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ORIGINAL ARTICLE

Investigation of Microbial Contamination of the Mahabad Dam Reservoir in Different Seasons

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	ABSTRACT: Mahabad dam was constructed to supply drinking water, irrigation, and electricity generation on the
KEYWORDS	Mahabad river located West Azerbaijan, Mahabad. In this research, bacterial contamination of the water of mahabad
	dam was investigated. According to the geographical properties of the reservoir, 3 stations (near the dam crest, Koter
Mahabad dam reservoir;	branch, and Bitas branch) were selected and samples were taken via Rottner from different depths of the reservoir
Total coliforms; Fecal coliforms;	during 12 months. Bacterial contamination (Total Coliforms, Fecal Coliforms, and Fecal Streptococci) were studied
Fecal Streptococci;	after transferring the samples to laboratory. Results indicated that values of Total Coliforms, Fecal Coliforms, and
Seasons	Fecal Streptococci were varied between 2.33 \pm 1.20 and 1166.67 \pm 33.33, 0.0 and 30.33 \pm 6.74 and 0.0 \pm 16.67 \pm 6.33
	MPN/100 ml respectively during over all the year. These values were in the allowed range according to the drinking
	water and recreation standards of Iran.

INTRODUCTION

The limitation of water resources on the earth and the ever-increasing population, high living standards, industrialization of human societies, and the increasing need for water have required the construction of dams for supplying agricultural and drinking water. However, the mentioned solution has resulted in extensive changes in the ecosystem of rivers; consequently, human activities have increased the negative effects by applying negative management. In the past decades, scientists stated that the preservation and optimal use of water resources as the principles of sustainable development in any country and claim that water resource planning should be based on the potential of surface and underground water resources. Surface water includes only 0.02% of the total water on the earth [1]. It will be enough for human consumption that low level of surface water is distributed equally and kept safe and healthy using appropriate management. Otherwise, water quality will become one of the biggest problems for humans. Today, waterborne diseases are a serious threat to developing countries [2]. The critical sources of water contamination include urban, industrial, and agricultural sewage [3]. Therefore, most countries have developed some guidelines for maintaining the appropriate quality of water and their sustainable use [4]. Currently, in order to control the quality of water, in addition to physicochemical properties, water is analyzed for microbial contamination (total fecal bacteria, coliforms, and *Escherichia coli*) [5]. Serotype O157 is one of the most important verotoxigenic strains of *E.coli* that are found in many regions of the world [6]. Europe, America, and Asia have caused epidemics or individual cases of infection in humans [7, 8].

Coliforms are used as an appropriate microbial index to indicate fecal contamination in water sources. The type of bacteria to be used as a microbial index due to coliform population increases is the digestive system of warm-blooded animals. Thus, they are highly present in feces and their durability in water is longer than intestinal pathogenic bacteria. Thus, intestinal pathogenic bacteria along with coliforms will be present in the water contamination. They do not reproduce considerably in water since water is not their natural habitat. Hence, the relative number of coliforms will indicate the proportion of fecal contamination in various water samples. On the other hand, their cultivation, counting, and isolation in the laboratory are easier compared to intestinal pathogenic bacteria. However, it was found that all the bacteria which are in the coliform category do not isolate from feces, but some coliforms have another origin such as soil. Hence, a sub-category was defined for coliforms and since they isolate from feces, they are called fecal coliforms. All the coliforms including fecal and nonfecal are called total coliforms [5]. Awareness of the quality of water resources is one of the significant requirements in developing, planning, preserving, and controlling water resources. Obviously, evaluations and monitoring should be conducted for collecting information about the quality of water sources since having comprehensive and reliable information about water resources with appropriate time can be a key factor in the goals, policies, and management plans of water resources [9].

Mahabad dam was constructed to supply drinking water, agricultural, and power generation in the West Azarbaijan province and Mahabad city. Due to the significance of the reservoir in the supply of drinking water, irrigation, and the environment, the presentation study evaluated the amount of bacterial contamination at Mahabad dam reservoir.

MATERIALS AND METHODS

The studied area

The mahabad dam reservoir is located one kilometer west of Mahabad on the Mahabad river in the West Azarbaijan province at longitude 45 degrees and 25 minutes to 46 degrees and 45 minutes east and at latitude 36 degrees and 23 minutes to 37 degrees and 03 minutes north. The volume of the reservoir in normal mode is 197.8×10^6 m³ and its useful volume is 170.3×10^6 m³. The type of dam is gravel with clay core and the height of the crest from the bed is 45 meters while the length of the crest is 700 meters. The drainage basin of the Mahabad dam reservoir [10] is one of the major sources of drinking and agricultural water in Mahabad and includes three sub-basins named Bitas, Koter, and Dehbakr [11]

Sampling

In order to the water quality assessment of the reservoir, three stations were selected in the Mahabad dam reservoir including Kouter related to the Kouter branch, Bitas related to the Bitas branch, and the station near the dam crest close to the axis of the dam reservoir. Sampling of the dam reservoir was performed from four depths according to the prepared guideline. The depth of the sampling points was not constant in different months and was determined based on the temperature profile of the reservoir water. In this regard, the temperature of the reservoir was determined before sampling and the appropriate depth for sampling was selected as one meter below the water level, just above the upper limit of the thermocline, just below the lower limit of the thermocline, and one meter above the bed sediments. Then, sampling was conducted by Ruttner sampler. About 100 ml of samples were transported to the laboratory at 4°C. Sampling of reservoir water was carried out over 12 months.

Microbial quality assessment

In this study, the microbial quality of water samples from 3 stations were randomly evaluated to determine the presence of coliforms by the MPN technique. In this test, first of all three tubes of lactose broth culture medium (26 g L⁻¹) containing 10 ml of lactose broth culture medium and six tubes of lactose broth culture medium (13 g L⁻¹), each containing 10 ml of culture medium, are used. Then, 10 ml of the sample, which has been prepared in a sterile condition, is added to each of the first three tubes. After that, one ml of the sample is added to each of the tested sample is added to the next three tubes. Then, they are incubated at $35^{\circ}C \pm 0.5$ for 24 hours. According to the MPN method, the presence of gas in all Durham

tubes shows a positive result, while the absence of gas and the presence of gas in some tubes causes the samples to be autoclaved for 24 hours. Finally, Durham tubes based on the presence of gas were evaluated after 48 hours [5].

Statistical analysis

ANOVA and Duncan's test were applied to means of total coliform, fecal coliform, and fecal streptococcus at 5% significance level. Statistical analyses were conducted using SPSS 22.

RESULTS

Total coliform

Total coliform values were between 2.70 ± 1.72 and $\pm 0.780\pm184.75$ per 100 ml in the station near the dam crest between, 3.67 ± 2.02 and 1166.67 ± 33.33 per 100 ml in Kouter station, and between 2.33 ± 1.20 and 1150 ± 28.87 per 100 ml in Bitas station (Table 1). The total coliform values of the Kouter station were significantly

higher than Bitas station and the station near the crest in April and February months. The value in Kouter and Bitas stations was significantly higher than the station near the crest in March month (p<0.05). No significant difference was found between the stations in other months of the year (p>0.05). The values at all three stations from July to January were significantly less than other months of the year (except April and February months at the station near the crest) (Figure 1). At the station near the crest, the values were significantly more in May and June months than in other months. In addition, total coliform at Kouter and Bitas stations in June and March had a significant difference than other months of the year (p<0.05).

Figure 2 shows the effect of depth changes on total coliform value at the station near the dam crest of Mahabad dam during different months of the year. As can be observed, the values have experienced deep changes in some months of the year. However, there have been some changes with increasing depth in some months of the year.

Month	Bitas branch	Koter branch	Near the dam crest
April	77.33±8.45a	234.33±114.03b	30.25±6.06a
May	403.33±56.67a	248.33±112.78a	702.5±238.06a
June	673.33±213.33a	460±0a	780.37±184.75a
July	2.4±1.20a	7.67±7.67a	2.7±1.72a
August	4.23±2.65a	13.87±5.63a	4.55±2.63a
September	25.03±9.84a	31.07±21.97a	7.1±5.35a
October	2.33±1.2a	26.67±6.17a	35.03±19.53a
November	4.33±2.6a	7.33±4.33a	3.38±1.97a
December	4.33±2.6a	3.67±2.02a	4.25±1.84a
January	10.33±6.33a	10.33±6.33a	5.5±2.18a
February	77.33±8.45a	200±26.46b	38.5.±6.34a
March	1150.67±28.87b	1166.67±33.33b	265±67.64a

Data are mean±standard error. Different letters in each row show a significant different (p<0.05)



Figure 1. Total coliform count of the the Mahabad dam reservoir during different month of the year in different stations based on the average of various depths.



Figure 2. Total coliform of the Mahabad dam reservoir during various months (near the dam crest) at different depths.

Fecal coliform (digestive)

Table 2 shows that the water of the Mahabad dam reservoir didn't have fecal coliform contamination at the station near the dam crest in April, May, September, December, and February months, at Kouter station in April, May, September and February months, and at Bitas station in April, May, October, September, and February months.

Kouter and Bitas stations indicated a significant difference compared to the station near the dam crest only in March month (p<0.05) and there was no significant difference between the stations in other months (p>0.05). At the station near the dam crest, the highest amount of fecal coliform was found in June

month (10.75 \pm 6.29 per 100 ml) which was significantly different from the other months of the year. Nevertheless, its value at Kouter station in March (21.0 \pm 0.7 per 100 ml) had a significant difference than other months (p<0.05). At Bitas station, the amount of fecal coliform in June and March months (30.33 \pm 6.74 and 0.25 \pm 11.36 per 100 ml, respectively) was more than in the other months of the year (Figure 3). Figure 4 shows the effect of depth changes of fecal coliform at the station near the dam crest during different months of the year. The maximum changes occurred in June, July, and August months.

Month	Bitas branch	Koter branch	Near the dam crest
April	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
May	0.0±0.00	0.0±0.00	0.0±0.00
June	30.33±6.74a	8.00±8.00a	10.75±6.29a
July	2.2±1.11a	3.03±3.03a	1.8±1.8a
August	1.2±1.2a	13.87±5.63a	3.17±2.15a
September	0.0±0.00	0.0±0.00	0.0±0.00
October	0.0±0.00a	4.03±2.68a	1.35±0.86a
November	2.67±1.33a	1.00±1.00a	0.75±0.75a
December	2.67±1.33a	2.33±2.33a	0.0±0.00
January	1.33±1.33a	3.00±3.00a	1.00±1.00a
February	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
March	25.00±11.36b	21.00±7.00b	0.75±0.75a

Table 2. Fecal coliform count different stations of the Mahabad dam reservoir

Data are mean±standard error. Different letters in each row show a significant different (p<0.05).



Figure 3. Fecal coliform count of the Mahabad dam reservoir during different month of the year in different stations based on the average of various depths.



Figure 4. Fecal coliform of the Mahabad dam reservoir during various months (near the dam crest) at different depths.

Fecal streptococcus

The water of Mahabad reservoir dam at the station near the dam crest had no fecal streptococcal contamination in April, May, July, February, and March months, at Kouter station in April, May, December, and February month, and at Bitas station in April, May, July, August, September, January, and February months(Table 3). Results indicated a significant difference between the stations in October and December months and its value was significantly more at the station near the dam crest in October month than in the other two stations. Furthermore, the value was higher at Kouter station and in December month. Comparing the data of different months indicated that the station near the dam crest had the highest amount of fecal streptococci in October month (7.27 \pm 1.82 per 100 ml), which was not significant in comparison to June, November, and December months (p<0.05). (Figure 5). The highest value was found at Kouter station in March month (10.0 \pm 5.77 per 100 ml), which was significantly different from other months except for June, July, November, and December months. The values at Bitas station in June, November, and March months (13.33 \pm 3.33, 16.67 \pm 6.33, and 10.0 \pm 0.10 per 100 ml, respectively) were significantly more than the other months (p>0.05). Figure 6 displays the effect of depth changes of fecal streptococci at the station near the dam crest during different months of the year. The most changes were observed in June, October, and November months.

Month	Bitas branch	Koter branch	Near the dam crest
April	0.0±0.00	0.0±0.00	0.0±0.00
May	0.0±0.00	0.0±0.00	0.0±0.00
June	13.33±3.33a	3.33±3.33a	5.00±2.89a
July	0.0±0.00a	2.53±2.53a	0.0±0.00
August	0.0±0.00a	1.00±1.00a	0.9±0.9a
September	0.0±0.00a	1.2±1.2a	0.9±0.9a
October	1.2±1.2a	1.2±1.2a	7.27±1.82b
November	16.67±6.33a	6.00±3.00a	2.75±2.75a
December	2.33±1.2ab	0.00±0.00a	4.00±1.08b
January	0.0±0.00a	3.00±3.00a	1.00±1.00a
February	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
March	10.00±10.00a	10.00±5.77a	0.00±0.00a

Table 3. Fecal streptococcus count different stations of the Mahabad dam reservoir.

Data are mean±standard error. Different letters in each row show a significant different (p<0.05).



Figure 5. Fecal streptococcus count of the Mahabad dam reservoir during different month of the year in different stations based on the average of various depths



Figure 6. Fecal streptococcus count of the Mahabad dam reservoir during various months (near the dam crest) at various depths. DISCUSSION

Water contamination by fecal bacteria, which is a result of the addition of sewage of humans and animals into water sources, is regarded as one of the main factors damaging the ecological quality of water and causing human and animal diseases. The origin of surface water contamination can be important points such as factories or urban wastewater. Decentralized contamination such as surface water leaching during rains or the direct effect of humans and animals into the water basin, as well as the lack of suitable efficiency of their purification systems often play a more essential role in surface water contamination [12]. Since identifying all pathogenic bacteria is highly difficult, time-consuming, and expensive, some bacteria are used to determine water contamination. Fecal coliforms and streptococci are the natural intestinal flora of humans and warm-blooded animals, which enter nature continuously in large numbers through feces. Hence, determining Escherichia coli and Enterococcus faecalis bacteria in water sources along with total coliform is used as an appropriate method to determine the water contamination [13]. In the present study, the amount of coliform in the reservoir increased with the increase of input to the reservoir in March, May, and June months, and the low level of coliform value were found in the other months. The highest amounts of coliforms were mainly found under the layer in May and June months, the main cause of which was the transport of coliforms along with sediments and suspended materials to the river and the sedimentation process in the reservoir.

In terms of recreational consumption, the maximum allowed total coliform is 2000 per 100 ml according to the presented standard. Since the maximum measured value was over 1000 per 100 ml in May and June months, the total coliform of the Mahabad dam reservoir was within the allowed limits for 1 year. However, the depletion of water quality in May and June months can be occurred due to the majority of recreational consumption is conducted in summer; there is no problem in total coliform contamination in summer season. Fecal coliform is the result of contamination with human sewage. In terms of the drinking standard, its value should be slight, although it is not specified in the Iranian drinking water standard. Compared to the recreational standard, the maximum value of fecal coliform in the present study was much less than the recreational standard for direct contact (400 per 100 ml) [14]. The increase of fecal coliform values up to 30 per 100 ml in June month could be the result of swimming and recreational consumption of the reservoir. The increased fecal coliform value in March month may be related to rains and the washing effect of surface water. Fecal streptococcus shows water contamination with animal waste. The mentioned parameter originated from the upstream area in spring and March month. The increase in Fecal streptococcus value can be explained based on cattle grazing the reservoir around and the dryness of the rivers entering the reservoir in summer and October month.

Evaluation of Escherichia coli and enterococci bacteria values, which are indicators of fecal bacteria, indicated that the microbiological quality of water in the main rivers of the Scheidt watershed was poor. The value of E. coli was 1.4×10^3 to 4×10^5 and the value of Enterococci was 3.4×10^2 to 7.6×10^4 per 100 ml [15] which was much more than the present study. Evaluating the physicochemical and microbial characteristics of the water at the Owena multi-purpose reservoir in the dry and rainy seasons showed numerous seasonal changes in the studied parameters. The bacterial load was slightly higher compared to the drinking water standard [4]. In this study, the total coliform was between 0.1×10^2 and 0.69×10^2 in the dry season and also between 3.03×10^2 and 87.87×10^2 per 100 ml in the rainy season, which was consistent with the present study. Hence, in this study bacterial contamination of water has been affected by seasonal rainfall that is consistent with the results of mentioned studies

Mahmoudi and Javanmardi [16] studied the fecal bacteria population in the Parishan Lake for 12 months based on the standard of the World Health Organization. The results indicated that the fecal coliforms population in April, November, February, and March months (127, 108, 355, and 133 CFU per 100 ml, respectively) was more than the allowed limit stated by the World Health Organization. The lowest amount (7 CFU per 100 ml) was observed in August month. Hence, the contamination with fecal coliform at the Mahabad dam reservoir in the present study was much less than that of the Parishan Lake. The trend of changes in fecal streptococci and total coliforms was consistent with fecal coliforms. The water contamination of the Parishan lake had a similar trend with the amount of rainfall resulting in microbial contamination increased when the amount of rainfall was more frequent due to the leaching of environmental pollution and the transfer of fecal matter through seasonal streams from the beach into the lake [16]. The maximum value of fecal coliform (400 colonies per 100 ml) of water sample from the Shirin Dareh dam reservoir was related to September month while the minimum value (184 colonies per 100 ml) was related to June month. In general, the amount of fecal coliform in summer season is more than the in other

seasons, but its values in all months are less than the standard discharge of fecal coliform to surface water which is due to the long length and sedimentation of the lake according to the other works. However, the result showed that the quality water in the present study was much higher than Shirin Dareh dam, which may be attributed to the large number of villages around the reservoir dam mentioned [17].

CONCLUSIONS

The preservation and optimal use of water resources are one of the principles of sustainable development in any country. Most water resource planning in a country is according to the potential of surface and underground water resources of that country. Awareness of the quality of water resources is one of the significant requirements for developing, planning, preserving, and controlling water resources. Also, evaluations and monitoring should be conducted for collecting information about the quality of water sources since having comprehensive and reliable information about water resources with appropriate time can be a key factor in the goals, policies, and management plans of water resources. The water quality of the Mahabad dam reservoir showed that its bacterial load was within the allowed limits. Nevertheless, the possibility of violating the allowed limits is not far from expected in some months of the year, especially in late spring. Nevertheless, it is possible to increase contamination to more than the allowed limits in some months of the year, especially in late spring

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Conflict of interests

The authors declare that there is no conflict of interest.

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