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ARTICLE

Spatio-Temporal Variability in Water Chemistry of Mediterranean Coastal Lagoons and its Management Implications

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Abstract Temporal variability is widely acknowledged as a main source of uncertainty and error in ecological studies. Despite this, conservation management tends to overlook the temporal dimension of ecosystems due to financial and operational constraints. The main aim of this study is to identify potential biases in the implementation of the European Water Framework Directive due to temporal variability in water-chemistry parameters. Specifically, we focused on the establishment of water-bodies typologies and the assessment of water quality in transitional waters. We measured water-chemistry (salinity and nutrients) in 34 coastal lagoons on a monthly basis over a full year (March'08–February'09) in a Mediterranean archipelago (Balearic Islands, Spain). We found large differences in nutrient levels among both the coastal lagoons of different islands (Majorca, Minorca, Ibiza and Formentera) and lagoons of different salinity types (euhaline, mesohaline and oligohaline). From an applied perspective, our results show that monthly values were inadequate for determining salinity-based typologies, as lagoon salinity values varied widely from month-to-month. Additionally, we found a lack of agreement between ecological assessments based on invertebrates and those based on nutrient thresholds often used for transitional waters. These facts highlight the need for a definition of specific thresholds for coastal lagoons which are not currently available.

Keywords Transitional waters · Nutrient concentrations · Salinity · Typology · Ecological status assessments · Water framework directive · Balearic Islands

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Introduction

Coastal lagoons are transitional waters that are normally connected to the sea through one or more channels. In some cases, they may be connected to supply from inland freshwaters. They are considered to be an ecotone between marine, freshwater and terrestrial ecosystems, sharing characteristics with each of these ecosystem types. Many coastal lagoons are naturally eutrophic as they receive nutrient loadings from the whole river basin (Nixon 1995; Roselli et al. 2009 and references therein) and/or from internal recycling processes between their sediment and water column because of their shallowness (Kormas et al. 2001 and references therein; Ji 2008). These properties have led to controversy in establishing water quality standards for these ecosystems (Newton et al. 2003). There are no specific benchmarks for nutrient-based water quality of Mediterranean coastal lagoons, despite this issue having been already claimed by some authors (Giordani et al. 2009) and several projects during the '90s that focused on the water quality of transitional waters, most including Mediterranean coastal lagoons (e.g., Coastal Lagoon Eutrophication and ANaerobic processes (CLEAN) Caumette et al. 1996; "ROle of BUffering capacities in Stabilizing coastal lagoon ecosystems" (ROBUST) de Wit et al. 2001; Development of an Information Technology Tool for the Management of European Southern Lagoons under the influence of river-basin runoff (DITTY) Marinov et al. 2007).

A main goal for the 2010 international biodiversity targets was to promote the conservation of biological diversity of ecosystems, habitats and biomes (Xu et al. 2010). Achieving this aim requires basic knowledge about the properties and functions of ecosystems, particularly those under threat. In general, aquatic ecosystems have largely benefited from this initiative, by providing new opportunities and funding for research (Revenga et al. 2005). However, some categories of aquatic ecosystems, such as transitional waters and more specifically coastal lagoons, have received little attention compared with rivers or lakes. This lack of attention is likely due to the coastal lagoons being positioned between purely marine and purely freshwater ecosystems (Razinkovas et al. 2008). However, coastal lagoons are ecosystems impacted by human activity to the extent of severe, and even irreparable, damage and thereby their retrieval must be immediate (HELCOM 1998). Due to their vulnerability, coastal lagoons have been listed as a priority habitat in the European Union Habitats Directive (92/43/EEC) as well as included in the European Water Framework Directive (WFD - 2000/60/EC) under the category "transitional water".

The WFD was adopted by the European Union in 2000 and a primary goal was achieving "good ecological status" for all surface waters by 2015. The WFD classification scheme for water quality includes five classes of ecological status: high, good, moderate, poor and bad. "High status" is defined as the biological, chemical and morphological conditions associated with no or very low human pressure. This is also called the "reference condition" as it is the best status achievable - the benchmark. Assessment of quality is based on the extent of deviation from this reference condition. According to the Directive, "good status" means 'slight' deviation, "moderate status" means 'moderate' deviation, and so on (see WFD Annex V for more complete information). The WFD has affected water management policies throughout Europe since its inception, as it puts aquatic ecology at the heart of management decision-making (Hering et al. 2010). Unfortunately, the implementation of the WFD in the context of transitional waters has been slow to materialize. This delay is likely to prevent the achievement of WFD environmental objectives by 2015 (Basset et al. 2004, 2006).

For transitional and coastal waters, the WFD identifies five "general chemical and physiochemical elements supporting the biological elements". The five elements are transparency, thermal conditions, oxygenation conditions, salinity and nutrient conditions. Although some work has been done to define the standards for some of these elements (Borja et al. 2004; Bald et al. 2005; Best et al. 2007), these standards are still under development and may be subject to further revision. Moreover, most of these attempts to define the standards for physicochemical elements in transitional waters have been carried out in estuaries, not specifically in coastal lagoons, which differ from estuaries despite both being transitional waters.

In general terms, ecological theory implicitly assumes that biotic and abiotic components of ecosystems change over time (Preston 1960; Levin 1992). This temporal variability is accentuated in Mediterranean regions because of seasonal variations in weather conditions and the unpredictable character of the disturbances (Blondel et al. 2010). Temporal fluctuations in coastal lagoons are expected to be large due to their intermediate position between marine and freshwater ecosystems and the influence of physical and meteorological drivers, such as wind, tides and seasonal variation in runoff (e.g., Elliott and Quintino 2007; Dauvin 2007). Additionally, there are temporal variations in the structure of fish (Pérez-Ruzafa et al. 2007), invertebrate (Casagranda et al. 2006) and plankton communities (Drake et al. 2010) of coastal lagoons and in their physico-chemical characteristics (Wilke and Boutiere 2000). Despite its importance, the temporal dimension of ecosystems is often overlooked, raising the risk of the misapplication of management actions.

The Balearic Islands are an archipelago located at the Mediterranean Sea (east Spanish coast) which have a large number of permanent coastal lagoons. All are protected under Spanish law (Ley de Aguas – Spanish Water Law (RDL 1/2001)) and were recently included in the River Basin Management Plans of the Balearic Islands. The Balearic Islands can be used as a model system for ecological studies on coastal lagoons as they have a large number of coastal lagoons in a relatively small area. Additionally, since lagoons are located on different islands and experience different environmental conditions, they provide an opportunity to study physico-chemical and conservation gradients. In general, the four main islands in the Balearic archipelago (Majorca, Minorca, and Pitiüsas Islands, which include Ibiza and Formentera) present different anthropogenic activities and land use patterns. For instance, the Majorca Island has the highest percentage of agricultural activity (76.8 %), followed by the Pitiüsas Islands (Ibiza and Formentera) (17.8 %) and Minorca (5.3 %) (Conselleria d'Agricultura i Pesca 2009). Similarly Minorca is characterized by livestock (mainly sheep) production, (e.g., 64 % of organic cattle farms are located on Minorca island) (Conselleria d'Agricultura i

Pesca 2009). Hence, we expect to find the ecological impact of these particular land-use patterns on the lagoons of these islands rather than on the other islands (see Table 1).

The main aim of this study is to describe the variation in water-chemistry across time and space in a Mediterranean system of small coastal lagoons. Specifically, we: i) provide a basic description of the water-chemistry characteristics in Balearic archipelago coastal lagoons; ii) evaluate monthly variation in nutrient concentrations, and iii) assess differences in nutrient concentration among islands and lagoon salinity-types across time. Additionally, the applied interest of this study is iv) the evaluation of the robustness of salinity-based typologies inferred from monthly samplings and v) the comparison of nutrient-based and fauna-based ecological status assessments in the WFD context.

Material and Methods

Area of Study

This study was conducted on coastal lagoons located on the Balearic Islands (Spain), an archipelago located in the western Mediterranean Sea (31S 499562 E/4386674 N; 4.991,7 km^2 total area) (Fig. 1). The four main islands are (in decreasing size): Majorca (MA, which occupies 72 % of the total area), Minorca (MI, which occupies 14 % of the total area), the Pitiüsas Islands that include Ibiza (IB, which occupies 11.5 %) and Formentera (FO, which occupies 1.7 %). The islands have a Mediterranean climate characterized by warm, dry summers and mild, wet winters. Rainfall is concentrated during autumn-winter, and is scarce or negligible during late spring and summer (Bolle 2003).

We monitored 34 permanent coastal lagoons on a monthly-basis. We sampled at a single station for 20 of the lagoons and at multiple sampling stations (two to seven) on 14 lagoons (due to their large size) resulting in a total of 64 sampling stations (Table 2).

Table 1 Gross pollutant loads discharged from agricultural use per island

Island	N (Tons)	P_2O_5 (Tons)	K_2O (Tons)
Majorca	4940	2233	1811
Minorca	1098	442	216
Pitiüsas (IB and FO)	332	158	148
Balearic Islands	6370	2833	2175

Conselleria d'Agricultura i Pesca 2009

IB Ibiza, *FO* Formentera

The permanent coastal lagoons of the Balearic Islands are typically shallow (< 4 m depth) and are waterbodies affected by microtides (< 1 m). They range in size from 900 m^2 to 15.2 km^2, with most lagoons being small (< 2.5 km^2). The lagoons may have a direct connection with the sea, with adjacent streams (semi-closed lagoons), with both systems (lagoons locally called "golas"), or with none of them (closed lagoons). The salinity of the coastal lagoons ranges from oligohaline (≤ 5 ‰), mesohaline (6–30‰) to euhaline (> 30 ‰) (Lucena-Moya et al. 2009; Lucena-Moya and Pardo 2012). Their hydrology is dependent on the temporal variability of their main water inflows (i.e., rainfall, streams, runoff, sea storms and groundwater discharges) which may cause sudden and irregular flooding. Evaporation, infiltration and discharge to the sea are the main water outflows. The lagoons are generally surrounded by halophytic vegetation (i.e. phytosociological communities Arthrocnemetum fruticosi, Tamaricetum canariensis, Spartino-Juncetum maritimi and Salicornietum emerici) (Tomàs-Vives and Crespí-Ramis 2002). These lagoons differ in their ecological status, as shown in a previous evaluation conducted at 95 % of the sampling stations included in this study and based on a multimetric index (MIBIIN) that considered the invertebrate community (Lucena-Moya and Pardo 2012).

Field Sampling and Laboratory Analysis

Field sampling was carried out between March 2008 and February 2009. During this period, the mean annual temperature of the archipelago was 16.9 °C and the total rainfall was 471.9 mm (AEMET 2010).

Each sampling station was sampled during the first 15 days of a month (except in August for MI and November for IB and FO, when lagoons were not sampled due to logistic problems). Unfiltered (for salinity and total nutrients) and filtered (for dissolved nutrients) water samples were collected at each sampling station. Filtration was carried out in situ with a hand-pump through a glass fibre filter (Whatman GF/F with a pore size 0.45 μm). Samples were immediately frozen and transported to the laboratory. Salinity and nutrients (total nutrients: total phosphorus [TP mg/L] and total nitrogen [TN mg/L]; and dissolved nutrients: phosphate [P-PO_4^{-3} mg/L], nitrate [N-NO_3^- mg/L], nitrite [N-NO_2^- mg/L] and ammonium [N-NH_4^+ mg/L]) were analyzed following the specific ISO standard methods for water samples (ISO 1996, 2003, 2005) by means of a conductivity-meter (Model Orion 125) and a continuous-flow analyzer (Auto-Analyzer 3, Bran + Luebbe, Germany), respectively.

Dissolved inorganic nitrogen [DIN mg/L] was computed as the addition of N-NO_2^-, N-NO_3^- and N-NH_4^+. The limits of detection were: P-PO_4^{-3}=0.0016 mg/L; N-NO_3^-= 0.005 mg/L; N-NO_2^-=0.003 mg/L; N-NH_4^+=0.001 mg/L.

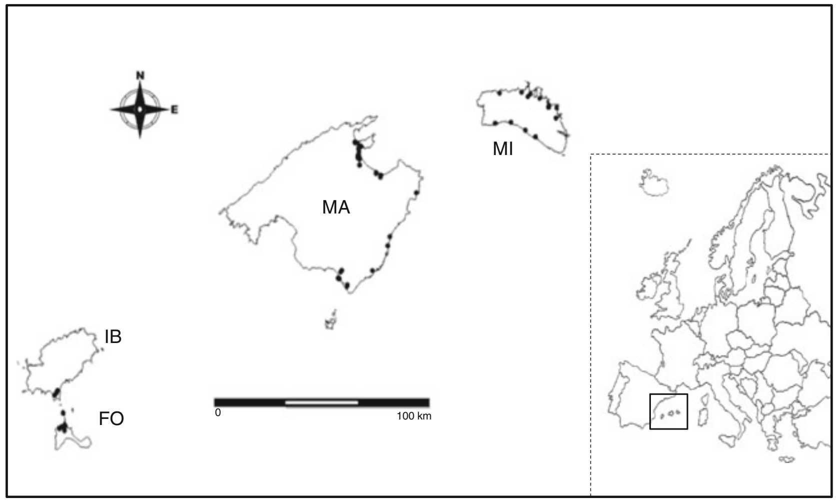

Fig. 1 Map of the Balearic Islands. *MA* Majorca, *MI* Minorca, *IB* Ibiza, *FO* Formentera. *Black dots* indicate sampling stations

Statistical Analysis

Main Nutrient Gradients in Coastal Lagoons

To identify the main environmental gradients for lagoon water chemistry a Principal Component Analysis (PCA) with varimax rotation was performed using the monthly values of log-transformed nutrient concentrations as variables (Statistica V7). The PCA also depicted the relationships among monthly nutrient concentrations and the abiotic similarities among sampling stations. Additionally, to evaluate differences in abiotic conditions of sampling stations among islands and salinity types, one-way analysis of similarities (ANOSIM) (Primer v6; Clarke and Warwick 2001) based on Euclidean distances were computed. The combination of the island and salinity type factors (e.g., MA-oligohaline, MA-mesohaline, MI-oligohaline, etc.) was used as a single grouping factor. Underrepresented combinations (number of sampling stations <4) were excluded (i.e., MI-euh, IB-oligo, IB-meso, FO-oligo and FO-meso). Bonferroni correction was applied to pair-wise ANOSIM R-statistics, resulting in a p-value for significance of 0.0056 (α=0.05; number of combinations=9).

Table 2 Number of sampling stations and coastal lagoons (in brackets) in each island (*MA* Majorca, *MI* Minorca, *IB* Ibiza and *FO* Formentera) and for each salinity type

Type	Island			
	MA	MI	IB	FO
Euhaline	8 (4)	2 (1)	4 (1)	7 (4)
Mesohaline	19 (9)	6 (2)	1 (1)	
Oligohaline	6 (2)	10 (9)		1 (1)
Total	33 (15)	18 (12)	5 (2)	8 (5)

Differences in Nutrient Concentration and Salinity Among Types, Islands and Time

Islands differed in the number of sampling stations per salinity type, resulting in an unbalanced dataset with euhaline lagoons being underrepresented on MI (n=2), and oligohaline and mesohaline lagoons being underrepresented on IB and FO ($n\leq1$; Table 2). This prevented the evaluation of all hypotheses using a single statistical test considering all salinity types and islands simultaneously. Comparison among all salinity types was only possible for MA, as it was the only island where all three salinity types were sufficiently abundant ($n\geq4$). For these reasons, three independent analyses were conducted: [i] a comparison of the three salinity types using only MA; [ii] a comparison of oligohaline and mesohaline types, using MA and MI; [iii] and a final analysis for the euhaline type, using MA, IB and FO.

For each nutrient and the DIN/P-PO_4^{-3} ratio, differences in concentration among salinity types and/or islands were evaluated taking into account temporal variation. Thus, one-way (type [i] or island [iii]) or two-way (island and type [ii]) Repeated Measures ANOVA (RMA) with post-hoc Tukey's Honestly Significant Difference test (Tukey-HDS) were conducted using monthly values as input variables. The Tukey-HSD test was applied to identify the differences between the levels (e.g., oligo-, meso-, eu-haline) of a factor (salinity type in this example) (Underwood 1997). The interaction with time (within-subjects factor) was evaluated, but only post-hoc significant differences corresponding to the same month were considered. Thus, we discarded significant differences where the parameter value for a given island (or type) in one particular month was significantly different from values in other months. All variables were transformed to meet the parametric assumptions of normality and homoscedasticity and we removed those that did not fulfill these conditions (referred to as "non analyzable data" in the text; maximum number of months removed for a given nutrient=5).

Ecological Status and Nutrient Concentration

For each lagoon salinity type, differences in nutrient concentrations (TP, P-PO_4^{-3}, TN and DIN) among lagoons with different ecological status classes were evaluated using one-way ANOVAs with post-hoc Tukey-HSD test. Ecological status class for each lagoon was determined using a multimetric index based on the invertebrate community (MIBIIN index, see Lucena-Moya and Pardo 2012). In brief, MIBIIN is a combination of metrics based on diversity, relative abundance and sensitive taxa of the invertebrate community and specific for each lagoon salinity type (oligo-MIBIIN = ∑ [Frequency of sensitive genera + Genera richness + Frequency of (*Cyprideis torosa* + Polychaeta)]; meso-MIBIIN = ∑ [Sensitive genera richness + Bray-Curtis Dissimilarity + Frequency of (Amphipoda + Gastropoda + Isopoda)] and euh-MIBIIN = ∑ [Sensitive genera richness + Frequency of *Artemia salina*)]. This multimetric is applied to reference sites (i.e., pristine or close to pristine state) and to non reference sites. The ratio between both, observed (non references)/expected (references), results in a value which is identified with an ecological status class (high, good, moderate, poor or bad). F- or Welch-statistic were used depending on whether the data met or did not meet the requirements of normality. A Bonferroni correction was applied, resulting in a p-value for significance of 0.0125 (α=0.05; number of nutrient parameters=4). Additionally, the correspondence between MIBIIN and nutrient-based ecological status classes was explored. The latter was obtained from two widely-used assessment guides: European Environment Agency (EEA 1999) for freshwater, marine and transitional environments and, the United States National Estuarine Eutrophication Assessment (Bricker et al. 2003). The correspondence was based on median values.

Results

Main Nutrient Gradients in Coastal Lagoons

A comprehensive description of the values of nutrients through coastal lagoons from the Balearic Islands is provided in Table 3. Mean and median values are relatively low, but the statistics descriptors such as maximum and minimum give an idea of how variable the data can be.

The PCA used to identify environmental gradients found two main gradients which explained 36.74 % of the variation in the water chemistry of the coastal lagoons (Fig. 2). The first gradient accounted for 21.47 % of the variability and was highly correlated (variable loading ≥0.7) with N-NO_3^- in March, April, June, January and February and the DIN/P-PO_4^{-3} ratio in April, June, January and February. The second gradient accounted for 15.27 % of the variability and was highly correlated with P-PO_4^{-3} in April, May, July and September to January (Fig. 2a). The ordination of lagoons based upon the PCA components was scattered and showed no clear segregation among islands or types (Fig. 2b). However, six sampling stations were clearly different from the rest: four in Majorca (two mesohaline and two oligohaline lagoons) and two in Minorca (both oligohaline lagoons). They were among the most eutrophic stations within their respective categories. The lack of differences among islands and/or types was also supported by the analysis of similarities (ANOSIM) which found no significant differences among coastal lagoons (p>0.0056, non-significant following Bonferroni correction, see ESM 1 for details).

Differences in Nutrient Concentration and Salinity Across Types, Islands and Time

Differences in Water-Chemistry Among Salinity Types (Oligohaline, Mesohaline and Euhaline): The Case of Majorca Island

Salinity values varied over time ($F_{11,330}$=10.05; p<0.0001). Salinity values for the oligohaline and the mesohaline lagoons were minimal from March to June and from November to February (high rainfall and low temperature) and were maximal from July to October (low rainfall and high temperature) (Fig. 3). Salinity values for the euhaline lagoons were high during both early spring and late summer (March–October) (Fig. 3). The salinity ranges of the three lagoon types over the course of the sampling period frequently overlapped (minimum value-maximum value) among types (oligohaline=0.3–21.1; mesohaline=0.2–65.3; euhaline=3.2–49.6). In fact, salinity values for the three lagoon types were similar each month, and differing significantly only in the case of the euhaline lagoon type from March to July and in December.

Except for N-NO_3^-, nutrient concentrations did not differ significantly among lagoon salinity types (Table 4[i] and see ESM 2 for details). N-NO_3^- concentrations were significantly higher in the oligohaline lagoon type than in the mesohaline and the euhaline lagoons (Tukey HSD, p<0.05). All nitrogen forms exhibited a significant increase during winter months, with the exception of N-NH_4^+ which lacked analyzable data for those months (Table 4[i] and ESM 2 and 3 (a)). Unlike nitrogen, the different forms of phosphorus did not show a strong seasonal trend, although there was a significant interaction with time (Table 4[i] and ESM 2 and 3(a)). In detail, significant differences were observed due to punctual high values of TP in February and July (ESM 2 and 3(a)). Similar to nitrogen, the DIN/P-PO_4^{-3} ratio increased significantly during the winter months (Table 4[i] and ESM 2 and 3(a)). In general, the interaction

Table 3 Statistical descriptors of nutrients of the coastal lagoons in the Balearic Islands. Data include 64 sampling stations sampled during 12 months (missing August in Minorca and November in Ibiza and Formentera). *dl* detection level. Note that data from all the lagoons of all salinities on the four islands have been combined to produce this summary

	TP (mg/L)	$P\text{-}PO_4^{-3}$ (mg/L)	TN (mg/L)	$N\text{-}NO_3^-$ (mg/L)	$N\text{-}NO_2^-$ (mg/L)	$N\text{-}NH_4^+$ (mg/L)
Mean	0.53	0.05	3.72	1.52	0.02	0.09
Standard Error	0.07	0.01	0.34	0.20	<dl	0.01
Median	0.07	0.01	1.52	0.04	<dl	<dl
Maximum	28.32	1.53	189.74	82.38	1.25	4.36
Minimum	<dl	<dl	0.02	<dl	<dl	<dl
Percentile 25	0.04	<dl	0.68	<dl	<dl	<dl
Percentile 95	2.3	0.29	13.88	8.01	0.09	0.4
N	715	715	715	715	714	715

term for type with time was significant for most nutrients (ESM 3(b)), but post hoc analyses showed that such significance did not correspond to differences among types within the same month.

Differences in Water-Chemistry Among Islands (Majorca vs. Minorca and Majorca vs. Ibiza vs. Formentera)

Lagoons in Majorca had significantly higher concentrations of TN, $N\text{-}NO_3^-$ and $DIN/P\text{-}PO_4^-$ than Minorca (Tukey HSD, $p<0.05$; Table 4[ii] and Fig. 4a–d). However, the interaction term for island with time indicated that these differences were not constant across time, but rather occurred only during the winter months (Tukey HSD, $p<0.05$). Minorca lagoons had significantly higher concentration of $P\text{-}PO_4^{-3}$ than Majorca lagoons (Tukey HSD, $p<0.05$; Table 4[ii]), but these differences were not detected across time (Fig. 4c and ESM 3c), that is they were not evident in any particular month. In the case of TP, significant differences were observed in particular months, as TP in February in Majorca was significantly higher than in Minorca in any given month (Tukey HSD, $p<0.05$).

Lagoons in Majorca also had significantly higher values of TN and $DIN/P\text{-}PO_4^{-3}$ than lagoons in Ibiza and Formentera (Tukey HSD, $p<0.05$; Table 4[iii] and Fig. 5). These differences were not constant across time in the case of $DIN/P\text{-}PO_4^{-3}$ (see ESM 3f); and, in the case of TN, these differences were not observed in any particular month. Regarding TP, concentrations in January were similar for Ibiza and Formentera and were significantly higher on those islands than on Majorca (Tukey HSD, $p<0.05$; Fig. 5 and ESM 3f).

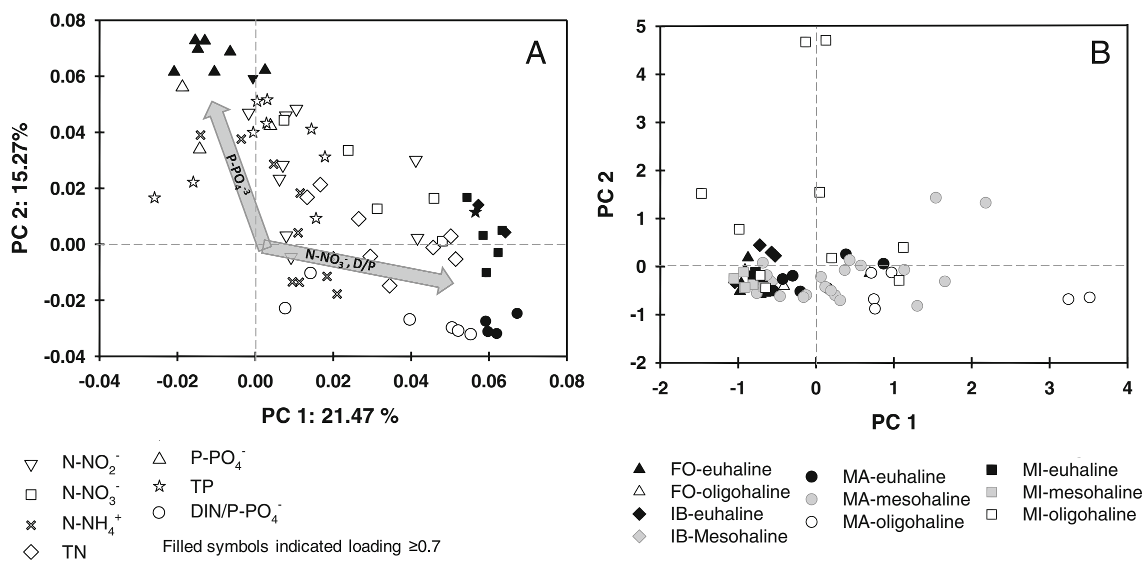

Fig. 2 Results from Principal Component Analysis using monthly nutrient concentrations and $DIN/P\text{-}PO_4^{-3}$ ratio as input variables. **a** Filled symbols are used for variables with loading≥0.7, which are also summarized by *two arrows* (D/P: $DIN/P\text{-}PO_4^{-3}$ ratio). **b** Ordination of coastal lagoons based on the first and second Principal Components, i.e., PC1 and PC2 (*MA* Majorca, *MI* Minorca, *IB* Ibiza and *FO* Formentera)

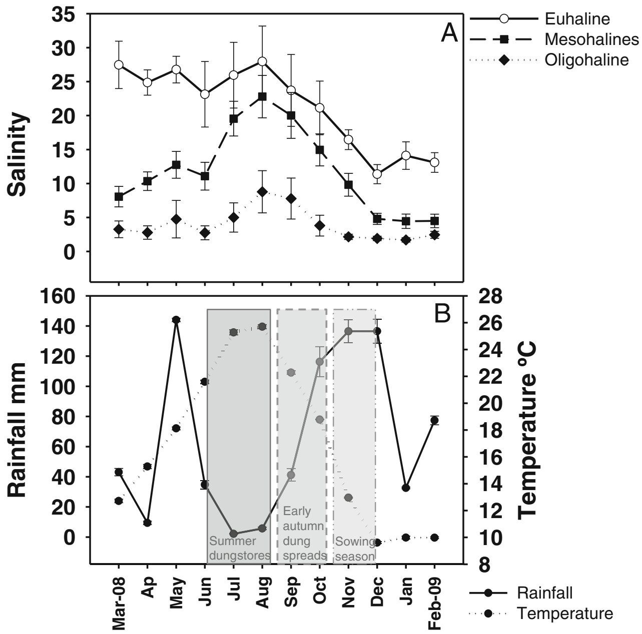

Fig. 3 (**a**) Mean salinity values ± standard error (SE) across time for all salinity types in Majorca Island (**b**) Climate diagrams (mean monthly temperature ± SE and mean monthly precipitation ± SE) of the study period in Majorca Island (data obtained from the closest meteorological stations to the coastal lagoons). Periods of agricultural activities (summer dung stores, early autumns dung spreads and sowing season) are shown

Differences in Water-Chemistry Between Types (Oligohaline vs.Mesohaline Types) and Islands (Majorca vs. Minorca)

Oligohaline coastal lagoons had significantly higher nutrient concentrations than mesohaline lagoons, with the exception of N-NH_4^+ and DIN/P-PO_4^{-3} (Tukey HSD, $p<0.05$; Table 4[ii] and Fig. 4e–h). The differences in P-PO_4^{-3}, TN and N-NO_2^- did not occur across time (ESM 3d). Differences in the interaction term between island and type were found for TP, P-PO_4^{-3} and N-NO_2^- concentrations (Table 4[ii]). P-PO_4^{-3} was significantly higher for the Minorca-oligohaline coastal lagoons than in the rest of the lagoons (i.e., Minorca-mesohaline and Majorca-oligohaline and mesohaline) (Tukey HSD, $p<0.05$) but these differences were not constant across time (Fig. 4k and ESM 3e). Differences in TP and N-NO_2^- were due to concentrations being significantly lower in the Minorca-mesohaline lagoons than in the Minorca-oligohaline lagoons (Tukey HSD, $p<0.05$; Table 4[ii], Fig. 4j and l), but not across time (ESM 3e).

Ecological Status and Physico-Chemical Conditions

Nutrient concentrations did not tend to increase with decreasing ecological status, as coastal lagoons with poor ecological status did not always have higher nutrient concentrations than lagoons with better ecological status (Fig. 6). In the oligohaline and mesohaline coastal lagoons, significant differences among ecological status classes were detected for all nutrient parameters, except for TP in mesohaline lagoons (Table 5). For the oligohaline lagoons, these differences were due to significantly lower nutrient concentrations in reference lagoons (Fig. 6 and Table 5). For the mesohaline lagoons, the differences were due to significantly higher nutrient concentrations in lagoons with *good* ecological status (Fig. 6 and Table 5). Conversely, for the euhaline lagoons only the forms of nitrogen showed significant differences, with lagoons of *moderate* ecological status having significantly lower TN and DIN concentrations (Fig. 6 and Table 5).

Ecological status inferred from the invertebrate community using the MIBIIN index did not correspond with ecological status inferred with nutrient-based assessments (European Environment Agency (EEA) 1999; United States National Estuarine Eutrophication Assessment (Bricker et al. 2003) (Fig. 6). Assessments based on DIN concentration evaluated most lagoons positively, even those classified as bad or poor by the MIBIIN index (Fig. 6). Assessments based on phosphorus differed; the one following Bricker (2003) evaluated all lagoons negatively (using TP); whereas the EEA (1999) evaluated almost all lagoons positively (using P-PO_4^{-3}).

Discussion

Spatial and temporal variability are key issues in understanding ecological systems (Levin 1992; Hastings 2010). However, addressing both sources of variability simultaneously is usually not feasible because of the associated costs, and often, one dimension is relinquished in exchange for a better understanding of the other. This is particularly true for studies of aquatic ecosystems, which tend to be snapshots of either time or space (Chainho et al. 2006; Plus et al. 2006 but see, for example, Giordani et al. 2005; Pérez-Ruzafa et al. 2007; Souchu et al. 2010). We have attempted to provide an integrated view of spatial and temporal variability in this study by sampling a large number of coastal lagoons across both space and time. This approach allows not only the understanding of the limnological functioning but also the evaluation of the consequences derived from spatial-temporal variation that may condition management decision-making in Mediterranean coastal lagoons.

In general, median values of nutrient concentrations for Balearic coastal lagoons were lower than concentrations for other Mediterranean lagoons (Picot et al. 1990; Quintana et al. 1998; Kormas et al. 2001; Pérez-Ruzafa et al. 2005; Roselli et al. 2009) although some measurements of nutrient concentrations in the Balearic Islands were up to three orders of magnitude higher. It should be noted that the level of anthropogenic stress experienced by these other Mediterranean lagoons (e.g., Mar Menor-Spain-, Pérez-

Table 4 Repeated Measures ANOVA for nutrient concentrations and DIN/PO_4^{-3} ratio: [i] for the three salinity types considering only Majorca Island; [ii] for the oligohaline and mesohaline types, comparing Majorca and Minorca Islands; and [iii] for the euhaline type, comparing Majorca, Ibiza and Formentera Islands. Results for the main factors (islands, salinity-types and time) as well as the interaction between island and type are shown (F statistics, df: degrees of freedom, p-values). Interaction with the time and Tukey HSD post-hoc test are shown in ESM 3

Islands	Types	Nutrients	Main Factors						Interaction	
			$F_{(df)}$	p-value	$F_{(df)}$	p-value	$F_{(df)}$	p-value	$F_{(df)}$	p-value
			Type				Time			
[i] Majorca	Oligo- Meso- Euh-	TP (mg/L)	0.24 $_{(2,25)}$	0.789			50.30$_{(10,250)}$	**<0.0001**		
		$P\text{-}PO_4^{-3}$ (mg/L)	0.06 $_{(2,24)}$	0.938			2.08 $_{(10,240)}$	**0.026**		
		TN (mg/L)	2.10 $_{(2,23)}$	0.145			7.04 $_{(10,230)}$	**<0.0001**		
		$N\text{-}NO_3^-$ (mg/L)	3.95 $_{(2,23)}$	**0.033**			13.88 $_{(9,207)}$	**<0.0001**		
		$N\text{-}NO_2^-$ (mg/L)	1.77 $_{(2,22)}$	0.193			3.16 $_{(11,242)}$	**0.0005**		
		$N\text{-}NH_4^+$ (mg/L)	0.16 $_{(2,25)}$	0.853			1.63 $_{(6,150)}$	0.141		
		$DIN/P\text{-}PO_4^{-3}$	1.42 $_{(2,21)}$	0.263			7.99 $_{(10,\ 210)}$	**<0.0001**		
			Type		Island		Time		Island*Type	
[ii] Majorca Minorca	Oligo- Meso-	TP (mg/L)	4.37 $_{(1,31)}$	**0.045**	0.59 $_{(1,31)}$	0.449	54.55$_{(10,310)}$	**<0.0001**	7.00 $_{(1,31)}$	**0.013**
		$P\text{-}PO_4^{-3}$ (mg/L)	8.18 $_{(1,31)}$	**0.007**	10.83 $_{(1,31)}$	**0.002**	8.08 $_{(9,279)}$	**<0.0002**	5.90 $_{(1,32)}$	**0.021**
		TN (mg/L)	4.98 $_{(1,32)}$	**0.033**	9.08 $_{(1,32)}$	**0.005**	1.17 $_{(10,320)}$	0.312	0.10 $_{(1,31)}$	0.744
		$N\text{-}NO_3^-$ (mg/L)	18.53 $_{(1,30)}$	**<0.001**	4.44 $_{(1,30)}$	**0.044**	13.88 $_{(10,300)}$	**<0.0001**	0.90 $_{(1,30)}$	0.351
		$N\text{-}NO_2^-$ (mg/L)	12.65 $_{(1,33)}$	**0.001**	0.78 $_{(1,33)}$	0.383	2.41 $_{(9,297)}$	**0.012**	6.75 $_{(1,33)}$	**0.014**
		$N\text{-}NH_4^+$ (mg/L)	1.57 $_{(1,\ 29)}$	0.220	0.10 $_{(1,29)}$	0.750	4.31 $_{(9,261)}$	**<0.0001**	2.97 $_{(1,29)}$	0.095
		$DIN/P\text{-}PO_4^{-3}$	2.48 $_{(1,32)}$	0.124	18.66 $_{(1,32)}$	**<0.0001**	6.67 $_{(9,\ 288)}$	**<0.0001**	0.23 $_{(1,32)}$	0.632
					Island		Time			
[iii] Majorca Ibiza Formentera	Euh-	TP (mg/L)			2.72 $_{(2,13)}$	0.103	33.96 $_{(8,104)}$	**<0.0001**		
		$P\text{-}PO_4^{-3}$ (mg/L)			1.41 $_{(2,11)}$	0.284	13.13 $_{(10,110)}$	**<0.0001**		
		TN (mg/L)			10.08 $_{(2,11)}$	**0.003**	1.87 $_{(8,88)}$	0.074		
		$N\text{-}NO_3^-$ (mg/L)			3.52 $_{(2,11)}$	0.065	1.39 $_{(7,77)}$	0.220		
		$N\text{-}NO_2^-$ (mg/L)			0.46 $_{(2,12)}$	0.641	1.24 $_{(8,96)}$	0.285		
		$N\text{-}NH_4^+$ (mg/L)			1.84 $_{(2,11)}$	0.204	2.08 $_{(8,88)}$	**0.046**		
		$DIN/P\text{-}PO_4^{-3}$			4.15 $_{(2,12)}$	**0.042**	3.08 $_{(7,84)}$	**0.006**		

Values in bold indicate significant results

Oligo—Oligohaline, *Meso*—Mesohaline, *Euh*—Euhaline

Ruzafa et al. 2005; Lesina lagoon-Italy-, Roselli et al. 2009) is probably higher than in the Balearic coastal lagoons. This does not diminish the importance of anthropogenic effects on Balearic lagoons, as anthropogenic activities appear to be one of the major causes of their spatial variability in water-chemistry, along with differences in internal processes such as vertical matter and energy flows. The orthogonal arrangement between phosphate and nitrate concentrations found in this study suggests that they probably originate from different sources (see Quintana et al. 1998). Nitrogen loading depends mainly on inputs such as agricultural fertilizers leaching and runoff from fields (Boyer and Howarth 2002). Phosphorus loading is mainly due to wastewater discharges from livestock and human sewage (Newton et al. 2003). This would explain why coastal lagoons on Majorca, primarily an agricultural island (Conselleria d'Agricultura i Pesca 2009) (see also Table 1) had the highest nitrogen concentrations; while lagoons on Minorca, primarily a livestock farming island (Conselleria d'Agricultura i Pesca 2009), had the highest phosphorus concentrations. In

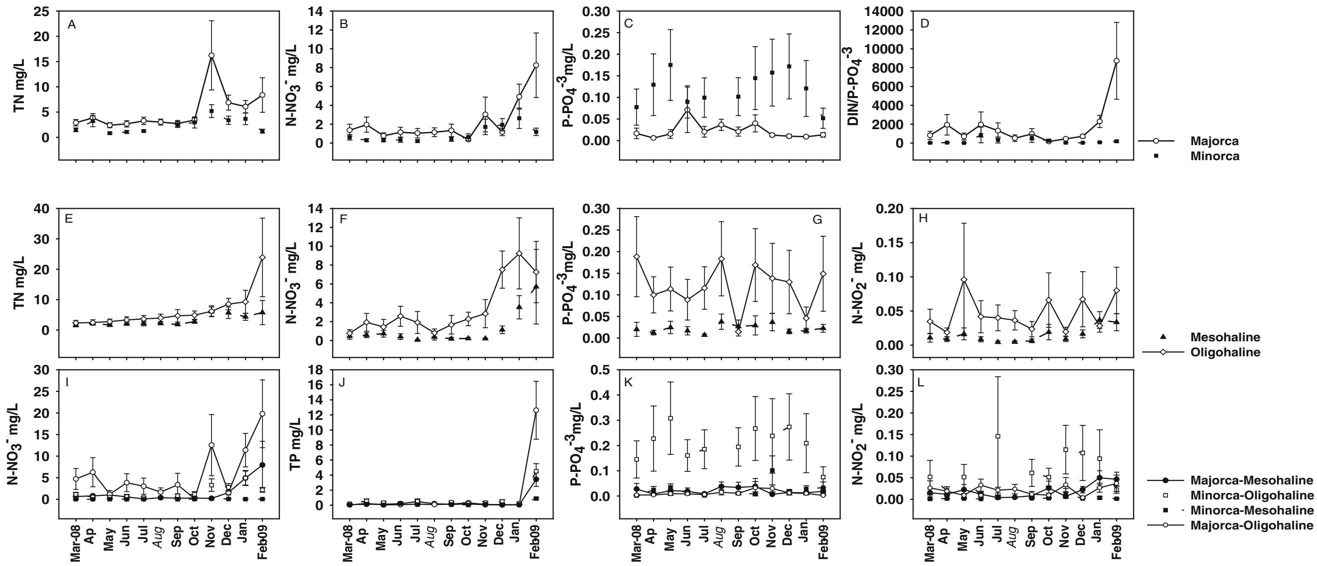

Fig. 4 Mean values and standard error of nutrient concentrations and DIN/P-PO_4^{-3} ratio in each month of survey. Data is shown for each island, Majorca and Minorca (**a**–**d**), for each salinity type, oligohaline and mesohaline (**e**–**h**), and for the interaction of both island and type factors (**i**–**l**). Only significant results are shown. *Months in italics* indicate non analyzable data

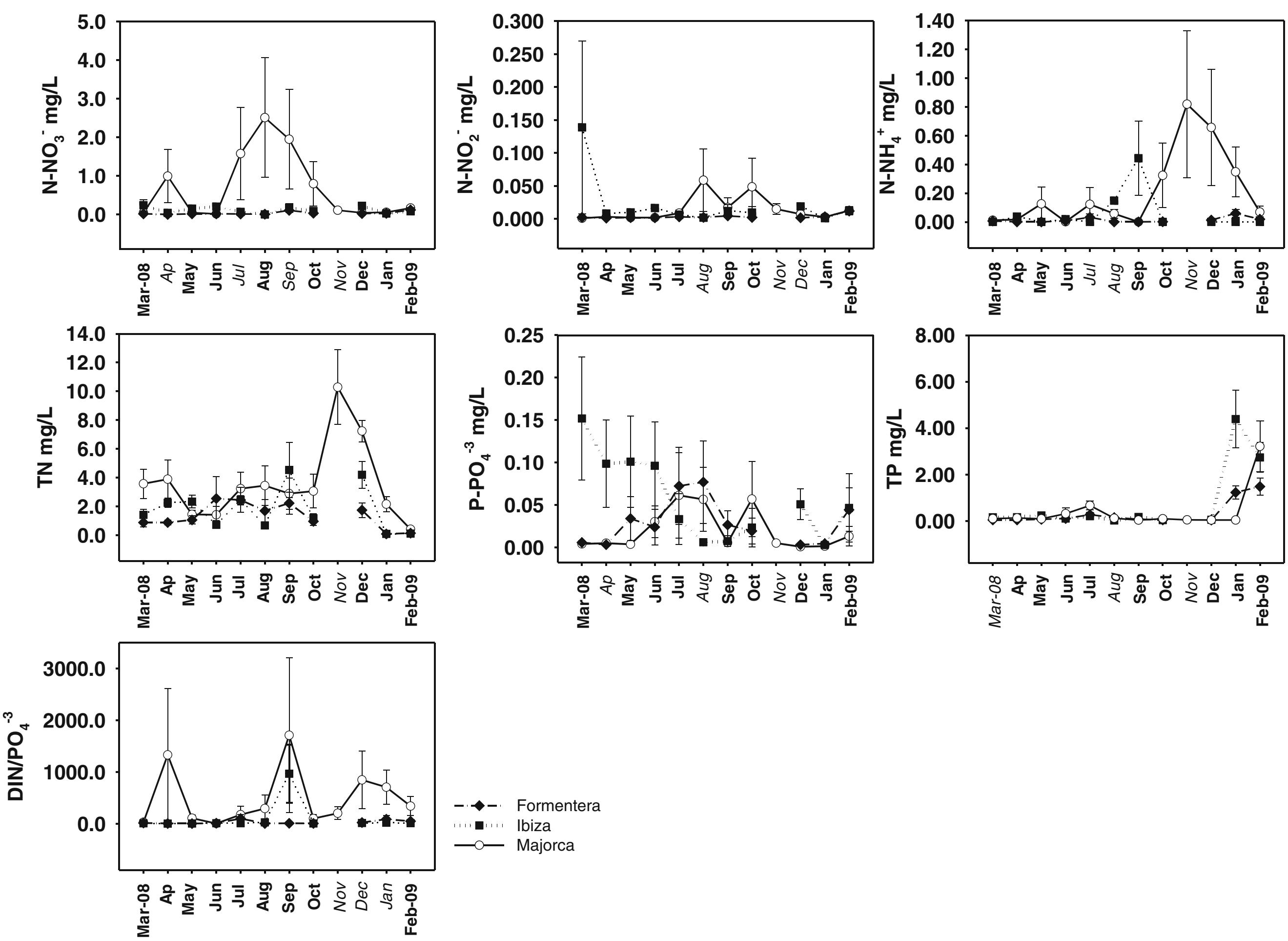

Fig. 5 Mean value and standard error of nutrient concentrations and DIN/P-PO_4^{-3} ratio in each month of the survey. Data is shown for the euhaline type in each island, Formentera, Ibiza and Majorca. *Months in italics* indicate non analyzable data

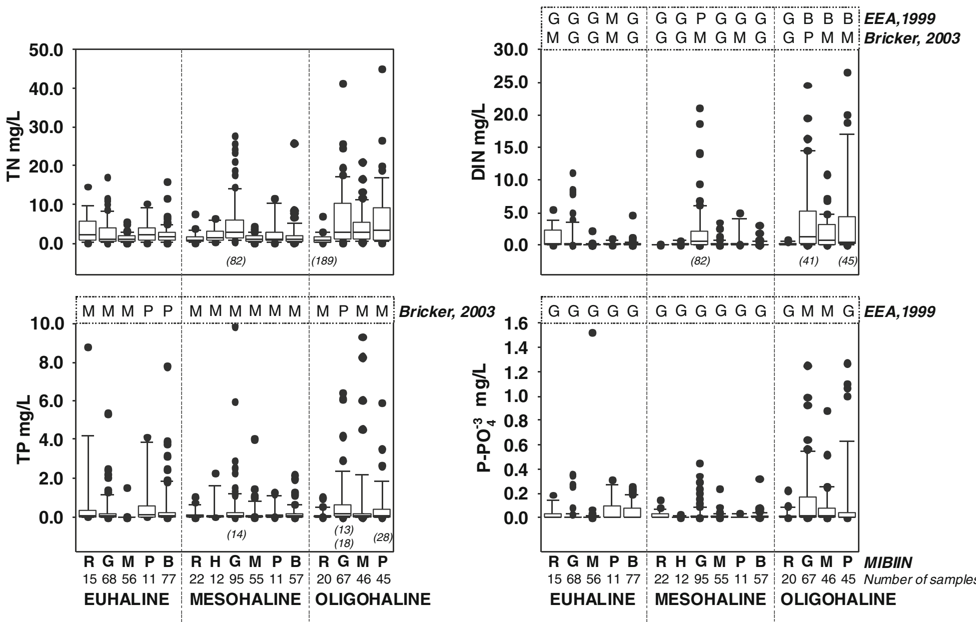

Fig. 6 Box plots showing nutrient concentrations for each salinity type and ecological status class inferred from the MIBIIN, from the EEA (1999) and from Bricker et al. (2003). *R* Reference; *H* High; *G* Good (Low trophic class for Bricker); *M* Moderate (Fair class for EEA, Medium class for Bricker); *P* Poor; *B* Bad (High trophic class for Bricker). Numbers of samples are shown. *Figures in brackets* indicate extreme values

the case of the Balearic Islands, it is estimated that sheep farming, the main livestock activity in Minorca, generates 44 % of the archipelago's total phosphorus (Conselleria d'Agricultura i Pesca 2009). Despite livestock activities producing both nitrogen and phosphorus, the former is removed quickly via denitrification and infiltration (Saunders and Kalff 2001; Boyer and Howarth 2002), while phosphorus is removed more slowly due to its sedimentary tendency (e.g., binding to clays) and slower remineralization rates, especially in closed and semi-closed coastal lagoons (Jenkins and Ives 1973; Golterman 2001).

Oligohaline coastal lagoons in both Majorca and Minorca showed the highest nutrient concentrations independent of the anthropogenic activity. This might be due to the high level of connection of this type of lagoons with freshwater flows (i.e., high waters turnover rate) (Comín and Valiela 1993; Roselli et al. 2009). Oligohaline lagoons seem to receive heavy nutrient loadings from the stream catchment area, via freshwater flows and runoff, which exceeds their metabolic capacity. In contrast, the water turnover rates in mesohaline and euhaline coastal lagoons were lower as they are generally shallower, and have limited exchanges with

Table 5 Results from the ANOVA evaluating differences in ecological status, derived from the MIBIIN index, among salinity types (F-statistic for normal and homoscedastic parameters or Welch-statistic for normal and heteroscedastic parameters, df: degrees of freedom, p-values and Tukey HSD post-hoc test)

Types	Nutrients	$F_{(df)}$	Welch $_{(df)}$	p-value	Post-hoc
Oligohaline	TP (mg/L)	3.14 $_{(3,174)}$		**0.027**†	R < G
	TN (mg/L)	5.31$_{(3,174)}$		**0.002**	R < G = M = P
	$P\text{-}PO_4^{-3}$ (mg/L)		4.21 $_{(3,93.7)}$	**<0.0001**	R < G = M = P
	DIN (mg/L)		17.86 $_{(3,\ 84.3)}$	**<0.0001**	R < G = M = P
Mesohaline	TP (mg/L)	1.32 $_{(5,\ 246)}$		0.254	
	TN (mg/L)	12.05 $_{(5,246)}$		**<0.0001**	G > R, M, B
	$P\text{-}PO_4^{-3}$ (mg/L)		4.59 $_{(5,71.5)}$	**0.001**	G > H, P
	DIN (mg/L)		4.38 $_{(5,50.1)}$	**0.002**	G > R
Euhaline	TP (mg/L)	1.10 $_{(4,222)}$		0.358	
	TN (mg/L)		5.85 $_{(4,42.6)}$	**0.001**	M < G, B
	$P\text{-}PO_4^{-3}$ (mg/L)		0.98 $_{(4,46.7)}$	0.425	
	DIN (mg/L)		4.47 $_{(4,\ 47.9)}$	**0.004**	M < G

Figures in bold indicate significant values after Bonferroni correction (α=00125)

R reference; *G* good, *M* moderate, *P* poor, *B* bad

† Significant value without considering Bonferroni correction

the sea, and only minimal connections with streams. For these lagoons, nutrient loadings appear to be related to natural internal processes (e.g. internal recycling) rather than external sources such as runoff from streams. These natural processes are associated with the lagoon's shallow depths and confined nature which causes the accumulation of organic matter or benthic flux. In this study, internal processes were not evaluated. However based on these results, future studies should focus on the driving forces relevant to each type of coastal lagoon in order to allow for a comprehensive comparison across lagoon types for the Balearic Island and other Mediterranean lagoons.

Temporal variability in water chemistry is one of the main characteristics of coastal lagoons and, at the same time, is a source of uncertainty when characterizing lagoons (Perillo et al. 2009). For this reason, spot-measurements of water chemistry may be inadequate for the characterization of a system (Gómez-Rodríguez et al. 2009). To avoid this problem, the characterization of Balearic lagoons was based upon median nutrient concentrations, which have the advantage of being less influenced by sudden peaks in nutrient concentrations that may affect other measures of central tendency, such as mean values. Notwithstanding, this is advisable to check extreme values to ensure that they respond to natural variability instead of to anthropogenic disturbances. During this study period, peaks and troughs in nutrient concentration were usually associated with specific weather events (e.g., droughts, downpours) that either diluted or concentrated nutrients. For example, the observed increase in DIN concentration in winter was likely related to increased rainfall during this period which led to increased runoff and leaching of nitrogen into lagoons. A TP peak was detected in February, which was probably related to changes in water levels. In January, unusually low precipitation for the winter season (comparable to the June or September) was recorded and led to decreases in the depth of many lagoons. This phenomenon can lead to a subsequent process of concentration and nutrient dilution. As mentioned above, this temporal variability in nutrient concentration (primarily nitrogen and to a lesser extent phosphorus) was not surprising, as it is one of the main characteristics of coastal lagoons (Nixon 1995; Cloern 2001). However, we think it is important to exhaustively depict temporal patterns (i.e. on a monthly basis) in this kind of lagoon while there is no consensus on how to deal with such large variability in practice.

Salinity has long been regarded as an important factor for the characterization of coastal lagoons and has been used broadly in abiotic-based typologies (IUBS 1959; WFD - 2000/60/EC; Lucena-Moya et al. 2009). However, our results show that typing lagoons using salinity should be done with caution since typologies done at different times of year could lead to different classifications due to temporal variation in salinity. This inconsistency in classification violates one of the basic principles of typology development: types must be invariant over time to ensure that any action based on waterbody types has a solid foundation (e.g., WFD implementation). Consequently, we recommend avoiding the use of spot-measurements of salinity to type lagoons and, instead, relying on measures of central tendency. Despite the shortcomings of salinity that have been noted, using central tendency values would allow the use of salinity as an appropriate descriptor of these lagoons and, thus, take advantage of its ecological properties. Salinity is considered a main driver of biological community and structure in transitional waters (e.g., Williams 1998). Moreover, from a practical standpoint, salinity is a parameter that is cheap and easy to measure and it has been widely used in the classification of transitional waters (e.g., Boix et al. 2005; Telesh and Khlebovich 2010) and thereby allows for comparisons between regions and systems.

The Balearic lagoons appear to have a good chemical status as their nutrient concentrations were below the thresholds set in the European Directives focused on water quality (74/440/EEC; 76/464/EC; 78/659/EC; 80/68/EEC; 98/15/EC). Notwithstanding this, it should be noted that some extremely high values were measured at particular sampling dates. High nitrogen concentrations (above European Directives thresholds) were found in 11 % of the coastal lagoons during winter (November-February), while phosphorus levels exceeding European thresholds were observed in 63 % of lagoons at some point in time, most often in February. The EEA (1999) and Bricker et al. (2003) classification systems, which are based on nutrient contents, evaluated most of the coastal lagoons positively. However, conservation assessments based on these water chemistry thresholds may be deceptive. A previous evaluation of these lagoons based on the invertebrate community (MIBIIN index from Lucena-Moya and Pardo 2012) showed a wide range of ecological classes, from very bad to highly preserved lagoons. This lack of correspondence between chemical and ecological classification systems might be due to (i) the temporal scale being different between both approaches as the invertebrate community reflects environmental condition across longer time scales (i.e., seasonal or even annual) than nutrients (Ritchie 2009). This cause is likewise influenced by a second factor, (ii) the MIBIIN index responds to a large variety of anthropogenic disturbances (e.g., salinization processes, hydro-morphological changes), and not just to nutrient loading. Alternatively, (iii) the MIBIIN index was specifically designed for coastal lagoons, while the thresholds of the EEA (1999) and Bricker et al. (2003) were not specifically designed for them, but for transitional waters in general, including estuaries, fjords and coastal waters. This is one of the main issues for nutrient assessments of coastal lagoons that needs to be solved. A large-scale study or a meta-analysis, including Balearic and other Mediterranean lagoons, should be the basis for providing specific nutrient benchmarks for this ecoregion.

Here, we have shown the need to explicitly account for temporal variability in the classification of coastal lagoons types and to infer their ecological status based on water chemistry. Inherent temporal variation of salinity and nutrient concentration in coastal lagoons should be considered a key issue for any management action, especially in the implementation of the WFD. Notwithstanding this, our study was carried out for only a 1 year period hence further research over an inter-annual period would provide a more general picture of the factors involved. Even so, the results presented in this paper suggest that the special characteristics of the Mediterranean climate (i.e. extreme temporal variability), in addition to the singular characteristic of coastal lagoons, make the Mediterranean coastal lagoons especially complex, unpredictable, and vulnerable to external forces (e.g. hydrometeorological factors, anthropogenic activities).

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