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# Environmental characterization of a Mediterranean protected shallow brackish coastal aquatic system, Klisova Lagoon, Western Greece: a case study

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**Abstract** The Klisova lagoon aquatic system belongs to the wetland of Messolonghi - Aetoliko – Klisova Lagoon Complex, located in the western Greece and represents one of the most important Mediterranean lagoon systems, as it is protected by international conventions and is listed in the Natura 2000 European Network. Water physicochemical parameters such as pH, temperature, salinity, dissolved oxygen, nutrients definition, TOC, TN and bacteriological indicators (E.coli and Enterococcus spp.) were analyzed in a time period of 1 year monthly monitoring in five sampling sites along the lagoon. The geographical distribution of these parameters show a clear zonation and partition of the lagoon as the result of: (a) the discharging of poorly treated wastes into the lagoon of Mesolonghi city waste water treatment plant and (b) the interplay between sea water influence via the lagoon inlet and the fresh water inflow via lagoon's perimeter channels. The lagoon is characterized by seasonal hypoxic conditions, which are responsible for several ecological socks in the past including fish mortalities. The system is threatened by human interference and sedimentological processes such as the longshore drifting and siltation of the lagoon inlet. Measurements should immediately be taken in order to prevent further downgrading of the water quality.

**Keywords** Brackish lagoon · Longshore drifting · Nutrients · Total nitrogen · Total organic carbon · Bacterial indicators

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## Introduction

Wetlands are among the most endangered environmental areas. A lot of detrimental impacts have been observed as results of intensive agriculture, aquaculture, industry, and overexploitation of the water resources, pollution due to human activities and urbanization, intense pasturing as well as over-fishing. As a result of the above anthropogenic activities, coastal lagoons are subjected to increased nutrient inputs, eutrophication and subsequent death and decomposition of the increased plant biomass which leads to dystrophic crises in the aquatic systems (Duarte 1995; Caumette et al. 1996).

The presence of organic matter in aquatic systems and liquid wastes has attracted an intensive research interest concerning environmental studies (Hoppe-Jones et al. 2010; Larsen et al. 2011). Thus, Total Organic Carbon (TOC) and Total Nitrogen (TN) are considered as the main factors giving quantitative information for the evaluation of water quality of an ecosystem (Vialle et al. 2011). Bottom sediments can reload the water column with nutrients via decomposition of sediment's organic matter (Golterman 2004). TOC content in water and sediments has been used as an indicator of pollution and eutrophication rate (Environmental Protection Agency, EPA, USA 2002).

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According to the European Directive 2006/7/EC of the European Parliament and of the council of 15 February 2006, concerning the management of bathing water quality and repealing Directive 76/160/EEC adapted by Greek legislation, it should be noted that a sea water sample is considered non comply with human use when it is not in accordance to at least one parameter of the ones mentioned in directives that it is to be free of microorganisms such as Escherichia coli and faecal Enterococci.

## Study area – previous studies

The Klisova lagoon is the south eastern part of the extensive wetland of Messolonghi - Aetoliko – Klisova Lagoon Complex, which is located in the western Greece and represents one of the most important Mediterranean lagoon systems and is the largest in Greece (Fig. 1). The Mesolonghi – Aetoliko Lagoon Complex is lying between the Acheloos and Evinos Rivers deltas. In the Mesolonghi – Aetoliko Lagoon complex, several times in the past anoxic events caused mass fish mortalities and release of hydrogen sulphide (Avramidis et al. 2010, 2015; Koutsodendris et al. 2015; Gianni et al. 2011, 2012, 2013; Chamalaki et al. 2013; Kehayias et al. 2013). The Klisova lagoon was separated by the main Mesolonghi – Aetoliko Lagoon Complex in 1885 by the construction of a road while at late 60s the lagoon was separated artificially in the West Klisova of about 1900 ha and East Klisova of about 600 ha (Fig. 1). It is a very shallow lagoon with maximum and mean depth of 1.5 and 0.5 m respectively, whereas the greatest depths occur in the central parts of the lagoon. Attempts for the drainage of the lagoon were made in the 1950–60s. The eastern Klisova lagoon is connected with the gulf of Patras via an artificial canal of 150 m width and 5000 m length.

The lagoon is of economically and ecologically significance as it is one of older and traditional 'extensive aquaculture' area of Greece. The lagoon is threatened by the malfunction of the waste water treatment plant (WWTP) from the city of Mesolonghi and irrigation water as well as by the limited water renewal as the result of siltation of the lagoon inlet by Evinos delta sediments. Several times in the past, environmental disturbances had taken place that resulted in massive death of fishes. The last ecological shocks took place on December 2006 and July 2008 where massive death of fishes was observed (Avramidis et al. 2010). During the last 20 years, studies have been made describing the environmental status of the Klisova lagoon in relation to sediments heavy metals concentrations (Papatheodorou et al. 2002; Marazioti et al. 2010; Karageorgis et al. 2012), water physicochemical parameters and macrophytes (Hotos and Avramidou 1997; Christia and Papastergiadou 2007; Christia et al. 2011), but without to focus on the East Klisova and specially to its nutrient, organic and bacteriological load which are the objectives of the present study. All the previous studies indicated that the main degradation of the ecosystem is the result of the catchment land use – agriculture activities and the periodical malfunctioning sewage treatment of the city of Mesolonghi. Studies of sediments heavy metal concentration indicate different results due to different methodologies and sampling and treatment procedure (Papatheodorou et al. 2002; Marazioti et al. 2010; Karageorgis et al. 2012). In Table 1 we summarized and present

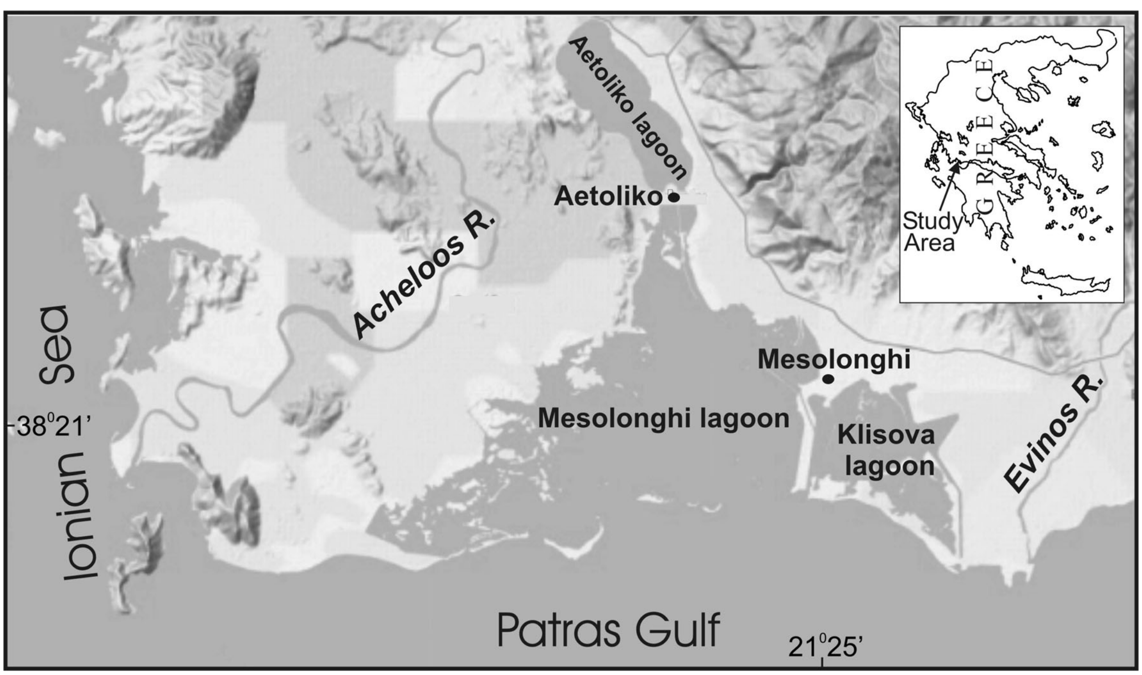


**Fig. 1** (a) Simplified map showing the Mesolonghi – Aetoliko Lagoon Complex and the study area of Klisova Lagoon, as well as the main deltas of Acheloos and Evinos Rivers

**Table 1** Examined environmental and ecological parameters of Mesolonghi - Aetoliko and Kleisova Lagoons complex from previous and present study

| | Sediments geochemistry heavy metals | Water heavy metals - physicochemical | Sediments and water TOC and TN | Ecological indicators macrophytes-zooplankton | Bacterial indicators | Reference |
|---|---|---|---|---|---|---|
| Mesolonghi lagoon | v | | | | | Karageorgis et al. (2012) |
| Aetoliko lagoon | | v | | | | Dassenakis et al. (1994) |
| | | v | | | | Papadas et al. (2009) |
| | | v | | v | | Gianni et al. (2012) |
| | | v | | v | | Kehayias et al. (2013) |
| | | v | v | | | Avramidis et al. (2015) |
| Klisova lagoon | | v | | | | Karageorgis et al. (2012) |
| | | v | v | | | Hotos and Avramidou (1997) |
| | v | | | | | Papatheodorou et al. (2002) |
| | | | | v | | Christia and Papastergiadou (2007) |
| | | | | v | | Christia et al. (2011) |
| | v | | | | | Marazioti et al. (2010) |
| | | v | v | | | Avramidis et al. (2010) |
| | | v | v | | v | This Study |

the existed environmental studies regarding the lagoons of Mesolonghi – Aetoliko and Klisova complex. This study as far as we know is the first complete research including microbiological parameters on the probable causes of this aquatic system water downgrading as well as an analysis of probable preventive actions.

## Materials and methods

One year monthly surveillance (December 2012 – November 2013) was carried out in five stations (S1-S5) in the longitudinal axis of the lagoon in a south to north direction (Fig. 2). Double water samples were collected from each station for

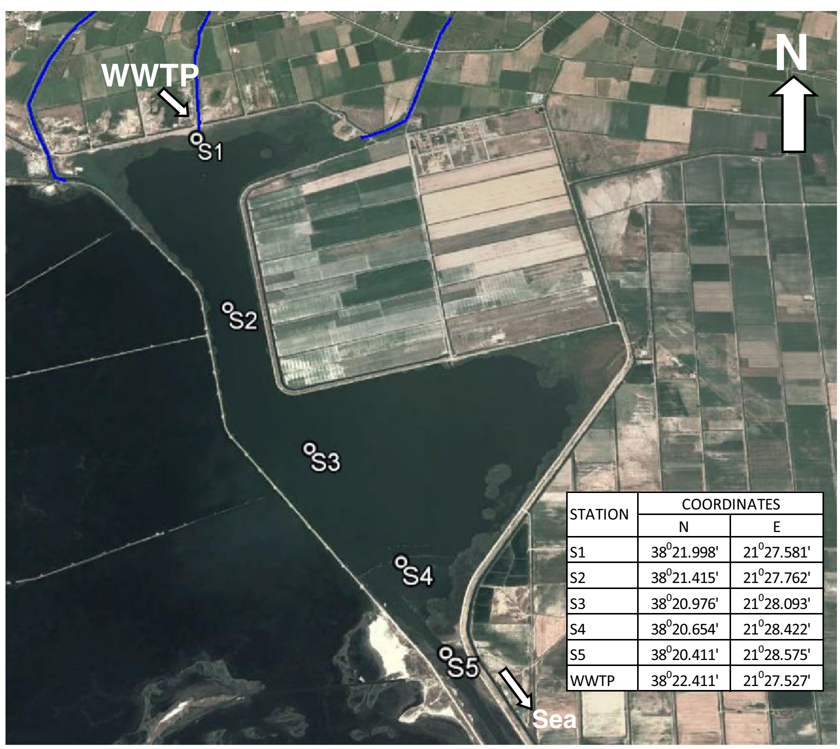


**Fig. 2** General aerial view of the study area, eastern Klisova lagoon, with the location of the water sampling stations (S1-S5), the perimeter channels and the location of the waste water treatment plant (WWTP) of Mesolonghi city

**Table 2** Directive [2006](#)/7/EC of the European Parliament and of the council of 15 February 2006 concerning the management of bathing water quality

| | | | | |
|---|---|---|---|---|
| For coastal waters | | | | |
| 1 | Intestinal enterococci (cfu/100 ml) | 100 ([a]) | 200 ([a]) | 185 ([b]) |
| 2 | Escherichia coli (cfu/100 ml) | 250 ([a]) | 500 ([a]) | 500 ([b]) |
| For inland | | | | |
| | Parameter | Excellent quality | Good quality | Sufficient |
| 1 | Intestinal enterococci (cfu/100 ml) | 200 ([a]) | 400 ([a]) | 330 ([b]) |
| 2 | Escherichia coli (cfu/100 ml) | 500 ([a]) | 1000 ([a]) | 900 ([b]) |

[a] Based upon a 95-percentile evaluation

[b] Based upon a 90-percentile evaluation

physicochemical and bacteriological analysis. Samples are indicating as 'S1', 'S2', 'S3', 'S4' & 'S5' (regional map) in regards to different chosen regional locations, starting from the closest (S1) and ending to the most distant (S5) from the Waste Water Treatment Plant (WWTP) (Fig. 2).

### Physicochemical analysis

The water samples were collected in one liter polypropylene bottles which were previously treated with HCl 5% *w/w*. The samples were transferred within 1 h of collection in the laboratory and were immediately analyzed. pH, salinity (‰), temperature (T, °C) and dissolved oxygen (DO, mg/L) were field measured with a portable instrument HACH HQ40D (Loveland, Colorado USA). Ammonium ions were measured using a HACH DR2800 (Berlin, Germany) absorption spectrophotometer, using HACH cuvette test (LCK), where ammonium was reacted with salicylate and hydrochlorite ions in the presence of sodium nitroprusside as a catalyst and the absorbance of indophenol blue at 690 nm was measured. All anion analyses were carried out using the ICS-1100 integrated ion chromatographic system of Dionex (IC pump which is a microprocessor-based isocratic delivery system, six-port electrically activated injection valve, precolumn and separator column including chemical suppressor device assembly). Before running a sample the ion chromatography system is calibrated using a Dionex standard solution. By comparing the data obtained from the sample to that obtained from the known standard, sample anions are identified and quantitated automatically. Total Organic Carbon (TOC) and total nitrogen (TN) were measured using a Schimadzu TOC analyzer (TOC-VCSH) coupled to a chemiluminescence detector (TNM-1 TN unit) (APHA 2005; ASTM D5176 2008; Bekiari and Avramidis 2014).

### Bacteriological analysis

All the water samples were analyzed according ISO standard methods for the detection of Total coliforms (ISO 9308–1:2000), E. coli (ISO 9308–1:2000), and Enterococcus spp. (ISO 7899–02:2000). The results were compared to the reference criteria contained in Directive 2006/7/EC of the European Parliament and of the council of 15 February 2006 concerning the management of bathing water quality and Directive 2009/54/EC of the European Parliament and the Council of the European

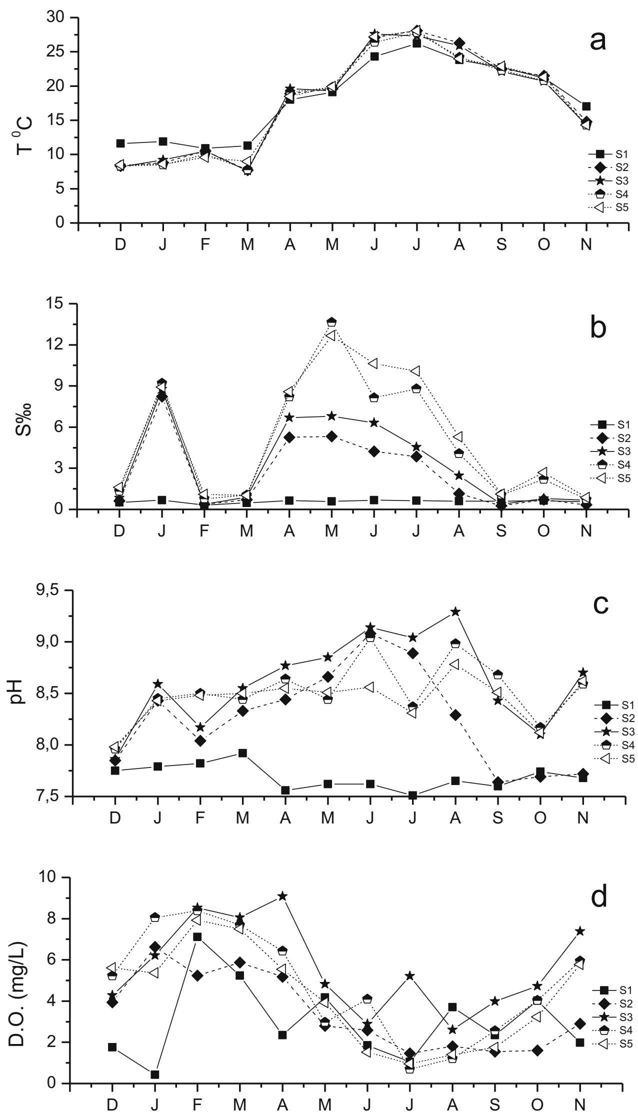


**Fig. 3** Monthly values of **a** temperature, **b** salinity, **c** pH and **d** dissolved oxygen in monitoring stations S1-S5

Union (2009) on the exploitation and marketing of seawater quality (Table 2).

### Statistical analysis

The microbiological and chemical results were evaluated using IBM SPSS 21.0 Pearson correlation and analysis of variance (ANOVA). Statistical comparisons of water quality between geographical stations and months were performed. The results were compared to the reference criteria contained in European Directive on the sea water quality.

## Results and discussion

### Physicochemical analysis

Due to the shallow lagoon depth, the monthly water temperature from all the monitoring stations (S1-S5), was close to the atmospheric temperatures around the year and varied from 7.6 to 28.1 °C with a mean value of 17.7 °C (Fig. 3a). Salinity values ranged from 0.27 to 13.63‰ (Fig. 3b) with a mean value of 3.35‰. In summer, due to the very low fresh water input, the highest salinity values were observed. The spatial distribution of salinity indicates relatively increased values in stations S5 and S4 as they are located proximal to the lagoon inlet and were influenced by the sea water intrusion and tide, while a gradual salinity decrease was observed from south to north. Station S1 which is located in the northern part of the lagoon, proximal to the channel which discharge fresh water into the lagoon, salinity values were below <1‰ during the monitoring year. A clear salinity zonation and partition of the lagoon in the north (low salinity) and south (high salinity) part were recorded as the result of the interplay and mixing between the fresh water influx via the north perimeter channels and the sea water intrusions via the lagoon inlet and tide activity. This interplay makes the eastern Klisova a typical example of a brackish coastal lagoon aquatic system.

The water pH ranges between 7.5 to 9.2 (with a mean value of 8.3) and, in general, had similar trends to all the stations around the year, with exception station S1 where values were slightly lower (Fig. 3c). Additionally, during autumn relative lower pH values were recorded in station S2 (Fig. 3c) indicating a partition of the lagoon in south part with relatively higher and north part with lower pH values.

Dissolved oxygen ranged between 0.42 and 9.08 mg/L indicating in general seasonal poor oxygenation and around saturation conditions with a mean value of 4.21 mg/L (Fig. 3d). Relatively lower DO values were observed from May until October, while during summer the lagoon is characterized by hypoxic conditions with DO concentrations <2 mg/L (Fig. 3d).

The monthly nutrients fluctuation (nitrates, nitrites, ammonium ions and phosphates) for the five monitoring stations (S1-S5) are presented in Table 3. The presence of nitrates was confirmed during winter in all monitoring stations while it was below the detection limit in several months during spring, summer and fall (Table 3). The mean seasonal concentration of nitrates varied from 0.026 to 0.42 mg/L, while the

**Table 3** Seasonal results of physicochemical analysis for nitrates, nitrites, ammonium ions and phosphates for monitoring stations S1-S5

| | | S1 | | | S2 | | | S3 | | | S4 | | | S5 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | min | max | aver | min | max | aver | min | max | aver | min | max | aver | min | max | aver |
| Winter | $NO_3^-$ (mg/L) | 0.05 | 2.54 | 1.21 | 0.13 | 0.48 | 0.25 | 0.14 | 0.42 | 0.24 | 0.15 | 0.29 | 0.2 | 0.1 | 0.25 | 0.2 |
| | $NO_2^-$ (mg/L) | <0.01 | <0.01 | <0.01 | <0.01 | 0.05 | 0.03 | <0.01 | 0.09 | 0.04 | <0.01 | 0.16 | 0.06 | <0.01 | 0.19 | 0.07 |
| | $NH_4^+$(mg/L) | 0.22 | 1.27 | 0.68 | 0.47 | 2.42 | 1.39 | 0.5 | 1.9 | 1.31 | 0.22 | 1.5 | 1.06 | 0.24 | 2.12 | 1.31 |
| | $PO_4^{3-}$ (mg/L) | <0.01 | 0.24 | 0.09 | <0.01 | 0.05 | 0.03 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | 2.63 | 0.88 |
| Spring | $NO_3^-$ (mg/L) | 0.11 | 0.43 | 0.23 | 0.07 | 0.44 | 0.24 | <0.01 | 0.25 | 0.14 | <0.01 | 0.17 | 0.1 | <0.01 | 0.3 | 0.17 |
| | $NO_2^-$ (mg/L) | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 |
| | $NH_4^+$(mg/L) | 7.55 | 10.05 | 8.7 | 0.07 | 2.91 | 1.23 | 0.07 | 0.63 | 0.44 | 0.06 | 3.87 | 1.43 | <0.01 | 3.37 | 1.27 |
| | $PO_4^{3-}$ (mg/L) | <0.01 | 2.86 | 1.22 | <0.01 | 0.3 | 0.12 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 |
| Summer | $NO_3^-$ (mg/L) | <0.01 | 0.09 | 0.04 | <0.01 | 0.07 | 0.03 | <0.01 | 0.07 | 0.03 | <0.01 | 0.08 | 0.03 | <0.01 | <0.01 | <0.01 |
| | $NO_2^-$ (mg/L) | <0.01 | <0.01 | <0.01 | <0.01 | 0.48 | 0.17 | <0.01 | <0.01 | <0.01 | <0.01 | 0.03 | 0.02 | <0.01 | 0.15 | 0.06 |
| | $NH_4^+$(mg/L) | 10.7 | 13.48 | 12.15 | 1.14 | 1.57 | 1.36 | 0.56 | 1.41 | 0.96 | 0.39 | 2.63 | 1.34 | 0.50 | 2.66 | 1.42 |
| | $PO_4^{3-}$ (mg/L) | <0.01 | 0.51 | 0.2 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 |
| Fall | $NO_3^-$ (mg/L) | <0.01 | 0.05 | 0.03 | 0.03 | 0.04 | 0.03 | <0.01 | 0.04 | 0.03 | <0.01 | 0.04 | 0.03 | <0.01 | 0.03 | 0.02 |
| | $NO_2^-$ (mg/L) | 0.03 | 0.04 | 0.03 | <0.01 | <0.01 | <0.01 | 0.03 | 0.08 | 0.06 | <0.01 | 0.15 | 0.06 | 0.11 | 0.41 | 0.22 |
| | $NH_4^+$(mg/L) | 4.82 | 9.44 | 6.59 | 0.35 | 1.56 | 1.1 | 0.74 | 1.3 | 1 | 0.77 | 1.21 | 1.04 | 0.61 | 1.3 | 1 |
| | $PO_4^{3-}$ (mg/L) | <0.01 | 1.35 | 0.49 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 |

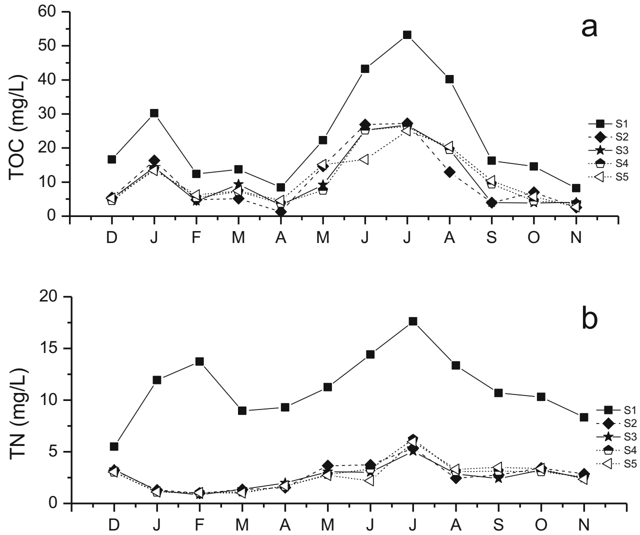


**Fig. 4** Monthly values of: **a** total organic carbon (TOC) and **b** total nitrogen (TN) in monitoring stations S1-S5

mean seasonal concentrations for winter, spring, summer and autumn were 0.42, 0.176, 0.026 and 0.028 mg/L respectively, indicating that the highest presence of nitrates takes place during winter (Table 3). Nitrites were recorded during fall, winter and summer with relatively very low values and were almost absent in spring (<DI) in all monitoring stations (Table 3). Ammonium ions were present to all monitoring stations during the whole year reaching the highest values during summer (13.48 mg/L station S1) while the seasonal mean and the lowest one during winter (0.22 mg/L). Phosphates were recorded in few monitoring stations only in winter and fall and it was the limited nutrient (Table 3).

In Fig. 4a and b, we present the monthly fluctuation of TOC and TN that were measured during the 1 year monitoring, while their geographical distribution is presented in Fig. 5a and b, respectively. TOC values ranged from 1.26 to 53.80 mg C/L with an average 13.9 mg C/L. The higher values were observed during July and the lower on April and November (Fig. 4a). The highest concentrations were measured in station S1 while a gradual decrease in TOC concentration was observed moving from station S1 (near the WWTP) towards the sea station S5 (Fig. 5a and b). This uniform decrease of TOC from station S1 to station S5 was recorded during the whole monitoring year as it was systematically observed in all months. The total nitrogen (TN) values ranged from 0.86 to 17.61 mg N/L with an average 4.2 mg N/L (Fig. 4b). The geographical distributions of TN as well as the monthly TN fluctuations reveal similar trends with TOC (Fig. 5a and b). Concerning the seasonal distribution the higher TOC and TN values were observed during summer (Fig. 4a). This can be explained by the possible dilution of pollutants in winter because of increased rain water flowing into the lagoon. Moreover, the fact that in summer the local population increases the use of water and as a result there are higher rates of discharge can also be related to the higher organic load measured during summer months. The above described spatial distribution and the high values for both

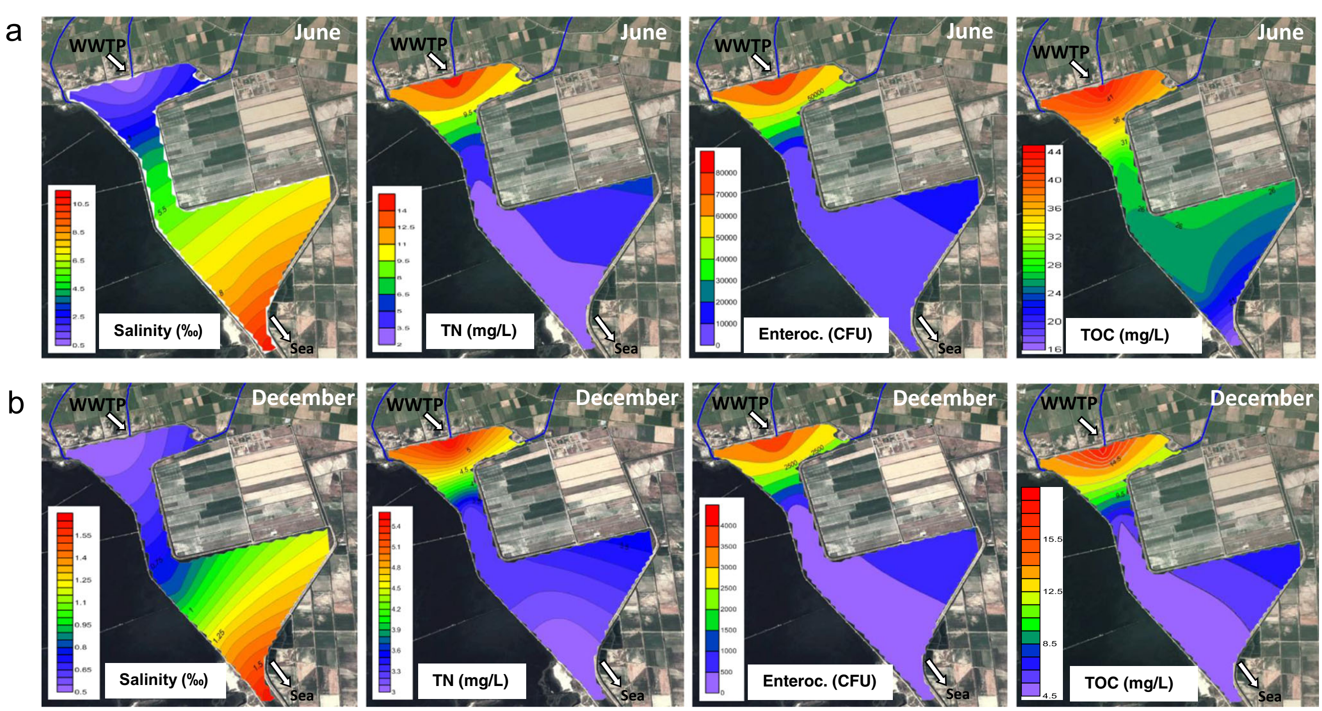


**Fig. 5** Representative geographical distribution of lagoon water properties salinity, total nitrogen (TN), Enterococci and total organic carbon (TOC) for **a** June and **b** December

**Table 4** Descriptive statistics of Log/cfu of Enterococci and total coliforms

| | Sampling stations | N | Mean | Std. deviation | Std. error | 95% confidence interval for mean | | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Lower bound | Upper bound | | |
| LOGEN | 1 | 12 | 4.198 | 0.654 | 0.188 | 3.782 | 4.614 | 3.23 | 4.90 |
| | 2 | 12 | 2.760 | 0.320 | 0.092 | 2.556 | 2.963 | 2.40 | 3.31 |
| | 3 | 10 | 2.154 | 0.564 | 0.178 | 1.750 | 2.558 | 1.30 | 2.91 |
| | 4 | 11 | 2.118 | 0.568 | 0.171 | 1.737 | 2.500 | 1.00 | 2.94 |
| | 5 | 12 | 1.905 | 0.553 | 0.159 | 1.553 | 2.256 | 1.00 | 2.98 |
| LOGTC | 1 | 12 | 5.256 | 0.155 | 0.044 | 5.157 | 5.355 | 4.76 | 5.30 |
| | 2 | 12 | 4.217 | 0.195 | 0.056 | 4.093 | 4.342 | 3.87 | 4.56 |
| | 3 | 12 | 3.773 | 0.760 | 0.219 | 3.290 | 4.256 | 1.78 | 4.30 |
| | 4 | 12 | 3.700 | 0.708 | 0.204 | 3.250 | 4.150 | 2.30 | 4.30 |
| | 5 | 12 | 3.534 | 0.845 | 0.244 | 2.996 | 4.071 | 1.70 | 4.30 |

TOC and TN in the northern part of the lagoon, can be explained mainly by a point source of pollution specifically by the central channel where the WWTP discharged its load, as well as the water discharged by the peripheral channels in this part of the lagoon. On the other hand, in the southern part of the lagoon the interplay between the sea via the lagoon inlet leads to dilution of pollutants and thus to lower TOC and TN values (Fig. 5a and b).

**Table 5** Descriptive statistics of Log/cfu of Enterococci and total coliforms (per month)

| | Month | N | Mean | Std. deviation | Std. error | 95% confidence interval for mean | | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Lower bound | Upper bound | | |
| LOGEN | 1 | 3 | 2.590 | 1.650 | 0.952 | −1.508 | 6.689 | 1.00 | 4.29 |
| | 2 | 5 | 2.571 | 0.484 | 0.216 | 1.970 | 3.173 | 1.95 | 3.23 |
| | 3 | 5 | 2.615 | 1.252 | 0.560 | 1.060 | 4.171 | 1.48 | 4.65 |
| | 4 | 4 | 2.267 | 1.297 | 0.648 | 0.202 | 4.333 | 1.00 | 3.47 |
| | 5 | 5 | 2.431 | 1.078 | 0.482 | 1.092 | 3.771 | 1.48 | 4.11 |
| | 6 | 5 | 2.885 | 1.190 | 0.532 | 1.407 | 4.363 | 1.85 | 4.90 |
| | 7 | 5 | 2.662 | 1.267 | 0.566 | 1.088 | 4.236 | 1.48 | 4.83 |
| | 8 | 5 | 3.192 | 0.930 | 0.416 | 2.037 | 4.348 | 2.36 | 4.76 |
| | 9 | 5 | 2.476 | 1.469 | 0.656 | 0.652 | 4.300 | 1.30 | 4.86 |
| | 10 | 5 | 2.969 | 0.906 | 0.405 | 1.844 | 4.094 | 2.15 | 4.46 |
| | 11 | 5 | 2.450 | 0.676 | 0.302 | 1.610 | 3.291 | 1.48 | 3.23 |
| | 12 | 5 | 2.617 | 0.565 | 0.252 | 1.916 | 3.319 | 2.11 | 3.59 |
| LOGTC | 1 | 5 | 3.015 | 1.578 | 0.705 | 1.055 | 4.975 | 1.70 | 5.30 |
| | 2 | 5 | 4.006 | 0.478 | 0.213 | 3.413 | 4.600 | 3.46 | 4.76 |
| | 3 | 5 | 4.057 | 0.852 | 0.381 | 2.998 | 5.115 | 3.32 | 5.30 |
| | 4 | 5 | 3.505 | 1.195 | 0.534 | 2.021 | 4.989 | 2.49 | 5.30 |
| | 5 | 5 | 3.949 | 0.794 | 0.355 | 2.964 | 4.935 | 3.30 | 5.30 |
| | 6 | 5 | 3.808 | 0.987 | 0.441 | 2.582 | 5.035 | 2.90 | 5.30 |
| | 7 | 5 | 4.425 | 0.493 | 0.220 | 3.813 | 5.038 | 4.13 | 5.30 |
| | 8 | 5 | 4.501 | 0.447 | 0.200 | 3.945 | 5.056 | 4.30 | 5.30 |
| | 9 | 5 | 4.501 | 0.447 | 0.200 | 3.945 | 5.056 | 4.30 | 5.30 |
| | 10 | 5 | 4.501 | 0.447 | 0.200 | 3.945 | 5.056 | 4.30 | 5.30 |
| | 11 | 5 | 4.501 | 0.447 | 0.200 | 3.945 | 5.056 | 4.30 | 5.30 |
| | 12 | 5 | 4.382 | 0.548 | 0.245 | 3.701 | 5.062 | 3.83 | 5.30 |

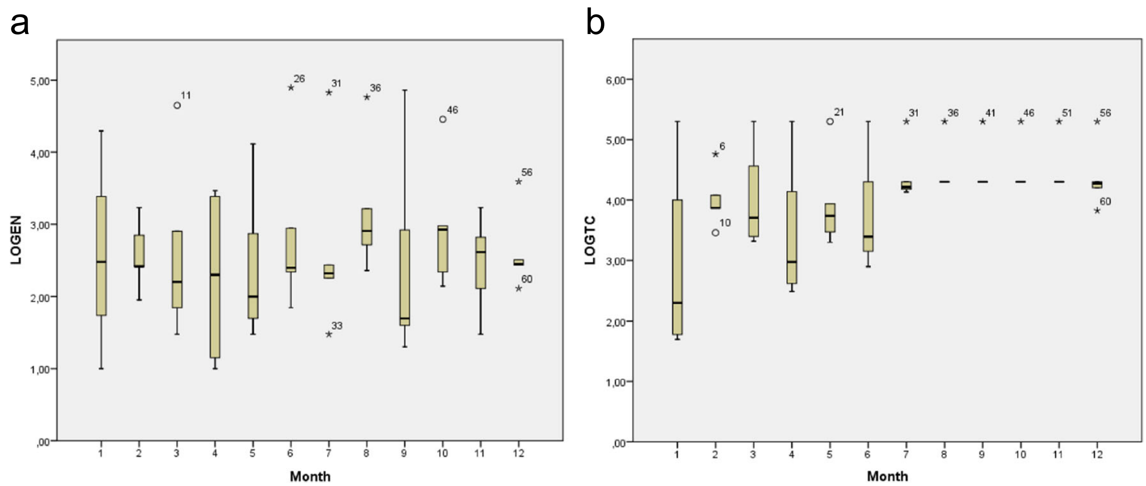


**Fig. 6** Monthly $\text{Log}_{10}$ of: **a** Enterococci and **b** E.coli in Klisova lagoon

## Bacteriological analysis

The individual, monthly, yearly, fluctuating values of Enterococci and E.coli collected from all five sampling stations have been recorded (Tables 4 and 5). Stations 1 and 2 areas were not complied with EU Bathing Water 2006/7/EC and 2009/54/EC Directives of the European Parliament and the Council of the European Union and considered of poor quality concerning enterococci limits for both inland and coastal water limits. Also, as it concerns monthly variation, in 8 of the 12 months, the enterococcal counts were exceeding the Directives limits. Our data proves the stable presence of human fecal material in lagoon environment as enterococci are considered as a common human fecal indicator.

Seasonal variation, during winter months, mean numbers of E.coli ranged from the upper 12.450 cfu/100 ml (*S1*) to a lower 3.206 cfu/100 ml (*S5*). Mean values of both bacterial indicators, were significantly decreased from December to February, with mean values ranging from 15.415 cfu/100 ml to a lower 7.405 cfu/ml respectively (Fig. 6). During spring time, bacterial counts fluctuate from a very high 19.663 cfu/ml (S1) down to 1.460 cfu/ml (S5) whereas during summer, probably due to absence of rainfalls, mean levels fluctuate from 20.000 cfu/ml (S1) to 12.023 cfu/ml (S5). An important finding is a constant high mean value of a 20.000 cfu/100 ml observed during fall, regardless of the sampling location although there is no significant variation in bacteriological load among the months ($p > 0.05$). Enterococci were more stable as their numbers were not correlated with month ($r^2 = 0{,}085$, $p > 0.05$). Total coliform numbers were slightly correlated with the month ($r^2 = 0.431$, $p < 0.05$). In 8 of the 12 months, the enterococcal counts were exceeding the Directive's higher limits.

In each individual month, it is clearly demonstrated a significant reduction ($p < 0.01$) in Total coliforms numbers as we move on away from the heavily polluted sampling site (S1) to

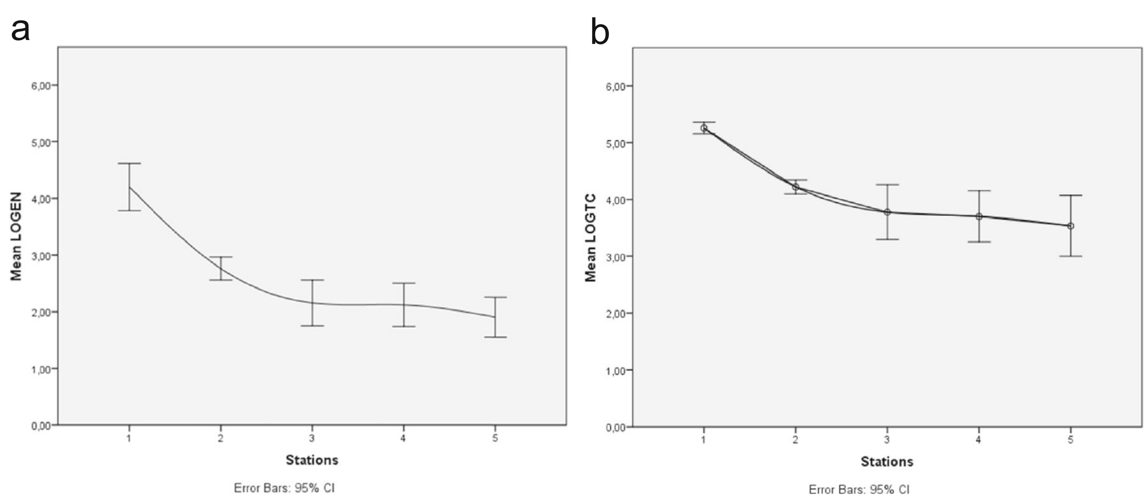


**Fig. 7** **a** Mean Log CFU and **b** Total coliforms per sampling station S1-S5

**Table 6** Correlations of physicochemical and bacteriological parameters in relation to the distance from the WWTP

| | | Distance | pH | T | TOC | S‰ | DO | $NO_3$ | $NO_2$ | $PO_4$ | $NH_4$ | TN | TC | EN | LOGTC | LOGEN |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Distance | Pearson Correlation | 1 | .594** | −.031 | −.352** | .467** | .262* | −.281* | .195 | −.227 | −.579** | −.645** | −.778** | −.561** | −.690** | −.800** |
| | Sig. (2-tailed) | | .000 | .811 | .006 | .000 | .043 | .029 | .136 | .081 | .000 | .000 | .000 | .000 | .000 | .000 |
| pH | Pearson Correlation | .594** | 1 | .239 | −.041 | .490** | .208 | −.211 | .090 | −.265* | −.555** | −.558** | −.665** | −.502** | −.553** | −.624** |
| | Sig. (2-tailed) | .000 | | .065 | .755 | .000 | .111 | .106 | .492 | .041 | .000 | .000 | .000 | .000 | .000 | .000 |
| T | Pearson Correlation | −.031 | .239 | 1 | .507** | .244 | −.669** | −.298* | .119 | −.064 | .165 | .352** | .083 | .199 | .198 | .117 |
| | Sig. (2-tailed) | .811 | .065 | | .000 | .060 | .000 | .021 | .365 | .625 | .208 | .006 | .527 | .127 | .130 | .386 |
| TOC | Pearson Correlation | −.352** | −.041 | .507** | 1 | .114 | −.491** | −.096 | −.078 | −.016 | .569** | .720** | .472** | .613** | .299* | .499** |
| | Sig. (2-tailed) | .006 | .755 | .000 | | .384 | .000 | .467 | .554 | .906 | .000 | .000 | .000 | .000 | .020 | .000 |
| S‰ | Pearson Correlation | .467** | .490** | .244 | .114 | 1 | −.029 | −.135 | −.075 | −.187 | −.313* | −.269* | −.421** | −.269* | −.664** | −.422** |
| | Sig. (2-tailed) | .000 | .000 | .060 | .384 | | .828 | .304 | .569 | .153 | .015 | .038 | .001 | .037 | .000 | .001 |
| DO | Pearson Correlation | .262* | .208 | −.669** | −.491** | −.029 | 1 | .077 | −.115 | .002 | −.209 | −.512** | −.352** | −.245 | −.450** | −.329* |
| | Sig. (2-tailed) | .043 | .111 | .000 | .000 | .828 | | .559 | .380 | .985 | .109 | .000 | .006 | .060 | .000 | .012 |
| $NO_3$ | Pearson Correlation | −.281* | −.211 | −.298* | −.096 | −.135 | .077 | 1 | −.137 | .047 | −.109 | −.072 | .225 | −.052 | .154 | .158 |
| | Sig. (2-tailed) | .029 | .106 | .021 | .467 | .304 | .559 | | .298 | .722 | .406 | .583 | .084 | .695 | .240 | .241 |
| $NO_2$ | Pearson Correlation | .195 | .090 | .119 | −.078 | −.075 | −.115 | −.137 | 1 | −.105 | −.154 | −.083 | −.134 | −.114 | .044 | −.104 |
| | Sig. (2-tailed) | .136 | .492 | .365 | .554 | .569 | .380 | .298 | | .426 | .239 | .531 | .308 | .385 | .740 | .443 |
| $PO_4$ | Pearson Correlation | −.227 | −.265* | −.064 | −.016 | −.187 | .002 | .047 | −.105 | 1 | .419** | .265* | .379** | .240 | .213 | .255 |
| | Sig. (2-tailed) | .081 | .041 | .625 | .906 | .153 | .985 | .722 | .426 | | .001 | .041 | .003 | .065 | .102 | .056 |
| NH4 | Pearson Correlation | −.579** | −.555** | .165 | .569** | −.313* | −.209 | −.109 | −.154 | .419** | 1 | .826** | .781** | .833** | .538** | .700** |
| | Sig. (2-tailed) | .000 | .000 | .208 | .000 | .015 | .109 | .406 | .239 | .001 | | .000 | .000 | .000 | .000 | .000 |
| TN | Pearson Correlation | −.645** | −.558** | .352** | .720** | −.269* | −.512** | −.072 | −.083 | .265* | .826** | 1 | .883** | .825** | .685** | .782** |
| | Sig. (2-tailed) | .000 | .000 | .006 | .000 | .038 | .000 | .583 | .531 | .041 | .000 | | .000 | .000 | .000 | .000 |
| TC | Pearson Correlation | −.778** | −.665** | .083 | .472** | −.421** | −.352** | .225 | −.134 | .379** | .781** | .883** | 1 | .741** | .741** | .830** |
| | Sig. (2-tailed) | .000 | .000 | .527 | .000 | .001 | .006 | .084 | .308 | .003 | .000 | .000 | | .000 | .000 | .000 |
| EN | Pearson Correlation | −.561** | −.502** | .199 | .613** | −.269* | −.245 | −.052 | −.114 | .240 | .833** | .825** | .741** | 1 | .506** | .764** |
| | Sig. (2-tailed) | .000 | .000 | .127 | .000 | .037 | .060 | .695 | .385 | .065 | .000 | .000 | .000 | | .000 | .000 |
| LOGTC | Pearson Correlation | −.690** | −.553** | .198 | .299* | −.664** | −.450** | .154 | .044 | .213 | .538** | .685** | .741** | .506** | 1 | .787** |
| | Sig. (2-tailed) | .000 | .000 | .130 | .020 | .000 | .000 | .240 | .740 | .102 | .000 | .000 | .000 | .000 | | .000 |
| LOGEN | Pearson Correlation | −.800** | −.624** | .117 | .499** | −.422** | −.329* | .158 | −.104 | .255 | .700** | .782** | .830** | .764** | .787** | 1 |
| | Sig. (2-tailed) | .000 | .000 | .386 | .000 | .001 | .012 | .241 | .443 | .056 | .000 | .000 | .000 | .000 | .000 | |

*Correlation is significant at the 0.05 level (2-tailed)

**Correlation is significant at the 0.01 level (2-tailed) the distant sampling site (S5). The main reason is that water becomes more dilute in organic matter, regardless of the seasonal changes. However, as reported above, fall results come as a dispute to the overall conclusions. The same reduction in numbers has been observed with Fecal Enterococci with a significant decrease ($p < 0.01$) from 'S1' to 'S5' locations, regardless the season. Bacterial reductions are shown in Fig. 7.

The correlations of physicochemical and bacteriological parameters are shown in Table 6.

### Sedimentological – physical processes in the lagoon inlet

The water renewal of eastern Klisova lagoon is made via the peripheral channels in the northern part of the lagoon and by an artificial canal of 150 m wide and 5 km length from the south (Figs. 1 and 2). The canal ensures the communication with the sea water and improves the lagoon water circulation. The mouth of the canal is 2.5 Km distance from the Evinos river delta and it is influenced by the river sediments load and its delta plain progradation (Fig. 8a). The sediments load and delta plain progradation as well as delta mouths development, is crucial for the conservation of the lagoon ecosystem. Siltation events of the lagoon inlet occur as the result of intense rainfall and the high bed and suspended river load. The siltation event which is described in the present study was occurred on winter 2010. The inlet of the lagoon was dammed by sediments of medium to coarse sand which were accumulated in the inlet mouth. In Fig. 8a and b we present the inlet morphology before and after the siltation event of 2010 (Google Earth 2008 and 2012). The above siltation process

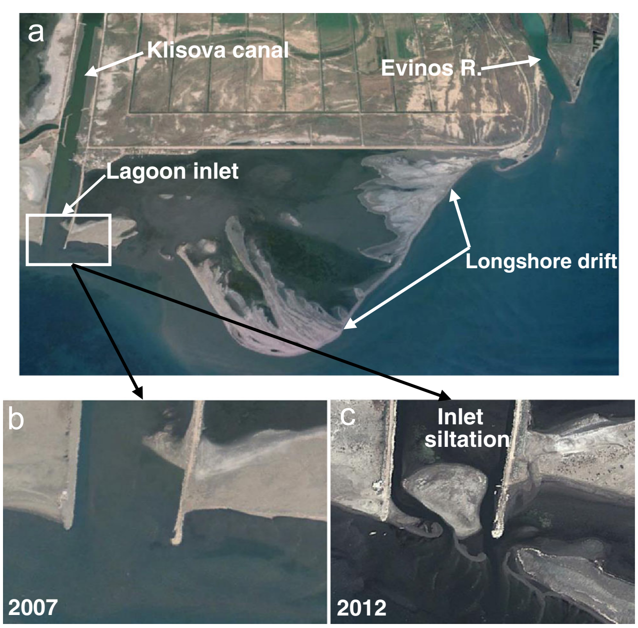


**Fig. 8** Aerial photographs showing: **a** the Klisova canal, the lagoon inlet and the Evinos R. delta, **b** the lagoon inlet in 2008 and **c** the lagoon inlet in 2012 with the sediments siltation as the result of longshore drifting

can be explained by the phenomenon of the longshore drifting, which is related to hydrodynamic influence, prevailing winds, sediment budget and coastal geomorphology. This assumption is in agreement with the hydrodynamic model of Patraikos gulf. Fourniotis and Horsch (2008) using the three – dimensional code MIKE 3 FM (HD) reveal a strong westward currents in the area of Rio-Antirio straits and in the study area, with a maximum current velocity of >5 m/s. The hydrodynamic regime of Patras Gulf described by Fourniotis and Horsch (2008; 2010) is the only available model where spatio-temporal parameters such as currents and water surface elevation drawing inferences, taking into account the tidal action, river discharge and prevailing winds and seems to explain very well the dammed of the Klisova Lagoon inlet mouth. The longshore drifting is physical and sedimentological process where the longshore currents transport quantities of as the results of prevailing winds and waves effects and) (Fig. 8a and b).

## Conclusion

The Klisova Lagoon is a protected coastal brackish aquatic system, threatened by human interference and physical – geological processes. The 1 year monitoring of the physicochemical lagoon properties reveal periods of hypoxic conditions with DO concentrations <2 mg/L, a clear zonation of total nitrogen (TN) salinity, pH and temperature, as the result of the interplay between the fresh water and the saline water coming from the sea via the lagoon inlet and the for years point source pollutant of waste water treatment plant of Mesolonghi city (Avramidis et al. 2010). The high total nitrogen concentrations, the low values of dissolved oxygen as well as the bacteriological load of the lagoon's water constitute an 'alarming situation' for the ecosystem of the eastern Klisova lagoon. Based on Avramidis et al. (2010) who studied the Klisova channels nutrient load and from the present study, it seems that the channels continue to discharge polluted water into the lagoon with high content of nutrients and bacterial load such as Enterococci and E.coli which both prove the point source of the pollution. Moreover, the sedimentological phenomenon of the siltation of the lagoon inlet, as the result of longshore drifting and the growth of the Evinos River delta, contributes to the limited renewal of the lagoon water and the deterioration of the ecological status of the lagoon. The triggering mechanism of the lagoons ecological socks seems to be both the human interference and physical processes. The above parameters have leaded the whole part of the lagoon to be characterized by oxygen depletion periods and hypoxic events and in combination with the bacteriological data, the aquatic area is characterized as inappropriate for the extensive use in fish farming.

Following the directives of the European Community, a permanent water monitoring procedure has to be installed in order to manipulate and limit the environmental risk of the protected and threatened ecosystem of Klisova lagoon. Moreover, artificial works have to carry out in order to maintain the lagoon water communication with the open sea and improve the circulation of the water inside the Klisova lagoon.

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