

## Effects of seasonal changes and environmental factors on bioindicator bacteria levels in Çardak Lagoon, Çanakkale Strait, Turkey

by

Ibrahim Ender Künili<sup>1,\*</sup>, A. Suat Ateş<sup>2</sup>

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<sup>1</sup>Department of Fishing and Processing Technology, Faculty of Marine Sciences and Technology, Çanakkale Onsekiz Mart University, 17100, Çanakkale, Turkey

<sup>2</sup>Department of Marine Science and Limnology, Faculty of Marine Sciences and Technology, Çanakkale Onsekiz Mart University, 17100, Çanakkale, Turkey

### Abstract

Çardak Lagoon is one of the most important marine environments in the Turkish Strait system, which is home to a variety of organisms. The lagoon is currently under stress and faces the risk of heavy pollution. For this reason, the present state of the lagoon was monitored in this study. During sampling from October 2018 to June 2019, the levels of indicator microorganisms fluctuated up to 4.04 Log<sub>10</sub> cfu 100 ml<sup>-1</sup> and their presence was found to be higher in warmer seasons. The highest positive correlations were observed for total coliform levels with salinity and chemical oxygen demand, whereas the highest negative correlations were found between the levels of fecal coliforms, pH and temperature. *E. coli* and fecal streptococci showed moderate correlations with the environmental factors in all seasons. Although nitrite and nitrate (NO<sub>2</sub> + NO<sub>3</sub>) were not significantly correlated with bacteria levels, they were present at elevated levels. Çardak Lagoon showed the lowest microbiological and chemical quality in the summer season, and this situation continued into the autumn season as a possible result of increased wastewater discharge and human activities. The lagoon should therefore be monitored regularly and precautions should be taken to prevent severe ecological deterioration.

**Key words:** Çardak Lagoon, pollution, bioindicator bacteria, environmental factors, Turkey

\* Corresponding author: [enderkunili@yahoo.com](mailto:enderkunili@yahoo.com)

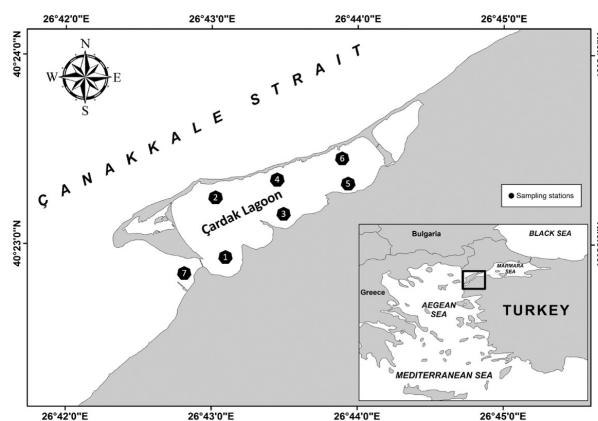
## 1. Introduction

Lagoons are naturally formed shallow waters found in coastal areas of the oceans and separated from the main body of water by sand and patch reefs (Kjerfve 1994). They are important forms in the marine environment as they provide shelter, feeding and breeding grounds for many organisms. However, these unique formations are at risk of deterioration caused by climate change and heavy pollution due to human activities, urbanization and industrialization. These factors are responsible not only for chemical and biological deterioration, but also for changes in the microbial balance of water in these areas (Scialabba 1998; Fiandrino et al. 2003; Yetis & Selek 2015; Freeman et al. 2019; Zoppini et al. 2020).

The microbial environment of seawater is composed of naturally occurring microorganisms and is generally not considered hazardous to the health of humans and marine organisms. However, discharges from various sources, such as sewage, ship ballast, domestic and industrial effluents, can lead to the introduction of fecal-derived microorganisms that pose a threat by altering the microbial ecology of seawater (Mansilha et al. 2009; Cabral 2010). Coliforms and fecal coliforms, along with *E. coli* and fecal streptococci, are widely used to assess environmental pollution and to classify the microbiological quality of seawater (EC 2006; WHO 2018; Anonymous 2019). In general, resistant microorganisms such as fecal streptococci are taken into consideration when monitoring fecal contamination and are often used together with coliforms to determine the origin of bacterial pollution due to their more resistant structure to unfavorable environmental conditions compared to coliform groups (Cabral 2010; Hassou et al. 2014; Rodriguez & Cunha 2017). The presence of fecal contamination in the marine environment can be an indication of a higher risk, such as the presence of other pathogenic microorganisms, e.g. pathogenic bacteria, enteric viruses, or even parasites (Anastasi et al. 2012; Steele et al. 2018; Vega et al. 2021). Therefore, the persistence of these microorganisms in the marine environment is important and not only largely depends on environmental factors such as temperature, salinity, pH, oxygen, and oxidation-reduction potential, but also on organic matter and nutrients, such as nitrogen, carbon, phosphorus, nitrite and nitrate, and ammonia (Cabral 2010; Aylagas et al. 2017; Gilfillan et al. 2018; Pala et al. 2018). These factors determine the fate of microorganisms in the marine environment by increasing their stability or survival (Cho et al. 2016; Pala et al. 2018; Korajkic et al. 2019a). Several studies

have addressed the survival of coliforms and fecal indicator bacteria in coastal conditions over extended periods of time and found that this is mostly related to optimal levels of environmental factors (Menon et al. 2003; Anderson et al. 2005; Ferguson & Signoretto 2011; Hennani et al. 2012; Katarzytė et al. 2018; Korajkic et al. 2019b). However, these conditions, which are affected by various factors such as precipitation, surface runoff from various sources, tides, domestic and industrial discharges, may be more effective in changing the survival characteristics of indicator bacteria in lagoon areas due to the fact that lagoons are semi-isolated and major receiving areas (Perini et al. 2015; Cho et al. 2016; Perkins et al. 2016). Previous studies conducted on the coast and in lagoons of the Turkish seas reported microbiological quality and changes in environmental factors (Kalkan & Altug 2015; Yetis & Selek 2015; Kacar & Omuzbuken 2017; Kalkan & Altug 2020; Zeki et al. 2021). Nevertheless, to our knowledge, this may be the first report analyzing the level of bacterial contamination and its correlation with various environmental parameters in Çardak Lagoon in different seasons.

Çardak Lagoon is an important coastal lagoon located on the southern coast of the Çanakkale Strait, the Marmara Sea, Turkey. A rich natural habitat has existed in and around the lagoon, the condition of which has deteriorated over the years and is now protected by the relevant regulations of the Ministry of Environment and Urbanization of Turkey. Before the deterioration and exposure to pollution, the lagoon supported commercial fish species, such as *Mugil sp.*, *Sparus aurata*, *Pomatomus saltatrix*, *Anguilla sp.* and *Solea sp.*, and commercial bivalve species, such as *Chamelea gallina*, *Flexopecten glaber*, *Mytilus galloprovincialis*, *Ruditapes philippinarum* and *Ostrea edulis*, through fishery and aquaculture (Vural & Acarlı



**Figure 1**  
Study area



2018). For some time now, the lagoon has suffered a marked deterioration reflected in a lack of diversity of organisms and structural changes due to being one of the main receiving coastal sites for surface runoff and discharges from continental sources. As microbiological quality and environmental factors are clear indicators of discharge and pollution in the coastal marine environment, our objective was to determine the levels of indicator microorganisms for microbiological quality and nutrients for chemical quality by comparing the environmental factors and their seasonal relationships.

## 2. Material and methods

### 2.1. Study area

The study area – Çardak Lagoon – is located on the southern coast of the Çanakkale Strait (Fig. 1). The lagoon is the third largest lagoon in the Turkish Strait system, with an area of 1.2 km<sup>2</sup>. The lagoon has a coastline of 4.3 km and an average depth of 1.3 m depth (Caliskan et al. 2011; Cataudella et al. 2015). The study area was divided into seven sampling locations (also referred to as sites), one of which was used as a reference site. The reference site was located outside the lagoon to indicate the quality of water entering the system. The coordinates and depths of the sampling locations are summarized in Table 1.

**Table 1**

Coordinates and maximum depths of the sampling locations

Sites	Coordinates		Max depth (cm)
1	40°22'906"N	26°43'103"E	150
2	40°23'053"N	26°43'264"E	100
3	40°23'203"N	26°43'491"E	150
4	40°23'345"N	26°43'399"E	100
5	40°23'278"N	26°42'988"E	180
6	40°23'236"N	26°42'800"E	150
7 (reference)	40°22'931"N	26°42'768"E	100

### 2.2. Sampling

Water sampling for all analyses was conducted seasonally at the selected sites between October 2018 and June 2019. Samples were collected from the surface water (10–30 cm) at each site using a 1 l sterilized amber glass bottle. The collected water samples were transported to the laboratory at 2–7°C within 4 h.

### 2.3. Analysis of water samples

Physicochemical parameters such as temperature (°C), salinity (PSU), dissolved oxygen (mg l<sup>-1</sup>), pH and oxidation-reduction potential (ORP) were measured in situ using a YSI 650 MDS multiparameter water quality meter.

Chemical oxygen demand (COD) was determined using the open reflux method. Standard methods for the examination of water and wastewater (Eaton & Franson 2005) were used for spectrophotometric determination of nitrite (NO<sub>2</sub><sup>-</sup>), nitrate (NO<sub>3</sub><sup>-</sup>), ammonium (NH<sub>4</sub><sup>+</sup>), phosphate (PO<sub>4</sub><sup>-</sup>) and SiO<sub>2</sub><sup>-</sup> levels (Strickland & Parsons 1972).

Microbiological analyses were performed according to the most probable number (MPN) and plate count methods (FDA 1998; APHA 2005). To count coliforms, peptone-diluted water samples were inoculated in Lauryl Sulfate Tryptose (LST) Broth (Merck) and incubated at 37°C for 20 h. Positive tubes indicating turbidity and gas production were transferred to Brilliant Green Bile Lactose (BGBL) Broth (Merck) and levels of total coliforms (TC) were determined after incubation of inoculated tubes for 36 h at 37°C. Fecal coliforms were assessed after transferring loop full samples from total coliform positive tubes into *Escherichia coli* (EC) broth (Merck) after incubation for 24 h at 44.5°C. *E. coli* levels were determined by dripping Kovac's reagent into EC broth positive tubes. Tubes that changed color from yellow to maroon were *E. coli* positive. Total coliform, fecal coliform, and *E. coli* levels were then determined by the most probable number with a confidence interval of > 95%. Fecal streptococci were determined by the membrane filtration method using a 0.45 µm pore size filter membrane and D-Coccosel agar (Biomerieux; APHA 2005; USEPA 2009). A 100 ml portion of samples was filtered and placed onto the medium and incubated at 37°C for 24 h. Presumed colonies were selected randomly and verified by biochemical tests.

### 2.4. Statistical analysis

Seasonal differences between the sampling locations in terms of bacterial levels and environmental factors were compared using one-way ANOVA. The correlation between the bacterial levels and the factors were determined using Pearson's correlation analysis in Minitab 17 (Minitab, LLC, USA). The significance level for differences between the factors was set at 0.05.

### 3. Results

In this study, total coliform, fecal coliform, *E. coli*, and fecal streptococci levels were monitored seasonally between October 2018 and June 2019. Bacteria levels were related to environmental parameters measured in samples collected from seven locations in Çardak Lagoon, and correlations between the factors affecting the pollution levels were determined.

Values of environmental parameters at seven sites in each season are presented in Table 2.

Levels of bioindicator microorganisms and their threshold levels are presented by site and season in Figure 2.

The levels of detectable total coliform bacteria fluctuated during the study between 1.85 and 4.04  $\log_{10}$  cfu 100 ml<sup>-1</sup>. The highest levels of coliforms were detected at site 3 in the summer season. The level of total coliforms determined in samples collected from site 3 in October 2018 was higher than the hitherto applied national threshold value (4.0  $\log_{10}$  cfu 100 ml<sup>-1</sup>) for bathing water (Anonymous 2006). The minimum and maximum detectable levels of fecal coliforms were in the range of 1.47–4.04  $\log_{10}$  cfu 100 ml<sup>-1</sup>. The highest level of fecal coliforms was also found in samples collected from site 3 in October 2018. The national threshold level for fecal coliforms (3.03  $\log_{10}$  cfu 100 ml<sup>-1</sup>) was also exceeded in these samples (Anonymous 2006). The minimum and maximum detectable *E. coli*

levels were 1.47 and 2.18  $\log_{10}$  cfu 100 ml<sup>-1</sup>, respectively. No *E. coli* was found in samples from sites 5 and 6 in any of the seasons. All *E. coli* levels in all seasons and at all sites were below the national and international threshold level set at 2.69  $\log_{10}$  cfu 100 ml<sup>-1</sup> (EC 2006; WHO 2018; Anonymous 2019). The detectable levels of fecal streptococci were in the range of 1.30–1.85  $\log_{10}$  cfu 100 ml<sup>-1</sup> across the seasons and sites. Not all samples contained fecal streptococci during the winter and spring seasons. The highest contamination was determined in samples from sites 2, 5, and 7 in the summer season, however, the national threshold level set at 2.27  $\log_{10}$  cfu 100 ml<sup>-1</sup> (EC 2006; WHO 2018; Anonymous 2019) was not exceeded in any of the samples.

Changes in bioindicator bacteria and environmental factor levels at the sampling sites and their correlations with all parameters in the autumn season are summarized in Figure 3.

The primary factors affecting the survival of bacterial cells in seawater columns, such as temperature, salinity, pH and O<sub>2</sub>, were determined in the range of 13.2–16.9°C, 20.7–20.9 PSU, 7.85–8.36 and 6.40–9.51 mg l<sup>-1</sup>, respectively. The lowest oxygen level (6.40 mg l<sup>-1</sup>) was determined at site 3, which was characterized by the highest bacterial levels. The correlation between O<sub>2</sub> and bacterial levels, and between temperature (for site 7) and TC and FC were significant ( $p < 0.05$ ), while they were insignificant ( $p > 0.05$ ) with other parameters

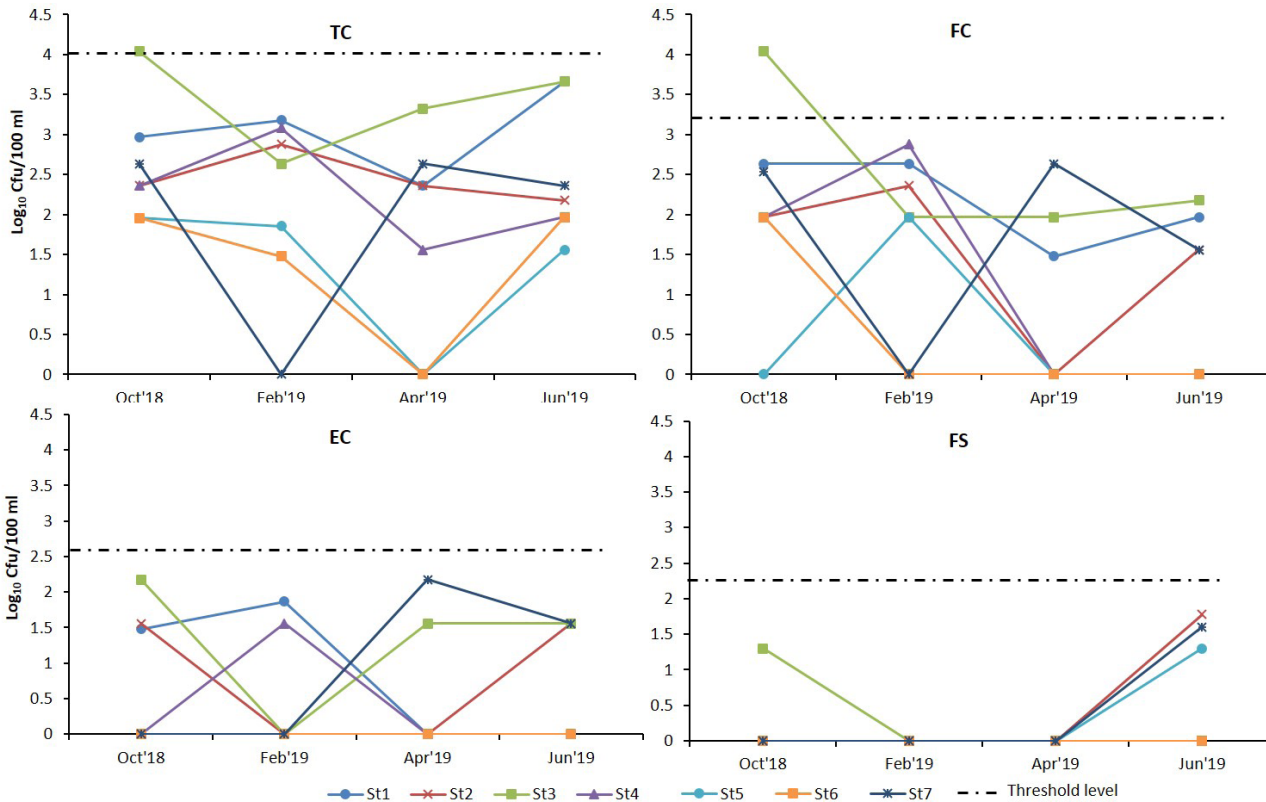
**Table 2**

Summary of environmental data by season

		Temp.	Sal.	pH	O <sub>2</sub> *	COD*	ORP	PO <sub>4</sub> -P*	TP*	NO <sub>2</sub> +NO <sub>3</sub> *	NH <sub>4</sub> *	TN*	SiO <sub>2</sub> *
Oct. 18	Min.	13.25	20.74	7.85	6.40	< 40.00	70.30	< 0.01	0.015	0.014	< 0.01	0.20	0.03
	Max	16.89	20.81	8.35	9.51	277.00	91.00	< 0.01	0.195	0.195	< 0.01	0.99	0.55
	Mean	14.77	20.78	8.14	8.08	197.83	80.41	< 0.01	0.019	0.129	< 0.01	0.61	0.34
	SD	1.15	0.02	0.17	1.13	84.92	7.78	n.a.	0.004	0.061	n.a.	0.32	0.19
Feb. 19	Min.	7.57	22.74	7.78	6.33	125.00	-87.60	< 0.01	0.016	0.060	< 0.01	0.10	0.25
	Max	8.72	23.47	8.27	8.95	296.00	-74.50	< 0.01	0.025	0.120	< 0.01	0.40	0.60
	Mean	7.99	24.40	8.16	7.46	178.86	-83.37	< 0.01	0.018	0.086	< 0.01	0.23	0.39
	SD	0.44	0.62	0.17	0.91	59.18	4.34	n.a.	0.004	0.024	n.a.	0.11	0.14
Apr. 19	Min.	13.7	22.14	8.04	5.90	< 40.00	-99.80	< 0.01	0.020	0.025	< 0.01	0.20	0.05
	Max	17.3	22.78	8.45	8.20	95.00	-78.70	0.030	0.070	0.085	< 0.01	0.60	0.25
	Mean	15.5	22.54	8.31	6.48	80.75	-92.24	0.025	0.040	0.059	< 0.01	0.44	0.17
	SD	1.44	0.22	0.14	0.82	28.54	7.51	0.006	0.021	0.021	n.a.	0.18	0.06
June 19	Min.	24.07	20.17	8.12	7.21	< 40.00	-111.0	< 0.01	0.040	0.015	< 0.01	0.15	0.17
	Max	27.34	22.17	8.56	8.47	106.00	-88.00	0.030	0.170	0.045	< 0.01	0.33	1.50
	Mean	25.67	21.04	8.30	7.84	74.00	-100.61	0.023	0.097	0.029	< 0.01	0.21	0.83
	SD	1.13	0.75	0.15	0.45	24.52	8.54	0.005	0.059	0.010	n.a.	0.06	0.63

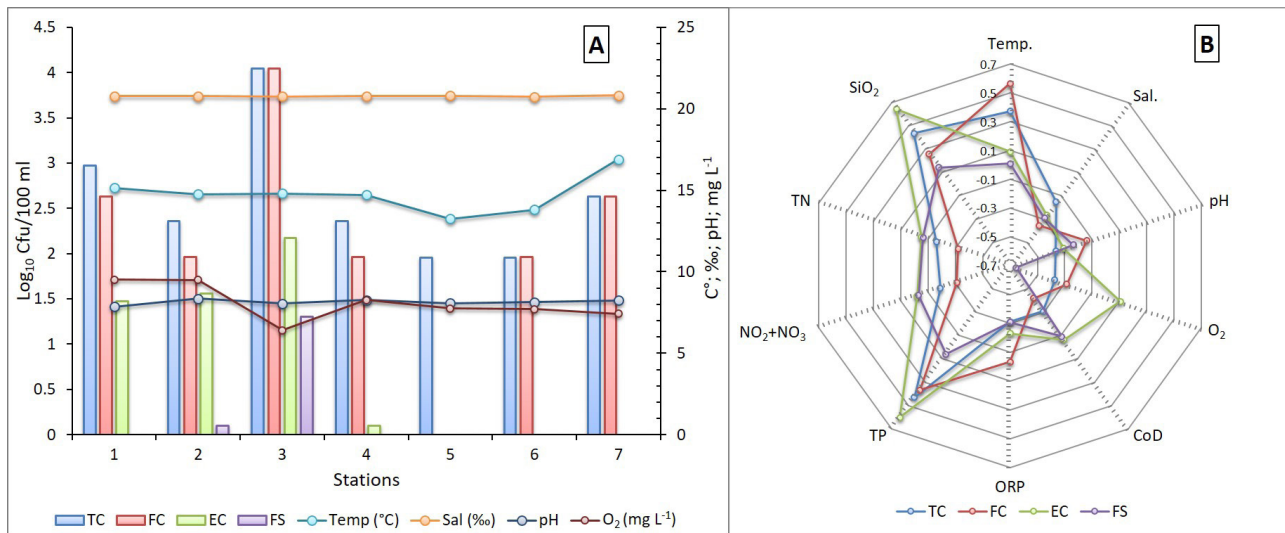
\* – mg l<sup>-1</sup>; n.a. – not applicable; St. – sites; Temp. – temperature (°C); Sal. – salinity (PSU); COD – chemical oxygen demand; ORP (mV) – oxidation-reduction potential; TP – total phosphate; TN – total nitrogen; SD – standard deviation





**Figure 2**

Summary graphs of bioindicator bacteria levels with their national and international threshold levels by season and location



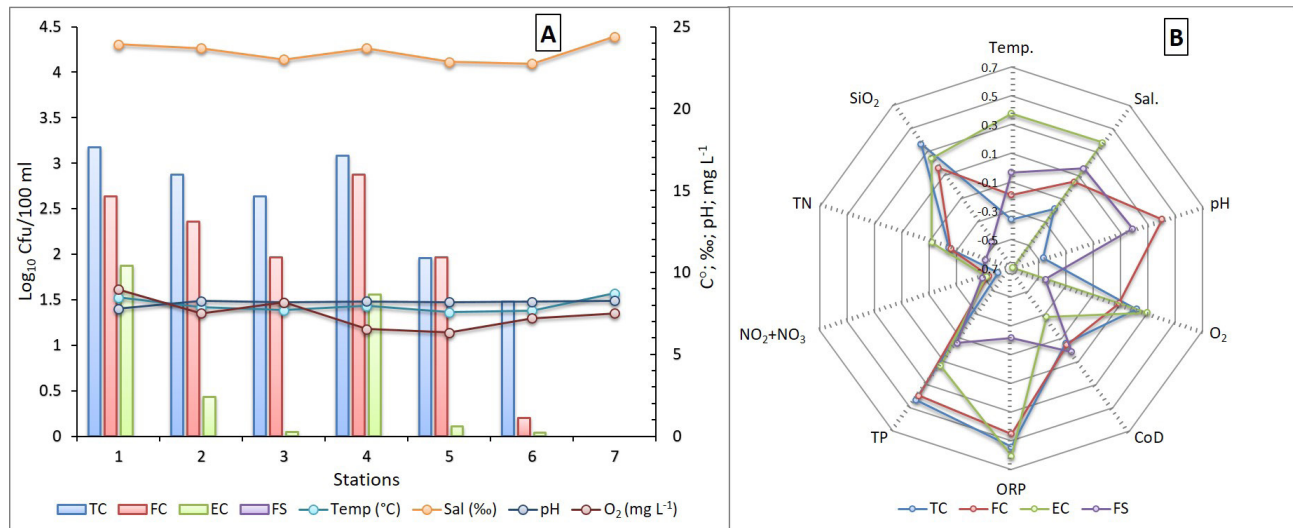
**Figure 3**

(A) Changes in total coliform (TC), fecal coliform (FC), *Escherichia coli* (EC) and fecal streptococci (FS) levels as a function of physicochemical parameters at different sampling locations in the 2018 autumn season. (B) Pearson correlations of environmental parameters with bioindicator bacteria levels for the 2018 autumn season



affecting the bacterial levels. The concentrations of orthophosphate ( $\text{PO}_4\text{-P}$ ), total phosphate (TP), nitrite + nitrate ( $\text{NO}_2 + \text{NO}_3$ ), ammonium ( $\text{NH}_4$ ), total nitrogen (TN) and silicon dioxide ( $\text{SiO}_2$ ) were in the ranges of  $< 0.01$ ,  $0.015\text{--}0.030$ ,  $0.014\text{--}0.135$ ,  $< 0.01$ ,  $0.20\text{--}0.99$ , and  $0.025\text{--}0.50 \text{ mg l}^{-1}$ , respectively. The highest positive and negative correlations between bacterial groups and physicochemical parameters were as follows: total coliforms with  $\text{SiO}_2$  ( $r = 0.435$ ) and  $\text{O}_2$  ( $r = -0.374$ ), fecal coliforms with temperature ( $r = 0.566$ ) and COD ( $r = -0.419$ ), *E. coli* with  $\text{SiO}_2$  ( $r = 0.644$ ) and pH ( $r = -0.308$ ), and fecal streptococci with  $\text{SiO}_2$  ( $r = 0.142$ ) and  $\text{O}_2$  ( $r = -0.655$ ).

Changes in bioindicator bacteria and environmental factor levels at the sampling sites and their correlations with all parameters in the winter season are summarized in Figure 4.



**Figure 4**

(A) Changes in total coliform (TC), fecal coliform (FC), *Escherichia coli* (EC) and fecal streptococci (FS) levels as a function of physicochemical parameters at different sampling locations in the 2019 winter season. (B) Pearson correlations of environmental parameters with bioindicator bacteria levels for the 2019 winter season

Values of physicochemical parameters – temperature, salinity, pH and  $\text{O}_2$  – were in the following ranges:  $7.57\text{--}8.72^\circ\text{C}$ ,  $22.85\text{--}24.40$  PSU,  $7.78\text{--}8.27$  and  $6.33\text{--}8.95 \text{ mg l}^{-1}$ , respectively. Differences between the parameters at the sampling sites and the correlation between the parameters and microorganism levels were insignificant in the winter season ( $p > 0.05$ ). The concentrations of orthophosphate ( $\text{PO}_4\text{-P}$ ), total phosphate (TP), nitrite + nitrate ( $\text{NO}_2 + \text{NO}_3$ ), ammonium ( $\text{NH}_4$ ), total nitrogen (TN), and silicon dioxide ( $\text{SiO}_2$ ) were as follows:  $< 0.01$ ,  $0.016\text{--}0.025$ ,  $0.060\text{--}0.120$ ,  $< 0.01$ ,  $0.10\text{--}0.40$ ,  $0.25\text{--}0.60 \text{ mg l}^{-1}$ , respectively. The highest positive correlations

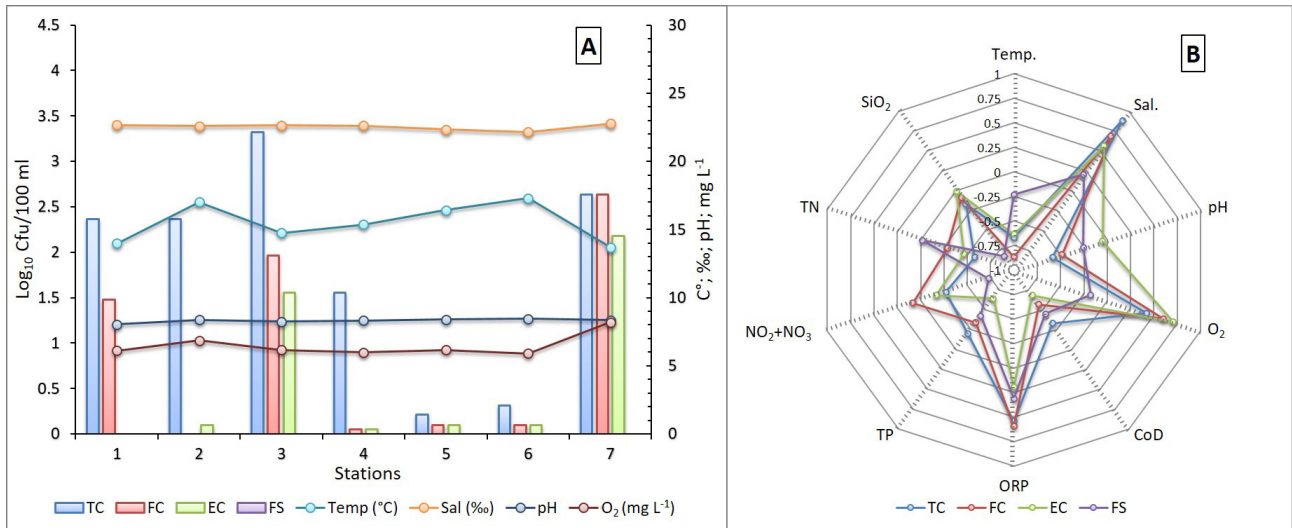
with *E. coli* levels were found for temperature ( $r = 0.377$ ), salinity ( $r = 0.377$ ),  $\text{O}_2$  ( $r = 0.296$ ) and ORP ( $r = 0.607$ ). The highest negative correlations between microorganism levels and nutrients were determined for the concentration of  $\text{NO}_2 + \text{NO}_3$  ( $r: -0.487$  to  $-0.603$ ).

Changes in bioindicator bacteria and environmental factor levels at the sampling sites and their correlations with all parameters in the spring season are summarized in Figure 5.

The values of temperature, salinity, pH and  $\text{O}_2$  were determined as  $13.70\text{--}17.30^\circ\text{C}$ ,  $22.14\text{--}22.78$  PSU,  $8.04\text{--}8.45$  and  $5.90\text{--}8.20 \text{ mg l}^{-1}$ , respectively. With the exception of the  $\text{O}_2$  level at site 7 ( $p < 0.05$ ), differences between the environmental parameters at individual sites and the correlation between the parameters and microorganism levels were found to be insignificant ( $p > 0.05$ ). The highest positive correlations were found

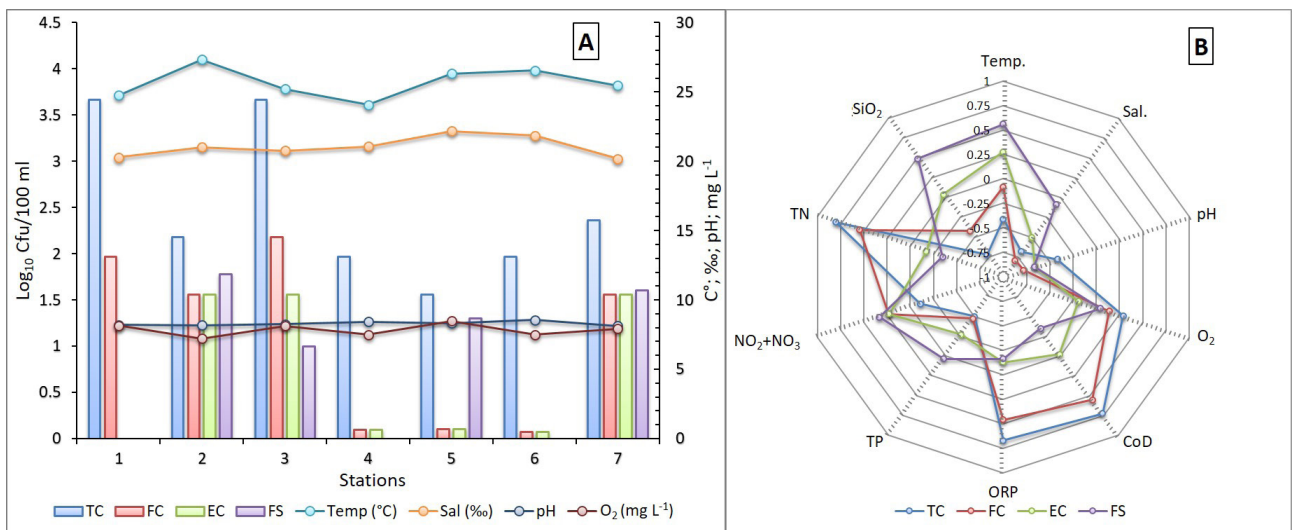
between salinity and total coliforms ( $r = 0.884$ ),  $\text{O}_2$  and *E. coli* ( $r = 0.709$ ), and between ORP and fecal coliforms ( $r = 0.591$ ). The highest negative correlations were determined between temperature and fecal coliforms ( $r = -0.865$ ),  $\text{SiO}_2$  and fecal streptococci ( $r = -0.828$ ), COD and *E. coli* ( $r = -0.685$ ), and between pH and total coliforms ( $r = -0.585$ ). Temperature, pH, TP, and TN were negatively correlated with all groups of bacteria in samples collected in spring 2019 (Fig. 5).

Changes in bioindicator bacteria and environmental factor levels at the sampling sites and their correlations with all parameters in the autumn season are summarized in Figure 6.



**Figure 5**

(A) Changes in total coliform (TC), fecal coliform (FC), *Escherichia coli* (EC) and fecal streptococci (FS) levels as a function of physicochemical parameters at different sampling locations in the 2019 spring season. (B) Pearson correlations of environmental parameters with bioindicator bacteria levels for the 2019 spring season



**Figure 6**

(A) Changes in total coliform (TC), fecal coliform (FC), *Escherichia coli* (EC) and fecal streptococci (FS) levels as a function of physicochemical parameters at different sampling locations in the 2019 summer season. (B) Pearson correlations of environmental parameters with bioindicator bacteria levels for the 2019 summer season

The values of temperature, salinity, pH and  $\text{O}_2$  were determined as 24.07–27.34°C, 20.17–22.17 PSU, 8.12–8.56 and 7.50–8.47  $\text{mg L}^{-1}$ , respectively. Differences between environmental parameters at each site and their correlations with microorganism levels were insignificant ( $p > 0.05$ ). The highest positive correlations were found between COD and TC ( $r = 0.725$ ), ORP and TC ( $r = 0.672$ ), and between temperature and FS ( $r = 0.559$ ). The highest negative

correlations were found between salinity and FC ( $r = -0.794$ ), pH and FC ( $r = -0.781$ ), and between salinity and TC ( $r = -0.682$ ).

#### 4. Discussion

Summer recreational activities in the study area continue through October, provided temperatures

are favorable (Fig. 3). Sites 1, 3 and 5 are located at the shoreline of the lagoon. Therefore, the effects of potential discharges from the shore were observed during the study period, especially at sites 1 and 3. The detection of fecal coliforms and *E. coli* at these sites, along with low  $O_2$  and elevated  $NO_2 + NO_3$  levels at site 3, may be related to these effects. Moreover, the negative correlation between  $O_2$  and elevated levels of coliforms and FS may also indicate wastewater discharge around sites 1 and 3, as most coliforms and FC are facultative anaerobic or microaerophilic microorganisms (Hill 2005; Cabral 2010). Although water circulation is lower at sites 4, 5 and 6 due to the shallow system, the bacterial load was considerably smaller compared to other sites. The main reason for this may be the low exposure to changes and the high level of pollution, especially in warmer months, as these sites are far from sites 1–3 and 7. Except for site 4, however, there were no significant differences in nutrient amounts between the sites during this season ( $p > 0.05$ ). The lowest amounts of nutrients in water samples from site 4 were determined for TP,  $NO_2 + NO_3$ , and TN, i.e.  $0.020 \text{ mg l}^{-1}$ ,  $0.014 \text{ mg l}^{-1}$ , and  $0.20 \text{ mg l}^{-1}$ , respectively. Elevated contamination levels in autumn, compared with mean annual values, were also reported by Avramidis et al. (2017) from Klisova Lagoon in Greece, who recorded TC levels at  $3.95\text{--}5.06 \log_{10} \text{ cfu } 100 \text{ ml}^{-1}$  and FS levels at  $0.66\text{--}4.30 \log_{10} \text{ cfu } 100 \text{ ml}^{-1}$ . The autumn season was also identified as the most polluted season by Kalkan & Altug (2020) based on the presence of fecal streptococci on the coast of the Black Sea, Turkey.

Elevated bacterial contamination was still observed in the winter season at sites 1 through 5. However, fecal contamination was not determined in samples from sites 6 and 7. As previously mentioned, site 7 was the reference site located outside the lagoon. Therefore, the absence of any bacteria may be related to lower temperatures, varied salinity and pH, precipitation, water flows, reduced recreational activity, and marine traffic in the harbor. On the other hand, the continued fecal contamination inside the lagoon may indicate that sewage discharge is an intense and persistent problem in the winter season, despite reduced temperatures. This situation is most likely related to flushing and upwelling effects of both stormwater runoffs and wind effects that may play an important role in transporting additional bacteria into the lagoon. In addition, resuspension of settled bacterial cells from sediment to water may also help these bacteria to maintain prolonged survival via suspended particles (An et al. 2002; Medema et al. 2003; Servais et al. 2007; Hong et al. 2010). Furthermore, the transmission of fecal contamination

at sites 1 and 3 during winter sampling is thought to be related to human-induced coliforms, because fecal streptococci were not detected at these sites, even though they are more resistant to environmental changes (Sinton et al. 1993; Rodriguez & Cunha, 2017). Among the chemical quality indicators, the levels of total phosphate and total nitrogen, which were at higher levels at sites 1 and 3 compared to other sites, indicated that domestic discharge may be concentrated in this part of the lagoon. Moreover, it was observed that a potential discharge may be an important factor in changing the water quality in the winter season, despite the evenly distributed precipitation and stormwater runoff flowing into the lagoon.

In the presented study, the spring was the season with the lowest fecal contamination at all sites except sites 3 and 7. Bioindicator bacteria were not found in samples collected at sites 5 and 6 that are located inside the lagoon and away from sites 1, 3 and 7. Salinity and pH were highly negatively correlated with all bacterial groups in the spring season. The observed negative correlations of pH and salinity with all bacterial groups can be explained by the negative synergic effect of increased surface evaporation and lack of precipitation as a consequence of elevated temperature in the summer season. While the differences in temperature between the seasons were significant ( $p < 0.05$ ), this was not the case for pH ( $p > 0.05$ ). This situation was also pointed out by Hong et al. (2010) who reported that TC and FC levels were closely correlated with pH and temperature in reservoirs. For coliforms, rising temperature may have both a negative effect on the survival and a positive effect on the growth, while nutrients directly positively affect both the survival and growth in the water column (LeChevallier 2003; Juhna et al. 2007; Hong et al. 2010). The highest bacterial pollution was observed in summer. Fecal contamination was determined at sites 2, 3, 5 and 7. The elevated bacterial load in this season may be due to increased recreational activity and wastewater discharge, harbor traffic, elevated temperature suitable for bacterial cells, and lack of precipitation. In addition, the findings also indicate that the origin of fecal contamination is related to sites 1, 3 and 7 and their surroundings. These sites are known to be exposed to sewage discharge, which may result in increased contamination during mild seasons such as autumn and spring. It can also be concluded that site 2 was contaminated with fecal bacteria because it is located close to sites 1 and 7. At site 5, fecal contamination was first observed in the summer season. This site was exposed to some extent to wastewater discharge due to increased human activity





in summer. Moreover, the source of the fecal load at this site may be the water running from site 7 as well as sites 1 and 3, which flows through the inner part of the lagoon along the shoreline.

Lagoons are important ecosystems due to their unique formation in coastal areas. Important studies were conducted in the Mediterranean aimed at monitoring the pollution of these unique systems. Bacterial levels in lagoons can vary from region to region due to climate differences, environmental factors dependent on geographical conditions, discharges, and other uncontrolled factors. In this study, however, coliform levels were similar to those reported by Zafiri et al. (2009) who determined the minimum and maximum levels in Koutavos Lagoon (Kefalonia, Greece) between winter and summer months as 2.0 and 2.65  $\log_{10}$  cfu 100 ml<sup>-1</sup>, respectively. Furthermore, our results were slightly different from the findings of another study conducted at Bizerte Lagoon (Tunisia) in the Southern Mediterranean, where coliform levels in lagoon water from autumn to summer ranged from 1.0 to 2.0  $\log_{10}$  cfu ml<sup>-1</sup> (Boukef et al. 2012). In both of these studies, bacterial levels in lagoon water were affected mainly by environmental factors. In our study, summer was the season most exposed to bacterial pollution. A similar finding was reported by Avramidis et al. (2017), who emphasized that bacterial levels (4.08–4.30  $\log_{10}$  cfu 100 ml<sup>-1</sup>) increased during warmer months due to the lack of precipitation, but were not correlated with months owing to the presence of contamination sources in the lagoon.

## 5. Conclusion

Lagoons are important coastal areas as they provide refuge, breeding and feeding habitats for many marine species. Çardak Lagoon, one of the most important lagoons in the Turkish Strait system, has experienced chemical and bacterial pollution in recent years. In this study, bacterial contamination levels were determined seasonally and the results were related and correlated with environmental factors. It was concluded that the pollution in the lagoon originates mainly from potential human-induced discharges in and/or around the lagoon. Moreover, the pollution varied seasonally depending on changes in environmental parameters, which showed significant correlations. The results show that bacterial pollution in the lagoon is at elevated levels compared with the national and international threshold values, and if it continues at this rate, more severe deterioration of the lagoon is inevitable. For this reason, it is important

that lagoons facing pollution problems, such as Çardak Lagoon, are closely monitored to protect their ecosystems and surrounding environment.

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