

original article

Evaluation of biological and physico-chemical quality of public swimming pools, Hamadan (Iran)

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ABSTRACT

Aims: The purpose of this study was to determine the types of fungal contamination and evaluation the fecal *Streptococcus*, total *coliform* and some physical and chemical parameters of swimming pool waters in Hamadan (Iran)

Materials and Methods: In this study, biological and physico-chemical quality were evaluated. Bacterial and fungal contamination of four public indoor swimming pools was evaluated by standard total coliforms fermentation and using membrane filtration and carpet and swab sampling method with a month interval. In addition, physical and chemical parameters such as residual chlorine, temperature and pH were measured.

Results: Results showed that the mean±SD water temperature, pH and residual chlorine were 29.3±1.3, 7.38±0.5 and 0.84±0.5 mg/L, respectively. Total count was 1.8±7.7 MPN/100ml and its maximum and minimum were 43 and 0 MPN/100ml, respectively. Fecal streptococcus count was 0.3±1.6 MPN/100ml and its maximum and minimum were 9 and 9 MPN/100ml, respectively. In the environment of pools, fungi were isolated in order of *Cladosporium* spp., *Penicillium* spp., *Aspergillus* spp., *Alternaria* spp., *Aspergillus.niger* spp., *Rhodotorula* spp and *Phoma* spp. In the collected water sample, no fungal growths were seen. In the studied pools, the microbial and fungi contamination showed significant and insignificant difference, respectively.

Conclusion: As results showed the residual chlorine in pools water was lower than standard level and as regard to microbial contamination in pool water, it can be concluded that the disinfection system has been impaired.

Key word: Fungi, physic-chemical parameters, swimming pool

INTRODUCTION

Swimming is an active and healthy way to spend free time and is one of the popular activities in Iran. It is also interesting and all friendly as a public sport and recreation. Swimming is performing in indoor pools in Iran country, approximately. The swimming pool water can be considered as a source of recipient and transmission of a wide range of pathogenic

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microorganisms. These microorganisms may be delivered by a variety of routes including direct excretion by bathers, transport on the body or growth within the filter bed. The swimming pools would become a major health hazard without positive steps to inhibit the existence of these microorganisms.^[1] The health risks associated with swimming in pools is always an importance issue because a large number of people in a limited time use of the pool simultaneously.^[2] Although, water of pools is not considered as drinking water can transmit diseases to humans via accidental ingestion. This occurs especially when the residual chlorine or other disinfectants in water is not enough.^[3] Wide range of people with different levels of economic, social and health use swimming pools. If there is no control over health standards for swimming pools, they can be a serious source of bacterial or fungal contamination.^[4] Many studies showed that absence of adequate disinfect in swimming pools water can lead to that water be considered as an important source of fungal diseases spreading. Presence of some infectious agents, saprophytic fungi and other microorganisms at floor or surfaces of many locations in swimming pools will lead to spread and transmit of certain diseases.^[1,4,5] Nanbkhsh *et al.* evaluated the fungal contamination of four public indoor swimming pools using membrane filtration and carpet sampling method in a period of one year and reported that the common identified fungi from water samples were *Candida* (22.9%), *Rhizopus* (4.16%) and *Aspergillus* (56.2%). In dressing rooms and bathrooms, *Alternaria*, *Cladosporium*, *Philophara* and *Trichophyton mentagrophytis* were isolated.^[4] Also, Shadzi *et al.*^[5] Identified the various fungal species from the studied pools. Identification of microorganisms, especially distribution and density of fungi species, can lead to preventing disease.

There are four indoor swimming pools in Hamedan city. Some individuals with different health level swim in this pools. This study was investigated biological and physico-chemical quality of indoor public swimming pools of Hamedan (Iran).

MATERIALS AND METHODS

In this study, the water samples were collected from four indoor public swimming pools including Laleh, Abiaran, Alvand, Kousar swimming pool of Hamedan city at February to May 2011. The samples were collected with 10 days interval. The sampling were carried out at the weekends; but the time of sampling was selected randomly.

Microbiological sampling

Samples were taken at 30 cm under the water surface with sterilised glass bottles (120 to 480 mL) according to standard methods.^[6] The bottles contain sodium thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3$) to quenching free chlorine residual. The added $\text{Na}_2\text{S}_2\text{O}_3$ in collected samples was provided a

final 100 mg/L concentration. Residual chlorine and pH was measured by colorimetric method and electrical pH meter, respectively. for experiments precision increment, these tests was measured triplicately and presented as Ave \pm Stdev.

Swimming pool contaminations were studied in respect of bacterial and fungal indicators. In order to bacteriological evaluation, the coliform bacterial and fecal streptococcus (FS) was examined according to multiple-tube fermentation technique and the results reported as most probable number (MPN) in 100 mL of sample.

For mycological studies, the sterile bottles with 250 mL volume were used and filled with 100 mL of sample. Other sampling points including foot washing sink, bathrooms, dry sauna rooms, wall and floor of dressing room and pool environment was carried using carpet (a piece of sterilized carpet in the size of 5 \times 5 cm). In soggy location, swab was used for sampling. The carpet and swab was swathed in sterile aluminum foil and transferred to the laboratory. In laboratory, all carpet pieces were shaken over culture media and the swab rubbed inoculating to agar media under a biological hood. For fungal water monitoring, samples were filtrated through Millipore filters with 0.45 micrometer pore size. Filters transferred to three different culture media including Sabouraud Dextrose Agar, Chloramphenicol, Cycloheximide (SCC) and without Cycloheximide (SC). The plates were incubated at 25°C for 3 weeks and examined at frequent intervals for fungal growth and determined by routine laboratory methods, especially slide culture.

RESULTS

The results of physico-chemical parameters in four indoor swimming pool water in Hamadan city showed that the average of temperature was 29.31°C and the minimum and maximum temperature were 27 and 32°C, respectively. As depicted in Table 1, the average of residual chlorine and pH in pools was 0.84 ± 0.52 mg/L and 7.38 ± 3 , respectively. The maximum and minimum of pH were 8 and 7.2 and for residual chlorine were 2 and 0.2 mg/L respectively.

The results of physicochemical parameters for each investigated swimming pool were presented in Table 2. The maximum and minimum of residual chlorine were measured in the Kousar and Alvand pool, respectively. Also, these values for water temperatures in Laleh and Kousar swimming pools were detected.

The results of statistical analysis (correlation coefficient and P. value) are summarized in Table 3. Test results for the incidence of identified fungal species from swimming pools samples are shown in Table 4.

According to the results no fungus was isolated from samples of pools water. Statistical analysis found insignificant relation between residual chlorine and

Table 1: Descriptive statistics results for physico-chemical characteristics of investigated pools

Characteristic	Residual chlorine (mg/l)	pH	Water temperature (°C)	TC MPN index/100 ml	FS MPN index/100 ml
Mean	0.84	7.38	29.31	1.84	0.28
Median	9	7.4	29	0	0
Standard deviation	0.52	0.5	1.28	7.72	1.59
Coefficient of variation	1.8	3	5	43	9
Minimum	0.2	5.2	27	0	0
Maximum	2	8.2	3.2	43	9

Table 2: Mean and standard deviations values of physico-chemical parameters of studied pools

Swimming pools	Parameters			TC MPN index/100 ml	FS MPN index/100 ml
	Water temperature (°C)	pH	Residual chlorine (mg/l)		
Abiaran	28.25±0.71	6.95±0.74	0.78±0.33	1.10±0.38	0
Alvand	28.00±1.31	7.48±0.18	0.59±0.51	15.1±6.50	3.18±1.13
Kousar	20.38±1.30	7.74±0.35	1.30±0.53	1.41±0.50	0
Laleh	29.63±0.74	7.38±0.17	0.70±0.45	0	0

Table 3: Results of the correlation coefficient and P. value between the parameters measured in swimming pools

	pH	FS MPN index/100 ml	TC MPN index/100 ml	Water temperature (°C)	Residual chlorine (mg/l)
pH					
The correlation coefficient	1.00	0.1	0.3	0.34	0.1
P.value		0.8	0.14	0.06	0.8
FS MPN/100 ml					
The correlation coefficient		1.00	0.04	0.2	0.1
P.value			0.8	0.3	0.8
TC MPN/100 ml					
The correlation coefficient			1.00	0.2	0.3
P.value				0.4	0.14
Water temperature (°C)					
The correlation coefficient				1.00	0.1
P.value					0.8
Residual chlorine (mg/l)					
The correlation coefficient					1.00
P.value					

TC: Total Coliform. FS: Fecal Streptococci

Table 4a: Number and percentage of fungal species isolated from swimming pools

Fungus type	Investigated swimming pools							
	Abiaran		Alvand		Kousar		Laleh	
	%	n	%	n	%	n	%	N
<i>Alternaria Spp.</i>	1.9	1	1.8	1	5	3	3.1	2
<i>Aspergillus Spp.</i>	1.9	1	1.8	1	3.3	2	10.8	7
<i>Aspergillus niger Spp.</i>	0.00	0	1.8	1	0.00	0	0.00	0
<i>Cladosporium Spp.</i>	18.5	10	17.5	10	25	15	43.5	12
<i>Penicillium Spp.</i>	1.9	1	7	4	10	6	6.2	4
<i>Phoma Spp.</i>	0.00	0	0.00	0	0.00	0	1.5	1
<i>Rhodotorula Spp.</i>	1.9	1	0.00	0	0.00	0	0.00	0

microbial contamination. Table 4a shows frequency and percentage of fungi species isolated from the sampling points except pool water.

As seen in Table 4b, highest and lowest fungal contaminants were isolated from dressing room and water samples.

DISCUSSION

In this study, the pool water pH was at range of 5.2 to 8.2 and the average pH over the sampling period was 7.83 ± 0.5 . Although, the gap between minimum and maximum values was high but also standard deviation is 0.5 units. According to guidelines, the recommended pH for swimming pool water is 7.2 to 8 ranges.^[7] Low pH of water can result in corrosive nature of water, skin and eye irritation, loss of chlorine, and skin stains in swimmers.^[8] In order to prevent eye irritation, the national standards recommended that in the water pH range from 7.2 to 8, free residual chlorine should be 0.5 to 2 mg/L and at range of pH 7.5 to 7.6, the amount of free residual chlorine should be 0.6 mg/L.^[8] In present study, the mean of free residual chlorine concentration was 0.84 ± 0.52 mg/L, hence it was lower than the national standard value.

The free chlorine consists of hypochlorous acid and hypochlorite ion. The HOCl form of chlorine is stronger disinfectant than OCl-. HOCl formation depends on pH and temperature, so

Table 4b: Prevalence and frequency of fungi isolated from the sampling points of the pools

Fungus type	Sampling locations											
	environment		Foot washing sink		Bathroom floor		Dry sauna rooms		Wall and floor of dressing room		Water	
	%	N	%	n	%	n	%	n	%	n	%	N
<i>Aspergillus spp.</i>	11	2	29	4	5	1	0.00	0	15	4	0.00	0
<i>Alternaria spp</i>	5	1	7	1	5	1	25	1	11	3	0.00	0
<i>Penicillium spp</i>	11	2	14	2	16	3	50	2	22	6	0.00	0
<i>Cladosporium spp</i>	68	13	50	7	74	14	25	1	44	12	0.00	0
<i>Rhodotorula spp.</i>	0.00	0	0.00	0	0.00	0	0.00	0	4	1	0.00	0
<i>Phoma spp</i>	5	1	0.00	0	0.00	0	0.00	0	0.00	0	0.00	0
<i>Aspergillus niger spp.</i>	0.00	0	0.00	0	0.00	0	0.00	0	4	1	0.00	0
Total	100	19	100	14	100	19	100	4	100	27	0.00	0

at pH value of 7.2, the available chlorine in form of HOCl is 65%.^[8] Kargar and colleagues,^[9] in their study reported the amount of residual chlorine was 1.3 mg/L, which is higher than the value obtained in this study. Neghb *et al.*,^[10] reported that 1.052 mg/L for residual chlorine, as well as showed significant relationship between residual chlorine and bacteriological contamination. The average water temperature of swimming pools was 29.31 ± 1.28 that is higher than the standard range (24.5 to 25.5°C).^[12] In this study, the average of MPN was at ranges 0 to 6.5 ± 1.15 MPN/100 mL and it was less than obtained results by Kargar and colleagues study.^[9] The standard value for total coliforms is 500 MPN in 100 mL and the standard value for fecal streptococci is 100 MPN in 100 mL.^[7] Based on the results of biological tests, the amount of total coliforms and fecal streptococci were lower than the standard values.

In this study, fungal contamination of pool water and other locations were investigated. Saprophytic fungi were isolated from the pool except pool water. No isolation saprophytic fungi from the pools water may be due to proper sanitation and health behaviors including use of foot washing sink by swimmers before entrance to pool water. Nanbakhsh *et al.*, and also Dindarloo and colleagues,^[4,11] have mentioned effectiveness improving sanitation and health behaviors in reducing fungal contamination in swimming pools. Nanbakhsh *et al.*, reported that the most common fungi species were *Aspergillus*, *Candida*, dermatophyte fungi, *Mucor*, *Phoma* Spp., *Penicillium*, *Rhizopus* and *Exophiala* species.^[4] In a similar study that was conducted in Isfahan-Iran by Shadzi *et al.*, *Penicillium spp*, *Rhizopus*, *Aspergillus*, yeasts, and fungi and *Trichophyton mentagrophytes* have been identified.^[5] According to a research on swimming pools of Bandar Abbas-Iran,^[11] the most common fungi isolated were *Aspergillus niger* (44.11%), species of yeast (35.8%), *Candida* (33.9%) and other species (21.09%). Other study on fungal contamination of swimming pools,^[12] in Zanjan city of Iran showed that the most isolated fungi were dermatophyte (including *Trichophyton mentagrophytes* and *Epidermophyton floccosum*) 0.4%, yeast 21.8%, filamentous fungi 77.7%, as well as non-saprophytic fungi isolated from the margins of pools, dressing room, shower floor and water were 24.8%, 22.6%, 11.4% and 16.8 %, respectively. In present study

as shown in Table 4, isolated no fungal species from water of pools which it may be due to residual chlorine and this issue has been mentioned by Shadzi and colleagues.^[5] In present study, *Rhodotorula* Spp and *Phoma* spp. were isolated, one time occurrence in Abiaran and Laleh swimming pool, respectively. This maybe is related to people used these swimming pools at a special time and it can be concluded that fungi contamination pattern of swimming pools can be different.

In a survey on fungal flora in mineral swimming pools of Sarein-Iran^[13] fungus species isolated were *Aspergillus fumigatus* 22.79%, *Aspergillus flavus* 15.5%, *Aspergillus niger* 15.54%, and *Penicillium spp* 14.5%, while did not isolate any dermatophytic fungi species from carpet or water samples. In present study highest percentage of fungal species isolated as *Cladosporium spp*.

Generally, the residual chlorine in pools water was lower than standard levels and according to microbial contamination in pool water, it can be concluded that the disinfection system was defective. In our study, bacterial contamination of investigated swimming pools was lower than the standard and fungal contamination of pools was not significant. According to the results presence of bacterial and fungal contamination may be related to hygiene level of pools, as well as physico-chemical parameters especially residual chlorine. In this study, physico-chemical parameters were lower or higher than standard levels, so it is necessary to pay more attention to the physico-chemical parameters, especially residual chlorine, by the managers of swimming pools. In general, monitoring of physico-chemical parameters in swimming pools can play an important role in control of pathogens.

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