



## Health Risk Assessment of Nitrate, Nitrite, and Fluoride Ions in Water Reservoirs of Mehriz, Iran in 2023

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### ABSTRACT

**Introduction:** High concentrations of nitrate, nitrite, and fluoride in drinking water can cause adverse health effects. This study aims to investigate concentration and health risks assessment of nitrate, nitrite, and fluoride ions in water reservoirs of Mehriz city and Bahadoran district.

**Materials and Methods:** Monthly sampling was done from water reservoirs of Mehriz city and Bahadoran district for 6 months. Nitrate, nitrite, and fluoride concentrations were measured in the samples using a spectrophotometer. Then, health risk assessment and sensitivity analysis were performed on the obtained data using Crystal Ball software and Monte Carlo simulation method.

**Results:** Nitrate, nitrite, and fluoride concentrations were lower than the standard limitation in all of the studied areas. Risk assessment findings indicated that hazard quotient (HQ) values of nitrite in Miankoh-Movahedin, Bidok and Bahadoran water reservoirs were less than 1 for all age groups. HQ values of nitrate were also below 1 for all age groups except children. HQ values of fluoride in Bahadoran water reservoir were below 1 for all age groups except children.

**Conclusion:** Health risk of consuming water containing nitrates and fluorides is high for children. Based on sensitivity analysis, the concentration of nitrate and fluoride in drinking water is the key factor in raising health risks. Reducing nitrate and fluoride concentrations in drinking water can reduce health risks in the population.

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### Introduction

Due to population growth, demand for water is increasing rapidly<sup>1</sup>. Fresh water is not only used for health purposes, but also in other sectors such as industrial and agricultural purposes, so providing healthy and hygienic water for humans is necessary<sup>2, 3</sup>. Human activities have contaminated underground water, making it temporarily or permanently unusable for sanitary purposes. Various elements can enter water naturally or artificially, and has affected the health

of the exposed human communities<sup>4</sup>. Among the pollutants, nitrate has increased the risk of water pollution due to its high solubility<sup>5, 6</sup>. The utilization of ground water for drinking and agricultural needs is rising substantially on a global scale in arid and semi-arid areas, and about 1.5 billion people worldwide depend on groundwater for these purposes<sup>7</sup>.

Nitrate and nitrite are considered to be stable forms of nitrogen chains in aerobic environments. Nitrate is commonly utilized as a mineral fertilizer

to enhance soil fertility in agriculture. It is additionally utilized as a food preservative. Sodium nitrate is utilized as a preservative, specifically in canned meat products.<sup>8</sup> Nitrate concentrations in surface waters are naturally low, ranging from 0 to 18 mg/L. Inputs of surface runoff, especially from agricultural land, can increase nitrate concentrations in surface and groundwater. Nitrate concentration in surface water usually varies in different seasons<sup>9</sup>. In many European countries, nitrate concentrations in sources have increased over the past few decades, and in some cases, have doubled over the past 20 years. For example, in some rivers in the UK, nitrate concentrations have increased by an average of 0.7 mg/L per year<sup>10</sup>. The levels of nitrate in groundwater are influenced by the composition of the soil and its geological features. In the US, levels of nitrate and nitrite in groundwater generally remain below 49 and 0.3 mg/L, respectively<sup>11</sup>. Nitrate concentration can easily increase significantly with the expansion of agricultural activities<sup>12</sup>. For example, a concentration higher than 1500 mg/L of nitrate has been reported in groundwater of some areas in India<sup>13</sup>. The toxicity of nitrate for humans is determined by the conversion of nitrate to nitrite. The primary impact of nitrite in humans is its ability to convert hemoglobin into methemoglobin, preventing the transport of oxygen to tissues in body. Methemoglobin anemia is typically diagnosed when over 10% of hemoglobin transforms into methemoglobin. High concentrations of methemoglobin can cause suffocation and death. Typically, the body's methemoglobin level is under 2%, and in children less than 3 months, its concentration is less than 3%<sup>14</sup>. Nitrate concentration can increase in groundwater due to many reasons, such as the entry of agricultural runoff and industrial wastewater<sup>15</sup>.

Fluoride is a naturally occurring element present in many kinds of food and beverages, and in most groundwaters<sup>16, 17</sup>. The most important source of fluoride entering water and soil is chemical weathering of some fluoride-containing minerals

such as fluorite, cryolite, apatite, fluoromica, biotite, epidote, tremolite, hornblende, and the rocks containing them such as granite, basalt, limestone and shale<sup>18</sup>. About 90% of fluoride is absorbed through digestive system. Around 65% of cases of fluorosis are caused by high levels of fluoride in drinking water. Despite being necessary, fluorine can be harmful through inhalation and absorption through the skin. Minor exposure to skin can result in notable chemical burns<sup>19</sup>. One of the main causes of tooth decay in countries is the consumption of large amounts of sugar and insufficient fluoride exposure. This element can enter human body through water, food, drugs, etc., among which, the main way to get daily fluoride is drinking water. The World Health Organization (WHO) has determined acceptable concentrations of fluoride in the range of 0.5-1.5 mg/L in drinking water. Consumption of water containing a concentration lower than the allowed range of fluoride (less than 0.5 mg/L) can cause tooth decay and cavities<sup>20</sup>. However, if the amount of fluoride is more than the allowed range (more than 1.5 mg/L), it can have health effects. Consumption of water containing fluoride more than the allowed limit increases the risk of theoretical diseases of skeletal fluorosis, non-skeletal fluorosis, and dental fluorosis<sup>21</sup>. In fluorosis, the beauty and transparency of the teeth are lost causing chalk-like lines. In this case, paper white areas scatter irregularly over the tooth and brown stains can be seen on them, which, in addition to the loss of beauty, cause the destruction of dental tissues and makes them brittle, and the teeth rot over time and gradually fall out. The optimal level of fluoride depends on various parameters such as local climate, food processing methods, cooking, amount of food and water consumption, and eating habits of the community. Since the main way fluoride gets into the human body is by consuming water, it is necessary to control it in water<sup>20</sup>. Drinking water containing fluoride levels above 6 mg/L can lead to a significant increase in all the three forms of fluorosis including skeletal fluorosis, non-skeletal fluorosis, and dental fluorosis among water

consuming communities. On the other hand, ingestion of water with elevated levels of fluoride (up to 6 mg/L) has resulted in dental fluorosis more than skeletal and non-skeletal fluorosis<sup>22</sup>.

The issue of fluoride in drinking water and its potential health risks remains a significant concern, particularly in developing nations where the population relies heavily on underground water sources for various health-related needs, including drinking water. According to the reports of the WHO, Iran is located in the global fluoride belt; therefore, extensive research on this anionic pollutant in water sources should be considered<sup>20</sup>. The present cross-sectional, descriptive-analytical study was carried out in Mehriz city. Considering the use of urban and rural water sources for drinking by 100% of the people of Mehriz city and Bahadoran district (based on the statistics provided by Mehriz city water department) as well as the importance of fluoride, nitrate and nitrite in consuming water to prevent cancer, methemoglobinemia, dental caries, dental fluorosis, skeletal fluorosis, and non-skeletal fluorosis, the present study was conducted in summer and autumn seasons in 2023. An evaluation of health risks relies on input data like chemical levels and other parameters of risk models. This evaluation technique is able to assess the amount of real risk, particularly in regions with minimal risk. Consequently, it is necessary to assess potential health risks associated with underground water containing nitrates, nitrites, and fluorides and provide necessary suggestions to reduce the effects of the mentioned harmful ions on different age groups (women, men, and children). Due to the importance of the issue, after determining nitrates, nitrites, and fluorides concentrations in water reservoirs, the present study determined the health risks of these Ions among residents of Mehriz city

and Bahadran district.

## Method

### Study area

The study area was Mehriz city and Bahadoran district in Iran. Mehriz city is located in the south of Yazd province, 30 km from Yazd city and next to the Tehran-Bandar Abbas (Yazd-Kerman) highway, with a longitude of 54.44515 and a latitude of 31.58299. The population of this area is 54,201 people, 35,399 of whom live in an urban area with an average water consumption of 2,800 m<sup>3</sup>/day (the area covered by Bidok reservoir) and 4,200 m<sup>3</sup>/day (the area covered by Miankoh-Movahedin reservoirs). The study area has a hot and dry climate and agriculture in the area is limited to pistachio cultivation in Bahadoran, which is far from the study area. The source of water supply in all areas are wells. The region has 18 water wells, which are used to transfer and supply water to the city. The depth of the wells is 50 meters in Mehriz and 140 meters in Bahadran. Bahadoran district has a population of 3,800 people and is located at a distance of 60 km from Mehriz city and has a longitude of 54.91685 and latitude of 31.32565 with an average water consumption of 550 m<sup>3</sup>/day. It has two water wells used to supply water to the district. The water of the studied wells is only used for domestic purpose and the only purification process is chlorination.

Sampling was done from water reservoirs of two water supply systems in Mehriz city and one water reservoir in Bahadoran district in a 6-month period (summer and autumn seasons) in 2023. The samples were taken monthly and transferred to the laboratory at 4°C, and nitrate, nitrite, and fluoride concentrations were measured. Figures 1 and 2 show the location of the studied stations.



**Figure 1:** The location of the studied stations in Mehriz city.

It should be noted that the three mentioned points are water reservoirs used for water supply, from which sampling was done (the upper two points are related to Miankoh and Movahedin

reservoirs, which were connected at the time of the research, and the lower point is related to Bidok reservoir and was sampled from the outlet).



**Figure 2:** The location of the studied stations in Bahadoran district.

### Measurement of parameters in water

To determine the concentration of nitrate, nitrite, and fluoride in water, the Palintest photometer (Model 8000, England) was used. The absorbance of samples and standard solutions was defined by a spectrophotometer to measure nitrate, nitrite, and fluoride at wavelengths of 220, 543, and 570 nm, respectively.

### Health risk assessment

To assess the health risks, the population in studied area was divided into three age groups including 3-10 years (children), 11-20 years (teenagers), and 21-72 years (adults).

Equation (1) from the USEPA was used to determine the daily intake of nitrate, nitrite, and fluoride content in water in this study. The parameters studied in health risk assessment are given in Table 1.

$$EDI_{ing} = \frac{C_w \times IR_w \times EF \times ED}{BW \times AT} \quad (1)$$

$EDI_{ing}$ : The amount of nitrate, nitrite, and

fluoride received daily through consumption of drinking water,

$C_w$ : Concentration of nitrate, nitrite, and fluoride in drinking water in mg/L,

$IR_w$ : Ingestion rate of drinking water per person per day in L/day,

$EF$ : Exposure frequency based on day/year,

$ED$ : Exposure duration in a year,

$BW$ : Body weight in Kg,

$AT$ : Average time in a day.

### Hazard quotient (HQ)

Non-carcinogenic risk estimation related to nitrate, nitrite, and fluoride exposure through consumption of drinking water was calculated using Equation (2).

$$(HQ = \frac{EDI}{RfD}) \quad (2)$$

The RfD represents the recommended daily dose of fluoride, nitrate, and nitrite for a particular exposure route, measured in mg/kg/day.

According to the Integrated Risk Information System (IRIS4) database related to the USEPA, the

RfD value through drinking water intake is 1.6 mg/kg/day for nitrate, 0.01 mg/kg/day for nitrite, and 0.06 mg/kg/day for fluoride<sup>23</sup>. Parameters

applied for the probabilistic risk assessment are presented in Table 1.

**Table 1:** Parameters used for the probabilistic risk model<sup>24</sup>

Parameter	Value (Mean ± St)		
	Children	Teenagers	Adults
Body weight (kg)	16.68 ± 1.48	46.25 ± 1.8	57.03 ± 10
Ingestion rate (l/day)	1.25 ± 0.57	1.58 ± 0.69	1.95 ± 0.64
Exposure frequency (day/year)	365	365	365
Exposure duration (year)	6.52 ± 2.32	14.82 ± 2.25	46 ± 15
Average time (day)	2190	5475	16790
Oral reference dose (mg/kg/day)	Fluoride	0.06	0.06
	Nitrate	1.6	1.6
	Nitrite	0.01	0.01

## Results

Overall, 36 samples were taken. The concentration of the parameters in summer and autumn in the studied reservoirs are shown in Table 2. According to the results, mean concentrations of nitrate, nitrite, and fluoride in water reservoirs of the studied areas were lower than mean values determined by the U.S. Public Health Service and Iran standard No.1053 (50, 3 and 1.5 mg/l for nitrate, nitrite, and fluoride respectively). The highest concentration of nitrate with the value of 36.126 mg/L was related to Bidok water reservoir in autumn, and the lowest

concentration of nitrate with the value of 20.66 mg/L was related to Bahadoran district water reservoir in autumn. Regarding nitrite, the highest concentration (0.02 mg/L) was related to Bahadoran district water reservoir in autumn, and the lowest concentration (0.0063 mg/L) was related to Bidok reservoir in Mehriz city. The highest fluoride concentration (1.053 mg/L) was related to Bahadoran district water reservoir in summer, and the lowest fluoride concentration (0.703 mg/L) was related to Miankoh-Movahedin reservoirs of Mehriz city in autumn.

**Table 2:** Concentration of studied chemical in different seasons and studied area

Place	Parameter	Summer			*Ave	**SD	Autumn			Ave	SD
		July	August	September			October	November	December		
Miankouh Movahedin	Nitrate	25.9	41.5	25.89	31.096	9.009	20.79	27.2	29.68	25.890	4.587
	Nitrite	0.007	0.014	0.006	0.009	0.004	0.005	0.006	0.017	0.009	0.006
	Fluoride	0.72	0.82	0.64	0.726	0.090	0.54	0.82	0.75	0.703	0.145
Bidok	Nitrate	31.08	20.86	29.8	27.246	5.567	45.16	25.01	38.21	36.126	10.235
	Nitrite	0.006	0.007	0.006	0.006	0.0005	0.016	0.017	0.026	0.0196	0.005
	Fluoride	0.84	0.62	0.75	0.73667	0.110	0.75	0.57	0.82	0.713	0.128
Bahadoran	Nitrate	15.5	33.25	34.02	27.590	10.477	34.25	15.24	12.5	20.663	11.845
	Nitrite	0.006	0.01	0.01	0.008	0.002	0.018	0.023	0.024	0.021	0.003
	Fluoride	1.15	0.96	1.05	1.053	0.095	0.79	0.73	0.75	0.756	0.030

\*Ave; Average  
 \*\*SD; Standard Deviation

In general, the concentration of the parameters in all samples were within the standard range (Iran Standard No.1053). Based on the results reported in Tables 3, 4, and 5, HQ values of nitrate in Miankoh-Movahedin, Bidok, and Bahadoran reservoirs in summer and autumn seasons were

greater than 1 for the children. Moreover, HQ values of fluoride in Bahadoran reservoir in summer and autumn seasons were more than 1 for children. HQ values of nitrite in the study areas for all age groups were less than 1.

**Table 3:** EDI and HQ for different seasons and chemicals in the studied areas for adults (21-72 years)

Studied areas	Season	Result	Fluoride	Nitrate	Nitrite
Miankouh Movahedin	Summer	EDI	0.0261	1.0850	0.0003
		HQ	0.4352	0.6781	0.0319
	Autumn	EDI	0.0256	0.8979	0.0003
		HQ	0.4273	0.5612	0.0327
Bidok	Summer	EDI	0.0251	0.9437	0.0002
		HQ	0.4188	0.5898	0.0211
	Autumn	EDI	0.0247	1.2962	0.0007
		HQ	0.4109	0.8101	0.0715
Bahadoran	Summer	EDI	0.0367	0.9492	0.0003
		HQ	0.6120	0.5932	0.0286
	Autumn	EDI	0.0265	0.7433	0.0007
		HQ	0.4416	0.4646	0.0743

**Table 4:** EDI and HQ for different seasons and chemicals in the studied areas for teenagers (11-20 years)

Studied areas	Season	Result	Fluoride	Nitrate	Nitrite
Miankouh Movahedin	Summer	EDI	0.0240	1.0779	0.0003
		HQ	0.4002	0.6737	0.0297
	Autumn	EDI	0.0236	0.8845	0.0003
		HQ	0.3940	0.5528	0.0307
Bidok	Summer	EDI	0.0250	0.9273	0.0002
		HQ	0.4173	0.5795	0.0200
	Autumn	EDI	0.0240	1.2241	0.0006
		HQ	0.3992	0.7651	0.0622
Bahadoran	Summer	EDI	0.0352	0.9627	0.0003
		HQ	0.5873	0.6017	0.0270
	Autumn	EDI	0.0257	0.6477	0.0007
		HQ	0.4291	0.4048	0.0708

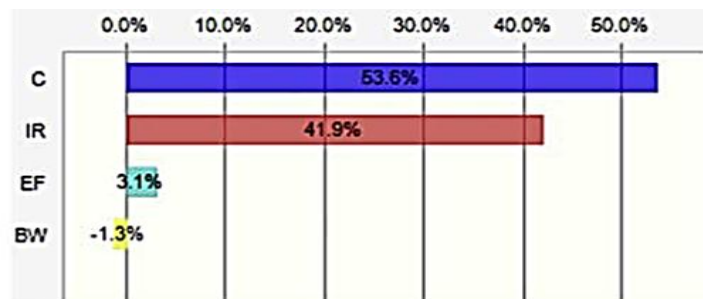
**Table 5:** EDI and HQ for different seasons and chemicals in the studied areas for children (3-10 years)

Studied areas	Season	Result	Fluoride	Nitrate	Nitrite
Miankouh Movahedin	Summer	EDI	0.0595	2.5680	0.0008
		HQ	0.9917	*1.6050	0.7600
	Autumn	EDI	0.0579	2.1132	0.0007
		HQ	0.9655	*1.3208	0.0747
Bidok	Summer	EDI	0.0585	2.1836	0.0005
		HQ	0.9746	*1.3647	0.0485
	Autumn	EDI	0.0581	2.9276	0.0016
		HQ	0.9688	*1.8298	0.1600
Bahadoran	Summer	EDI	0.0845	2.3554	0.0007
		HQ	*1.4089	*1.4721	0.0657
	Autumn	EDI	0.0621	1.6295	0.0017
		HQ	*1.0355	*1.0180	0.1693

\*HQ > 1; high non-cancer risk

Based on the findings of the sensitivity analysis (Figure 3), the most critical variable contributing to the growth of non-carcinogenic risk of nitrate and

fluoride in children is the concentration of these elements in drinking water of the study areas.



**Figure 3:** Diagram of sensitivity analysis in children



## Discussion

### *Health risk assessment of fluoride*

The average values of non-carcinogenic risks for every age range except children in Bahadoran district water reservoir were estimated to be less than 1, showing a significant non-carcinogenic risk value for children. The reason for higher non-carcinogenic risks for children is their low body weight compared to other age groups<sup>25</sup>. The greatest mean value and 95th percentile for HQ calculated in the study areas related to Bahadoran reservoir and in summer and autumn seasons were 1.408 and 1.035, respectively, for children, indicating the high non-carcinogenic risk of fluoride in this district. Within the designated research zone, the non-carcinogenic risk of fluoride for three groups of exposed population was as follows: adults < teenagers < children. Based on the findings of the health risk assessment, children were the population potentially at risk, which is in line with the study by Guissouma et al. (2017)<sup>26</sup>. Yousefi et al. carried out research to evaluate the health risk assessment of fluoride in the drinking water of rural inhabitants in Poldasht city, located in the northwest of Iran. The study's findings revealed that maximum risk of contact with fluoride in different areas of Poldasht city was in young, children, and teenage consumers<sup>27</sup>. Dehghani et al. evaluated fluoride contamination in underground water sources in southern Iran and the risks related to human health in 2018. Health risk assessment results showed that in 70.6, 48.2, and 34.4% of the collected samples, the health risks of fluoride for children, teenagers, and infants were higher than 1, indicating that these age groups are at risk by consuming drinking water. The health risks were in the order of children > women > men<sup>28</sup>. Mirzabeigi et al. (2018) also conducted a study entitled "Fluoride concentration data and health risk assessment in drinking water of Ardakan city, Yazd province". The results of risk assessment showed that HQ values in 46.4% of samples of groundwater sources exceeded 1 for infants, children, teenagers and adults<sup>29</sup>. Yan Hu et al. carried out an investigation to assess the

geographic spread and potential health risks associated with fluoride in drinking water sources of Huabei and Anhui provinces of China in 2020. Their results showed that HQ values of fluoride were different in three exposed groups (infants > children > adults) and infants and children were at higher risk than adults<sup>30</sup>. The results of similar studies have shown that children are more sensitive to health effects of fluoride in water compared to other age groups, and necessary measures should be taken in this regard.

### *Nitrate health risk assessment*

The average values of non-carcinogenic risk exists for every age group except children in Miankoh-Movahedin, Bidok and Bahadoran water reservoirs were less than 1. HQ values for the 95th percentile in teenagers and adults were under 1 and for children in all of the studied areas was higher than 1, indicating a high non-carcinogenic risk value for children. In all of the studied areas, the non-carcinogenic risk of nitrates for the three groups of exposed population was as follows: adults < teenagers < children. Based on the findings of the health risk assessment, children are at potential risk, which is in accordance with the study by Guissouma et al. (2017)<sup>26</sup>. Moreover, Pirasteh et al. carried out research to assess the risks of nitrates in drinking water in urban and rural areas of Zahedan city in 2018. The results showed that health risk values of the samples in 44% of the children group, 14% of the women group, and 8% of the men group and in urban areas for 55% of the children group, 22% of the women group, and 18% of the men group were higher than 1<sup>31</sup>. Mohammadi et al. assessed non-carcinogenic risks caused by nitrates in the potable water in both urban and rural regions of Bostan city. The results of health risk assessment showed that HQ values in 5.55% of urban water samples and 11.25% rural water samples were more than 1 for children. According to the results, in most cases, the amount of nitrate in drinking water was below than the national standard of Iran and the WHO (50 mg/l). However, children are exposed to non-carcinogenic risks caused by nitrates in drinking

water<sup>32</sup>. Narsimha Adimalla et al. also conducted a study in 2018 to assess the health risks of nitrate and fluoride in the area with water scarcity located in Nirmal city located in southern India. The health risk assessment findings indicated that children compared to men and women in the study area are at a higher risk<sup>33</sup>. The results of similar studies have shown that children are more sensitive to the health effects of nitrates in water compared to other age groups.

#### **Health risk assessment of nitrite**

The average values of non-carcinogenic risks for all groups were estimated to be less than 1. HQ values for the 95th percentile in teenagers, adults, and children were below 1, showing a low non-carcinogenic risk for all age groups.

#### **Sensitivity analysis**

For all age groups, in all studied areas, the most crucial factor influencing health risk values was the concentration of nitrate and fluoride in drinking water, with an impact factor of 53.6% (Figure 3). Based on the sensitivity analysis, next parameters included the amount of daily consumption of drinking water (41.9%), number of exposures (3.1%), and finally body weight (-1.3%). In Mehriz city and Bahadoran district, the concentration of the mentioned elements in drinking water is the key factor that influences the level of health risks within different age categories.

#### **Conclusion**

In the current study, the concentration of nitrate, nitrite, and fluoride and its health risks in drinking water reservoirs of the study areas were investigated in two seasons. Of the 36 samples taken from these reservoirs, 100% were within the standard range (Iran Standard No.1053). The results showed that HQ values of nitrate and fluoride for all age groups except children were less than 1; therefore, children are at risk. Based on the findings of the sensitivity analysis, the most important variable increasing non-carcinogenic risks of nitrate in children is nitrate concentration in drinking water of the study areas. Moreover, the most important variable increasing non-carcinogenic risks of fluoride in children is

fluoride levels in drinking water of Bahadoran district. Therefore, it is possible to reduce the amount of risks in the population at risk by reducing nitrate and fluoride concentrations.

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#### **Conflict of Interest**

The authors declare no conflict of interest.

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#### **Ethical Considerations**

The authors obtained permission from the Ethics Committee of Shahid Sadoughi University of Medical Sciences.

#### **Code of Ethics**

The study was approved by the university's Ethics Committee with ethics code of IR.SSU.SPH.REC.1402.053

#### **Authors' Contributions**

All the authors contributed to the study's conception and design. Preparation of material, data collection, and analysis were performed by Ali Ghasemzadeh, Reza Ali Fallahzadeh, Vahid Jafari Nodoshan, Hossein Fallahzadeh and Arash Dalvand. The first draft of the manuscript was written by Ali Ghasemzadeh, Reza Ali Fallahzadeh, Mohammad Taghi Ghaneian and Arash Dalvand. All the authors commented on the previous versions of the manuscript and read and approved the final manuscript.

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