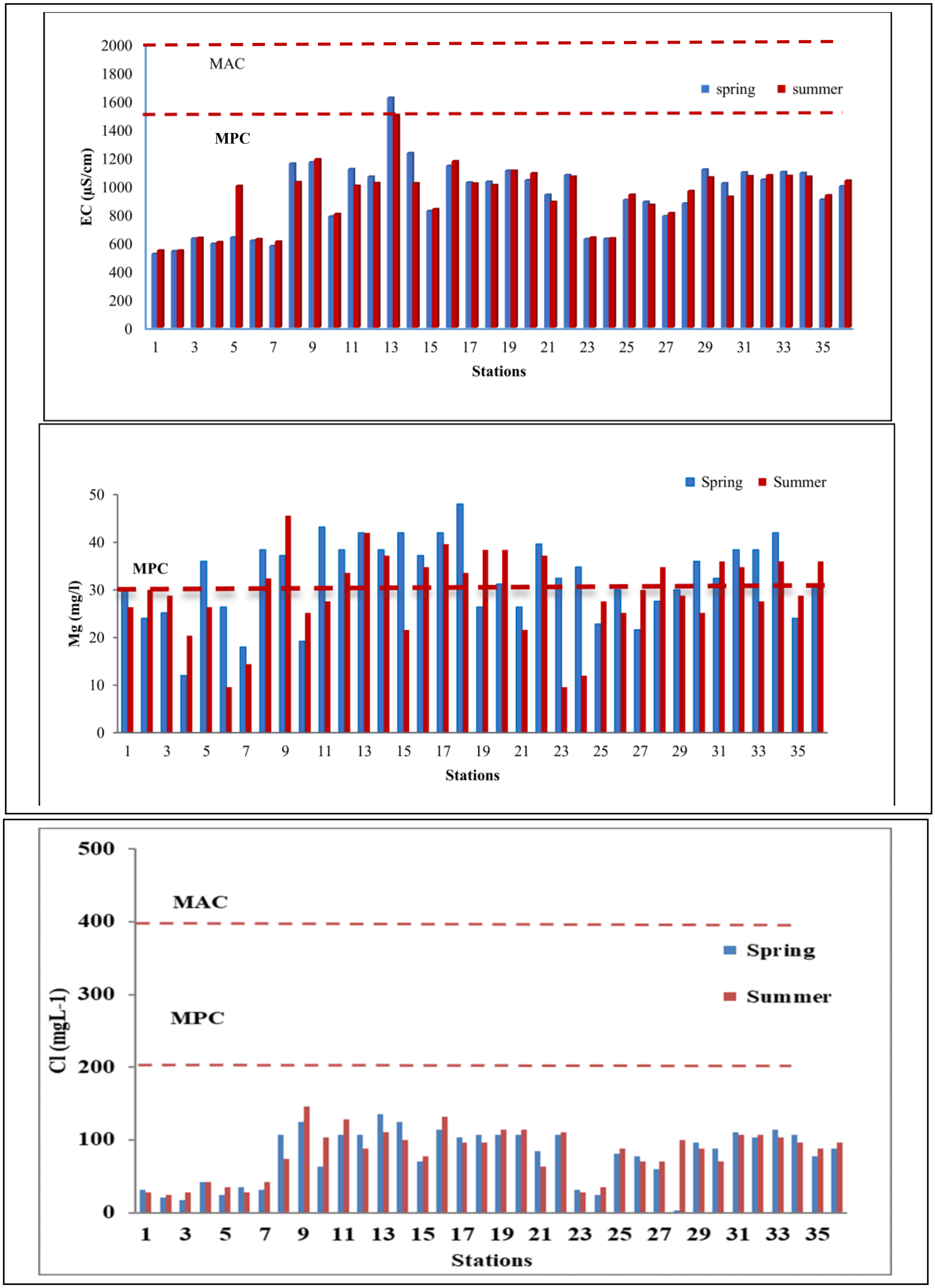

Figure 6. Comparison of physicochemical parameters in drinking water of Mashhad with standard Maximum Acceptable Concentration (MAC); Maximum Permissible Concentration (MPC)

DISCUSSION

In this study, toxic heavy metals and some physicochemical parameters in drinking water have been estimated to evaluate contamination level and distribution. There was a wide variation in the concentration of heavy metals and physicochemical parameters in drinking water either at stations or in sites. This is probably due to differences in water supplies in each study area, poor purification system, pipeline corrosion, and different geological texture, and composition [36, 37]. The highest concentrations of Pb, Ni, As, and Hg was measured in North zone, at the 4 and 6 stations, and in West zone, at the 22 and 23 stations. The drinking water from West cite originated from wells and groundwater. According to the geochemical studies, two different sources of heavy metals cover this region that includes the acidic rocks as the origin of As, Cd, and Cu, as well as Ophiolite rocks as the origin of Ni, V, Fe, and Pb. Therefore, these natural metals from acidic and Ophiolite rocks could be released into groundwater sources, and contaminates drinking water supplies in this region. Thus, the geochemical texture could be considered as the main route of heavy metals contamination in this region.

The highest concentrations of Pb, Ni, As, and Hg was also observed in the North zone (4, and 6 stations). Drinking water at these stations has been supplied through groundwater resources resource, which mainly distributed to the distribution network without refining and regular monitoring heavy metals. This may influence the quality of drinking water in this region [31, 32]. Thus, it is essential to use suitable sanitation improvement programs to protect the health of the residents in these regions.

The highest Cr levels in drinking water samples were observed in the East site (12, 16, and 20 stations), North site (7 station), and Center site (32 and 34). Likewise, the highest concentration of Pb was observed at the 33, 34 and 29 stations. It was evidenced that pipeline transports in these stations were old and processes of water transporting pipelines have corrosion problems. This process may increase concentration Pb and Cr in the distribution network of drinking water and the consequence of badly conditioned water [38]. This result also consistence with studies that was conducted in other metropolitan cities in Iran such as Tehran [39], Mashhad[40] and Ahvaz [37]. According to EPA and the American Water Association, large amounts of leakage of heavy metals into drinking water occur through the distribution network and house plumbing such as lead pipes, tin, valves and brass fittings, copper tubes, etc.[41, 42]. According to reports from the US Water Board, distribution network has a contribution of 29% in water pollution[43]. Application of chemicals in water treatment, leakage of these elements through water pipes, corrosion of pipes, inappropriate storage containers, and poor filtration can lead to the entry of heavy elements from the distribution network into water[37, 44]. Furthermore, the high concentration of As, Cr and Pb in East and South sites, (at 11, 12, 17, 18, and 21 stations) might be related to the water quality of Doosti dam (at the border of Turkmenistan and Iran) because the drinking water from South and East regions originated from this dam. According to recent studies, several contaminants such as municipal effluents, agricultural processes, industrial wastes, and untreated waste are being discharged into Doosti dam [45] [40].

Physicochemical parameters almost at all stations were approximately at the level of Iran's standard values and the WHO guidelines. The highest values of electrical conductivity, calcium, and magnesium are observed in the south, east, and center sites. This could be due to the fact that the waters of these areas are likely to be supplied through Doosti dam. It was evidenced that several sources have affected quality of water in this dam such as successive droughts, thermal layering, severe evaporation from the surface of the water, chemical reaction of the reservoir floor with the water of dam, and the construction of the Salma dam on the upstream of the dam in Afghanistan [46] and lead to increase suspended and dissolved materials, particularly inorganic materials in this Dam [40, 45].

Our finding showed the insignificant temporal variation of Pb, Cr, and As concentration, while the concentration of Hg and Ni in spring significantly were higher than the summer.

Overall, this significant difference in the concentration of Hg and Ni is unusual during this short sampling period [45]. However, it was evidenced that chemical properties of water and heavy metal are associated with other environmental factors, such as seasonal fluctuations, atmospheric deposition, and change of pollution load from anthropogenic source, which lead to this temporal change in enrichment, bioavailability, and mobility of metals and other elements during a short time [47]. According to more studies, in the rainy season (Spring), the concentration of metals in environmental media (e.g., water and soil) is higher than dry season (Summer). This could be related to heavy rainfall that causes increase river discharges and land-based runoff. It can enter a large number of chemicals into water supplies for hundreds of a mile far from their place of origin [48, 49]. It has been confirmed in various studies that the concentration of Cr and As in water resources (dams, basins, wells and springs) depends on the soil texture of the area and its leak rate. In the study of water quality in Doosti Dam, it has been observed that in the rainy season, the concentration of water ions and salts in the water behind the dam increases[48, 49].

Our finding showed that the mean concentrations of all metals and physicochemical parameters (except EC and Mg) were lower than threshold levels stated in national and international guidelines to regulate drinking water quality and protect public health. According to this result, our hypothesis was rejected because, at all stations, the mean heavy metals concentration were lower than the guideline-threshold levels and rarely reached a concentration that lead to the adverse effect on human health. Furthermore, a significant decline in the concentration of heavy metals in drinking water was observed in this study compared with the previous study was conducted in 2013. Peiravi et al reported Pb and Cd concentrations were higher than threshold levels in some regions in Mashhad. This may attribute to the integrated management program in this city [50]. In 2014, integrated management programs were established, and responsible organizations (environment, Khorasan Razavi Regional Water Authority, wastewater agency and, NGOs) contributed to improve quality of freshwater supplies and drinking water in Mashhad. For example, useful rule ratified to prevent industrial waste derivatives from entering water resource, improve wastewater treatment and runoff control. This regulation and policy focused on strategies that were based on national agreements to improve wastewater treatment and runoff control. Contamination assessment of drinking water-based heavy metals compounds in this study was highlighted via two main limitations: the first limitation related to the scarcity of database on the heavy metals concentrations in drinking water of the Mashhad. The second limitation was related to the background information, as no local background information was available for the drinking waters.

CONCLUSIONS

Results from this study showed that no adverse health effect was associated with the exposure to heavy metals and other physicochemical parameters in the Mashhad, and only areas around the stations 1,4, 5, 6, 17, 18, 23, 22, 33, and 34 were dominated by heavy metals. Although the concentration of heavy elements in most locations is less than the permitted limit for drinking water that recommended by the national and international guidelines, more management and remediation efforts are needed to improve quality of drinking water in urban regions water supply network because of the inappropriate purification system in this city. Therefore, using the proper water treatment methods is essential to reduce health risks in local population.

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Availability of data and materials

The datasets generated and analyzed during this study are included in the main document of this manuscript.

Authors' contributions

BZGO involved in writing up of the research proposal, performed sampling and laboratory analysis, presented the results, and is the corresponding author. HA initiated the research concept, interpreted results, and finalized the manuscript document. SBTS presented the results and discussions, interpreted results, wrote up of the draft manuscript. MT and HSH performed laboratory analysis and analyzed the data. MF analyzed the data and involved in manuscript reviewing.

Compliance with Ethical Standards

We obtained ethical clearance from the Mashhad University of Medical Science.

Consent for publication

Not applicable

Conflict of Interest

The authors declare that they have no competing interests.

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