



**UNIVERSIDADE DO ALGARVE**  
**FACULDADE DE CIÊNCIAS E TECNOLOGIA**

*Environmental conditions and biotic communities  
in Foz de Almargem and Salgados coastal lagoons,  
Algarve (South Portugal)*

**Susana Isabel Eusébio Coelho**

Tese para obtenção do grau de Doutor em Ciências Biológicas  
(Especialidade em Ecologia das Comunidades)

Trabalho efectuado sob a orientação de:  
Professora Doutora Sofia Gamito, Universidade do Algarve  
Professor Doutor Angel Pérez-Ruzafa, Universidade de Múrcia

2013



*Environmental conditions and biotic communities  
in Foz de Almargem and Salgados coastal lagoons, Algarve  
(South Portugal)*

Declaração de autoria de trabalho

Declaro ser a autora deste trabalho, que é original e inédito. Autores e trabalhos consultados estão devidamente citados no texto e constam da listagem de referências incluída.

---

(Susana Isabel Eusébio Coelho)

Copyright de Susana Isabel Eusébio Coelho

A Universidade do Algarve tem o direito, perpétuo e sem limites geográficos, de arquivar e publicitar este trabalho através de exemplares impressos reproduzidos em papel ou de forma digital, ou por qualquer outro meio conhecido ou que venha a ser inventado, de o divulgar através de repositórios científicos e de admitir a sua cópia e distribuição com objectivos educacionais ou de investigação, não comerciais, desde que seja dado crédito ao autor e editor.



*Ao Pai Xico e Mãe Malyn,*

*Dedico esta tese, por todo o seu apoio incondicional, paciência, carinho e força, tendo ELES contribuído de forma fundamental para a sua conclusão.*

*Ao Avô Eusébio e Avó Joaquina,*

*Com quem, infelizmente, já não posso partilhar o fim de uma longa etapa e o início esperançoso de uma nova.*

## ACKNOWLEDGEMENTS

I would like to acknowledge my supervisors, Professor Sofia Gamito, from the University of Algarve and Professor Angel Pérez-Ruzafa, from the University of Murcia, for encouraging me to finish the writing of this manuscript.

This thesis was financed by Fundação para a Ciência e Tecnologia (PRAXIS XXI/BD/21521/99).

This thesis also had the logistic support of the former Direção Regional do Ambiente e Ordenamento do Território do Algarve (CCDR-ALGARVE), namely in the field trips for water samples collection and in some of the laboratory analyses performed on physical and chemical water parameters. A particular thank to Dr. Ana Paula Gaspar, who participated in the field trips and provided information and literature about the studied lagoons.

I would like to thank to:

- Professor Delminda Moura (CIMA), for giving permission to use the geology laboratory, for the advices on the methodology and for helping on the analyses of sediment grain-size data;
- Professor Rui Santos (CCMAR), for letting me use the laboratory equipment to perform phytopigments analyses;
- Paulo Pedro from ITUCA, for conducting nutrients analyses of water samples;
- Dr. Silvia Condinho, for helping on phytoplankton identification;
- Dr. Alexandra Marques, for helping on zooplankton identification;
- Dr. Carlos Afonso, for helping on benthic macroinvertebrate identification;
- Paulo Santana, who facilitated logistics on the geology laboratory;
- Miguel Madureira, who was always available to solve problems with laboratory equipment and materials.

I am grateful to all my friends and colleagues, who shared their time with me during the field work (Dinamene Sousa, Sónia Ólim, Monica Pescaru, Raluca, José António); the laboratory work (Camané, Cecile Godinho, Raquel Machás, Aschwin Engelen, François Hubert) and solving my computer and software problems (António Encarnação).

A special thanks to all those friends that, although “faraway”, gave me strength and support me, even if it was just in a long phone call (Verita – Lisboa; Racas – Ireland & Portugal; Jorge Safara, Marco Mirinha, Carla Janeiro, Sandra Bernardo – Évora; Sonucha – Bias do Sul, Ana Quaresma & Miguel Correia – Olhão; Rui Graça – Faro; Pedro Andrade & Ana Sara – Sevilha & Cerro do Guelhim).



## ABSTRACT

The present study intended to compare environmental conditions and biotic communities of two choked coastal lagoons located in the Algarve region, Foz de Almargem and Salgados, with the purpose of evaluating the effects of organic pollution from wastewater discharges in water quality and biotic communities from different levels of the food chain, namely phytoplankton and benthic macroinvertebrates.

Both lagoons were seasonally connected to the sea, but most of the year they were isolated receiving the freshwater input from small rivers and wastewater (in Salgados). Data were collected from June 2001 to July 2002 in three sampling stations, according to a gradient of proximity with the sea.

Characterization of environmental conditions was performed based on the study of hydrological parameters, physical and chemical water parameters, and sediment parameters.

Phytoplankton communities were analyzed in terms of phytopigments concentrations; *taxa* composition, richness, abundance, diversity and salinity tolerance. Relations between phytoplankton and water parameters were analyzed through bivariate and multivariate statistical techniques. Results indicated that the two lagoons presented different phytoplankton communities, which were associated to water parameters and particularly with organic pollution in the case of Salgados lagoon.

Taxonomic composition, richness, abundance and diversity of benthic macroinvertebrate communities were also characterized, just as ecological features concerning the salinity tolerance of *taxa*, trophic groups and sensitivity to an increasing stress gradient. Relations between benthic macroinvertebrate parameters, water and sediment parameters were analyzed with the same techniques used with phytoplankton. Results also revealed distinct benthic macroinvertebrate communities in Foz de Almargem and Salgados lagoon, which were related with the environmental parameters that indicated a greater organic pollution in Salgados lagoon.

Water quality and trophic state were evaluated using different indices and approaches that classified Salgados lagoon as a hypereutrophic system with bad water quality. Most indices pointed out to mesotrophic conditions in Foz de Almargem, although they were not always concordant.

Keywords: coastal lagoons, water quality, trophic state, phytoplankton communities, benthic macroinvertebrate communities



## RESUMO

O presente estudo teve como principal objectivo a comparação das condições ambientais e das comunidades bióticas em duas lagoas costeiras, Foz de Almargem e Salgados, localizadas na região do Algarve. Pretenderam-se avaliar os efeitos da poluição orgânica, derivada da descarga de efluentes de estações de tratamento de águas residuais (ETAR), na qualidade da água e nas comunidades de organismos pertencentes a diferentes níveis da cadeia trófica, nomeadamente no fitoplâncton e nos macroinvertebrados bentónicos.

A selecção destas lagoas como áreas de estudo teve que ver com o facto de serem as únicas lagoas da região com a mesma tipologia hidrológica e de uma das lagoas receber os efluentes de uma ETAR (lagoa dos Salgados), enquanto que na lagoa da Foz de Almargem, aparentemente não existiam fontes directas de poluição orgânica. Este tipo de lagoas permanece isolado durante a maior parte do ano, ficando temporariamente abertas ao mar aquando do rompimento natural ou artificial do cordão dunar, que acontece durante os períodos de maior pluviosidade e consequente aumento do nível de água nas lagoas, conjugado com as condições marítimas. Geralmente, os períodos de ligação das lagoas ao mar são de curta duração, devido à rápida recuperação do cordão dunar.

O estudo decorreu entre Junho de 2001 e Julho de 2002, tendo-se efectuado recolha de amostras e medições *in situ* dos diversos parâmetros em três locais seleccionados ao longo de um gradiente de proximidade com o mar. As amostragens foram realizadas com uma frequência aproximada de mês e meio.

Em termos hidrológicos, analisou-se a variação do nível de água nas lagoas em função da precipitação e da ligação das lagoas ao mar através das aberturas naturais ou artificiais da barra arenosa.

A caracterização da qualidade da água foi feita com base em parâmetros físico-químicos, tendo-se considerado igualmente as concentrações de clorofila *a*, por ser um dos indicadores utilizados na definição do estado trófico e da qualidade dos ecossistemas lagunares.

O sedimento foi caracterizado em termos de granulometria, conteúdo de matéria orgânica e concentração de fitopigmentos.

As comunidades fitoplanctónicas e as comunidades de organismos macroinvertebrados bentónicos foram estudadas em termos de composição taxonómica, abundância, riqueza, diversidade e tolerância à salinidade. No caso dos organismos bentónicos abordaram-se ainda alguns aspectos da ecologia das espécies, nomeadamente o regime trófico e a sua sensibilidade no que diz respeito a um gradiente de stress ambiental.

As relações entre os parâmetros ambientais e as comunidades bióticas foram avaliadas através de técnicas de análise bivariada e multivariada.

Na lagoa dos Salgados observaram-se maiores concentrações de fosfatos, fósforo total, nitritos, amónia, azoto inorgânico total, sólidos suspensos totais, clorofila *a* e feopigmentos. A salinidade, a concentração de nitratos e a diversidade pigmentar foram superiores na lagoa da Foz de Almargem.

As comunidades fitoplanctónicas diferiram consideravelmente nas duas lagoas. Na lagoa dos Salgados observaram-se maiores concentrações de fitopigmentos, uma maior abundância fitoplanctónica e a dominância de Cyanophyceae, em alternância com Bacillariophyceae e algas pico-nano flageladas. Na lagoa da Foz de Almargem dominaram as Dinophyceae, Bacillariophyceae e algas pico-nano flageladas. Em termos de tolerância à salinidade, a comunidade da lagoa dos Salgados apresentou maioritariamente *taxa* de ambientes dulciaquícolas e salobros, enquanto a lagoa da Foz de Almargem registou maiores abundâncias de *taxa* que ocorrem em ambientes salobros e marinhos.

As principais diferenças nas comunidades fitoplanctónicas das lagoas estão relacionadas com os parâmetros físico-químicos das lagoas. As abundâncias de Dinophyceae e Bacillariophyceae apresentaram uma relação positiva com a razão N:P e a concentração de nitratos, estando negativamente associadas à concentração de ortofosfatos. As Cyanophyceae e as Chlorophyceae apresentaram relações inversas com estes parâmetros. As maiores abundâncias de pico-nano flagelados estiveram associadas a menores valores de temperatura e maiores concentrações de nitritos.

Em ambas as lagoas foram identificados *taxa* potencialmente tóxicos, cuja abundância se correlacionou significativamente com as concentrações de compostos azotados e compostos fosfatados. A ocorrência destes *taxa* e o desenvolvimento de florescências é particularmente preocupante na lagoa dos Salgados, quer em termos de saúde pública quer em termos de impacto na fauna piscícola e avifauna locais.

Relativamente às comunidades de organismos macroinvertebrados bentónicos foram igualmente encontradas diferenças nas duas lagoas, ao nível da composição taxonómica, densidades e tolerância à salinidade. Na lagoa dos Salgados, os grupos mais representativos em termos de densidades foram os Insecta e os Crustacea, sendo a comunidade constituída principalmente por organismos de ambientes salobros e de influência continental. Na lagoa da Foz de Almargem foram determinadas maiores densidades de Gastropoda, Bivalvia e Polychaeta, tendo grande parte dos organismos afinidades eurialinas marinhas. Em termos tróficos, ambas as lagoas apresentaram comunidades maioritariamente compostas por organismos detritívoros, apesar da diversidade de grupos tróficos na lagoa da Foz de Almargem ter sido superior. Relativamente à sensibilidade dos *taxa* bentónicos, nas duas lagoas predominaram organismos tolerantes à poluição orgânica; porém na lagoa dos Salgados, durante os meses de Primavera e de Verão de 2002, observou-se um incremento notável na densidade de organismos oportunistas, indicadores de desequilíbrios ambientais e excesso de matéria orgânica.

As principais diferenças observadas nas comunidades bentónicas das lagoas estão relacionadas quer com parâmetros da água quer com parâmetros do sedimento. As densidades de Gastropoda, Bivalvia e Polychaeta apresentaram uma associação negativa com as concentrações de fósforo total e clorofila *a* na água, assim como com o conteúdo de argilas e a concentração de clorofila *a* no sedimento. Os Insecta, Oligochaeta e Amphipoda apresentaram associações positivas com estes mesmos parâmetros e com a concentração total de azoto dissolvido na água. Em ambas as lagoas foram observadas variações sazonais que estiveram associadas à temperatura da água.

Com base nos parâmetros estudados e na determinação de índices de qualidade da água e estado trófico, a lagoa da Foz de Almargem foi considerada mesotrófica, tendo como principal fonte de nutrientes de origem antropogénica a escorrência agrícola e eventualmente a contaminação difusa proveniente do lençol freático. A lagoa dos Salgados foi classificada como hipereutrófica, tendo o enriquecimento de nutrientes estado associado às descargas de efluentes da ETAR, às escorrências agrícola e de um campo de golfe.

Palavras-chave: lagoas costeiras, qualidade da água, estado trófico, comunidades fitoplanctónicas, comunidades de macroinvertebrados bentónicos





2.3.2.6. Trophic state and water quality	63
2.3.3. Comparison of the two coastal lagoons	68
2.3.3.1. Hydrological aspects	68
2.3.3.2. Water parameters	69
2.3.3.3. Sediment parameters	76
2.3.3.4. Relations between environmental parameters	80
2.3.3.5. Comparison of environmental parameters during isolation and connection of the lagoon with the sea	83
2.3.3.6. Trophic state and water quality	84
<b>3. PHYTOPLANKTON COMMUNITIES</b>	<b>89</b>
<b>3.1. Specific Aims</b>	<b>89</b>
<b>3.2. Material and methods</b>	<b>89</b>
3.2.1. Field procedures and laboratory analyses	89
3.2.2. Data analysis	90
<b>3.3. Results and discussion</b>	<b>91</b>
3.3.1. Foz de Almargem coastal lagoon	91
3.3.1.1. Phytoplankton communities	91
3.3.1.2. Environmental parameters and phytoplankton communities	100
3.3.1.3. Phytoplankton communities during isolation and connection of the lagoon with the sea	102
3.3.2. Salgados coastal lagoon	104
3.3.2.1. Phytoplankton communities	104
3.3.2.2. Environmental parameters and phytoplankton communities	117
3.3.2.3. Phytoplankton communities during isolation and connection of the lagoon with the sea	120
3.3.3. Comparison of the two coastal lagoons	121
3.3.3.1. Phytoplankton communities	121
3.3.3.2. Environmental parameters and phytoplankton communities	127
3.3.3.3. Comparison of phytoplankton communities during isolation and connection of the lagoons with the sea	132
3.3.3.4. Phytoplankton communities according to salinity preferences	134
3.3.3.5. Potentially harmful phytoplankton	139
3.3.3.6. Phytoplankton communities and water quality	144
<b>4. BENTHIC MACROINVERTEBRATE COMMUNITIES</b>	<b>146</b>
<b>4.1. Specific Aims</b>	<b>146</b>
<b>4.2. Material and Methods</b>	<b>146</b>
4.2.1. Field sampling and laboratory procedures	146
4.2.2. Data analysis	147
<b>4.3. Results and discussion</b>	<b>150</b>
4.3.1. Foz de Almargem coastal lagoon	150

4.3.1.1. Benthic macroinvertebrate communities	150
4.3.1.2. Environmental parameters and benthic macroinvertebrate communities	163
4.3.1.3. Comparison of benthic macroinvertebrate communities during isolation and connection of the lagoon with the sea	167
4.3.2. Salgados coastal lagoon	170
4.3.2.1. Benthic macroinvertebrate communities	170
4.3.2.2. Environmental parameters and benthic macroinvertebrate communities	180
4.3.2.3. Comparison of benthic macroinvertebrate communities during isolation and connection of the lagoon with the sea	183
4.3.3. Comparison of the two coastal lagoons	185
4.3.3.1. Benthic macroinvertebrate communities	185
4.3.3.2. Environmental parameters and benthic macroinvertebrate communities	193
4.3.3.3. Comparison of benthic communities during isolation and connection of the lagoons with the sea	197
4.3.3.4. Benthic macroinvertebrate community's preference/tolerance to salinity	199
<b>5. GENERAL DISCUSSION AND CONCLUSIONS</b>	<b>202</b>
5.1. Hidrological aspects	202
5.2. Water parameters	203
5.3. Phytoplankton	206
5.3.1. Chlorophyll <i>a</i> concentration	206
5.3.2. Phytoplankton communities	206
5.3.3. Potentially harmful phytoplankton	208
5.3.4. Phytoplankton salinity tolerance	210
5.4. Sediment parameters	211
5.5. Benthic macroinvertebrates	213
5.5.1. Communities structure	213
5.5.2. Benthic trophic groups	216
5.5.3. Benthic ecological groups	217
5.5.4. Benthic macroinvertebrate salinity tolerance	217
5.6. Lagoons inlets open <i>versus</i> closed	218
5.7. Trophic state and ecological quality	220
<b>6. REFERENCES</b>	<b>226</b>
<b>APPENDICES</b>	<b>235</b>

# LIST OF FIGURES

## 1. INTRODUCTION, GENERAL AIMS AND STUDY AREAS

Figure 1.1 – Location of Foz de Almagem and Salgados lagoons in the south coast of Algarve.	6
Figure 1.2 – Foz de Almagem coastal lagoon with high (left side) and low (right side) water level.	7
Figure 1.3 – Salgados coastal lagoon with high (left side) and low (right side) water level.	8
Figure 1.4 – Typology of transitional waters and sheltered coastal water.	10

## 2. ENVIRONMENTAL CONDITIONS: HYDROLOGY, WATER, SEDIMENT AND TROPHIC STATE

Figure 2.1 - Variation of daily rainfall registered in Loulé meteorological station and Foz de Almagem coastal lagoon openings to the sea.	17
Figure 2.2 - Seasonal variation of physical and chemical water parameters (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Foz de Almagem sampling stations.	19
Figure 2.3 - Daily variation of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) during spring (May 2002) in the upstream and downstream sampling stations from Foz de Almagem coastal lagoon.	20
Figure 2.4 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration and N: P ratio (DIN: orthophosphate) in Foz de Almagem sampling stations.	22
Figure 2.5 - Seasonal variation of photosynthetic pigments concentration (chlorophyll <i>a</i> and phaeo-pigments) and Margalef's pigment diversity index in Foz de Almagem sampling stations.	23
Figure 2.6 - Principal Component Analysis performed on the hydrological and water parameters from Foz de Almagem sampling stations.	25
Figure 2.7 - Ternary diagram for textural classification of sediments from Foz de Almagem coastal lagoon, based on sand, silt and clay ratios.	26
Figure 2.8 - Seasonal variation of individual grain size fractions in the tree sampling stations from Foz de Almagem coastal lagoon.	27
Figure 2.9 - Seasonal variation of water content, organic matter content, photosynthetic pigments concentration (chlorophyll <i>a</i> and phaeo-pigments), chlorophyll <i>a</i> degradation index (% phaeo-pigments) and Margalef's pigment diversity index in the sediment of the tree sampling stations from Foz de Almagem coastal lagoon.	29
Figure 2.10 - Principal component analysis performed on sediment parameters from Foz de Almagem coastal lagoon.	31
Figure 2.11 - Principal component analysis performed on water and sediment parameters from Foz de Almagem coastal lagoon.	33
Figure 2.12 - Seasonal variation of chlorophyll <i>a</i> in the sediment, chlorophyll <i>a</i> in the water and percentage of each relative to total chlorophyll <i>a</i> in Foz de Almagem sampling stations.	35
Figure 2.13 – Seasonal variation of trophic state and water quality indexes in Foz de Almagem coastal lagoon: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX.	39
Figure 2.14 – Linear regression between chlorophyll <i>a</i> concentration ( $\mu\text{g L}^{-1}$ ) and the concentrations of nitrites ( $\mu\text{M}$ ) and nitrates ( $\mu\text{M}$ ), during the period Foz de Almagem lagoon was closed.	43
Figure 2.15 - Variation of daily rainfall registered in Algoz meteorological station and daily water level measured in the deepest part of Salgados coastal lagoon.	44
Figure 2.16 - Seasonal variation of physical and chemical water parameters (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Salgados sampling stations.	46

Figure 2.17 - Daily variation of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) during spring (May 2002) in the intermediate and downstream sampling stations from Salgados coastal lagoon.	47
Figure 2.18 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration, N: P ratio (DIN: orthophosphates) and total solids in suspension (TSS) in Salgados sampling stations.	49
Figure 2.19 - Seasonal variation of photosynthetic pigments concentration (chlorophyll <i>a</i> and phaeo-pigments) and Margalef's pigment diversity index in Salgados sampling stations.	50
Figure 2.20 - Principal Component Analysis performed on the hydrological and water parameters from Salgados sampling stations.	52
Figure 2.21 - Ternary diagram for textural classification of sediments from Salgados coastal lagoon, based on sand, silt and clay ratios.	53
Figure 2.22 - Seasonal variation of individual grain size fractions in the two sampling stations from Salgados coastal lagoon.	53
Figure 2.23 - Seasonal variation of water content, organic matter content, photosynthetic pigments concentration (chlorophyll <i>a</i> and phaeo-pigments), chlorophyll <i>a</i> degradation index (% phaeo-pigments) and Margalef's pigment diversity index in the sediment of the two sampling stations from Salgados lagoon.	55
Figure 2.24 - Principal component analysis performed on sediment parameters from Salgados lagoon.	56
Figure 2.25 - Principal component analysis performed on environmental parameters from Salgados coastal lagoon.	59
Figure 2.26 - Seasonal variation of chlorophyll <i>a</i> in the sediment, chlorophyll <i>a</i> in the water and percentage of each relative to total chlorophyll <i>a</i> in Salgados sampling stations.	61
Figure 2.27 - Seasonal variation of trophic state and water quality indexes in Salgados coastal lagoon: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX.	65
Figure 2.28 - Variation of monthly rainfall registered in Foz de Almargem (Loulé meteorological station) and Salgados (Algoz meteorological station) and duration of the connection between the lagoons and the sea (number of days <i>per</i> month).	68
Figure 2.29 - Seasonal variation of physical and chemical water parameters mean values (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Foz de Almargem and Salgados coastal lagoons.	70
Figure 2.30 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration and N: P ratio (DIN: orthophosphate) mean values in Foz de Almargem and Salgados coastal lagoons.	72
Figure 2.31 - Seasonal variation of photosynthetic pigments mean concentrations (chlorophyll <i>a</i> and phaeo-pigments) and Margalef's pigment diversity index in Foz de Almargem and Salgados coastal lagoons.	73
Figure 2.32 - Principal Component Analysis performed on the hydrological and water parameters mean values from Foz de Almargem and Salgados coastal lagoons.	75
Figure 2.33 - Seasonal variation of mean sand, silt and clay contents in Foz de Almargem and Salgados coastal lagoons.	76
Figure 2.34 - Seasonal variation of mean values for water content, organic matter content, photosynthetic pigments concentration (chlorophyll <i>a</i> and phaeo-pigments), chlorophyll <i>a</i> degradation index (% phaeo-pigments) and Margalef's pigment diversity index in Foz de Almargem and Salgados coastal lagoons.	78
Figure 2.35 - Principal Component Analysis performed on the sediment parameters mean values from Foz de Almargem and Salgados coastal lagoons.	79
Figure 2.36 - Principal Component Analysis performed on the environmental parameters mean values from Foz de Almargem and Salgados coastal lagoons.	81

Figure 2.37 - Seasonal variation of chlorophyll <i>a</i> in the sediment, chlorophyll <i>a</i> in the water and percentage of each relative to total chlorophyll <i>a</i> in Foz de Almargem and Salgados lagoons.	82
Figure 2.38 – Seasonal variation of trophic state and water quality indexes (means) in Foz de Almargem and Salgados coastal lagoons: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX.	85

### 3. PHYTOPLANKTON COMMUNITIES

Figure 3.1 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Foz de Almargem sampling stations.	93
Figure 3.2 - Evolution of phytoplankton abundance <i>per</i> class and relative frequency of each class in Foz de Almargem sampling stations.	94
Figure 3.3 - Seasonal variation of Dinophyceae <i>taxa</i> abundance in Foz de Almargem sampling stations.	96
Figure 3.4 - Evolution of Bacillariophyceae <i>taxa</i> abundance in Foz de Almargem sampling stations.	97
Figure 3.5 - Evolution of Chlorophyceae ( <i>Cosmarinum</i> sp.; <i>Staurastrum</i> sp.), Cryptophyceae ( <i>Cryptomonas</i> sp.; <i>Rhodomonas</i> sp.), Euglenophyceae ( <i>Eutreptiella</i> sp.) and Cyanophyceae ( <i>Anabaena flos-aqua</i> ) <i>taxa</i> abundances in Foz de Almargem sampling stations.	98
Figure 3.6 - Canonical correspondence analysis performed with the phytoplankton groups (total density <i>per</i> station) from Foz de Almargem sampling stations.	102
Figure 3.7 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Salgados sampling stations.	106
Figure 3.8 - Evolution of phytoplankton abundance <i>per</i> class and relative frequency of each class in Salgados sampling stations.	107
Figure 3.9 - Evolution of Cyanophyceae <i>taxa</i> abundance in Salgados sampling stations.	109
Figure 3.10 - Evolution of Bacillariophyceae <i>taxa</i> abundance in Salgados sampling stations.	110
Figure 3.11 - Evolution of Chlorophyceae <i>taxa</i> abundance in Salgados sampling stations.	112
Figure 3.12 - Evolution of Euglenophyceae <i>taxa</i> abundance in Salgados sampling stations.	113
Figure 3.13 - Evolution of Cryptophyceae and Dinophyceae <i>taxa</i> abundances in Salgados sampling stations.	115
Figure 3.14 - Canonical correspondence analysis performed with the phytoplankton groups (total density <i>per</i> station) from Salgados sampling stations.	119
Figure 3.15 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) monthly mean values in Foz de Almargem and Salgados lagoons.	123
Figure 3.16 - Evolution of phytoplankton mean abundance <i>per</i> class and relative frequency of each class in Foz de Almargem and Salgados lagoons.	124
Figure 3.17 - Monthly mean abundances of phytoplankton <i>taxa</i> that occurred in both lagoons.	127
Figure 3.18 - Canonical correspondence analysis performed with the phytoplankton groups (mean density) from Foz de Almargem and Salgados lagoon.	130
Figure 3.19 – Evolution of taxonomic richness and phytoplankton abundance according to <i>taxa</i> salinity preferences in Foz de Almargem and Salgados lagoons.	138
Figure 3.20 - Monthly mean abundances of potentially harmful <i>taxa</i> in Foz de Almargem and Salgados lagoons.	141
Figure 3.21 - Monthly mean abundances of potentially harmful <i>taxa</i> in Foz de Almargem and Salgados lagoons, according to <i>taxa</i> occurrence in terms of <i>habitats</i> .	142

#### 4. BENTHIC MACROINVERTEBRATE COMMUNITIES

Figure 4.1 - Evolution of total benthos density, taxa richness, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Foz de Almargem sampling stations.	152
Figure 4.2 - Seasonal variation of the main taxonomic benthos groups densities and relative frequency of each group in Foz de Almargem sampling stations.	153
Figure 4.3 - Seasonal variation of Polychaeta <i>taxa</i> densities in Foz de Almargem sampling stations.	157
Figure 4.4 - Seasonal variation of Mollusca <i>taxa</i> densities ( <i>Gastropoda Ventrosia ventrosa</i> and <i>Hydrobia ulvae</i> ; <i>Bivalvia Abra segmentum</i> and <i>Cerastoderma glaucum</i> ) in Foz de Almargem sampling stations.	157
Figure 4.5 – Relative frequency of <i>taxa</i> in terms of constancy and fidelity in Foz de Almargem sampling stations.	158
Figure 4.6 - Seasonal variation of trophic groups densities and relative frequency of each group in Foz de Almargem sampling stations.	162
Figure 4.7 - Seasonal variation of ecological AMBI groups densities and relative frequency of each group in Foz de Almargem sampling stations.	163
Figure 4.8 - Canonical correspondence analysis performed with the benthic macroinvertebrate <i>taxa</i> (total density per station) from Foz de Almargem lagoon.	167
Figure 4.9 - Evolution of total benthos density, taxa richness, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Salgados sampling stations.	171
Figure 4.10 - Seasonal variation of the main taxonomic benthos groups densities and relative frequency of each group in Salgados sampling stations.	172
Figure 4.11 - Seasonal variation of Insecta <i>taxa</i> densities in Salgados sampling stations.	174
Figure 4.12 - Seasonal variation of Oligochaeta <i>taxa</i> densities in Salgados sampling stations.	175
Figure 4.13 - Seasonal variation of Crustacea <i>taxa</i> densities in Salgados sampling stations.	175
Figure 4.14 – Relative frequency of <i>taxa</i> in terms of constancy and fidelity in Salgados sampling stations.	176
Figure 4.15 - Seasonal variation of trophic groups densities and relative frequency of each group in Salgados sampling stations.	178
Figure 4.16 - Seasonal variation of ecological AMBI groups densities and relative frequency of each group in Salgados sampling stations	179
Figure 4.17- Canonical correspondence analysis performed with the benthic macroinvertebrate <i>taxa</i> (total density per station) from Salgados lagoon.	182
Figure 4.18 - Evolution of benthic macro invertebrate richness, total density, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) monthly mean values in Foz de Almargem and Salgados lagoons.	186
Figure 4.19 - Seasonal variation of the main taxonomic benthic groups' densities and relative frequency of each group in Foz de Almargem and Salgados lagoons.	188
Figure 4.20 - Monthly mean abundances of benthic macroinvertebrate <i>taxa</i> that occurred in both lagoons.	188
Figure 4.21 - Seasonal variation of trophic groups densities and relative frequency of each group in Foz de Almargem and Salgados lagoons.	192
Figure 4.22 - Seasonal variation of ecological AMBI groups densities and relative frequency of each group in Foz de Almargem and Salgados lagoons.	193
Figure 4.23- Canonical correspondence analysis performed with the benthic macroinvertebrate groups (mean density) from Foz de Almargem and Salgados lagoon	196
Figure 4.24 – Evolution of taxonomic richness and density of benthic macroinvertebrate <i>taxa</i> according to salinity preferences in Foz de Almargem and Salgados lagoons.	200

# LIST OF TABLES

## 1. INTRODUCTION, GENERAL AIMS AND STUDY AREAS

Table 1.1 – Resume of the general characteristics of Foz de Almargem and Salgados lagoons.	10
--	----

## 2. ENVIRONMENTAL CONDITIONS: HYDROLOGY, WATER, SEDIMENT AND TROPHIC STATE

Table 2.1- Water level in the lagoon and periods of isolation, semi-isolation and connection between Foz de Almargem coastal lagoon and the sea.	18
Table 2.2 – Minimum, maximum and mean values of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) monitored during a 24 hours cycle in spring (May 2002) at the upstream and downstream sampling stations from Foz de Almargem lagoon.	21
Table 2.3 – Annual mean values and standard deviation of water parameters in Foz de Almargem sampling stations.	24
Table 2.4 – Annual mean values and standard deviation of sediment parameters in Foz de Almargem sampling stations.	30
Table 2.5 - Significant correlations between water and sediment parameters from Foz de Almargem coastal lagoon.	32
Table 2.6 – Water and sediment parameters in January 2002 and variation between values when the lagoon was closed (December 2001) and opened to the sea (January 2002) in Foz de Almargem sampling stations.	37
Table 2.7 – Trophic state and water quality indexes mean values, standard deviation and classification in Foz de Almargem sampling stations, determined during all studied period (June 2001-July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).	40
Table 2.8 – Water quality in Foz de Almargem sampling stations, based on the 90 <sup>th</sup> percentile of chlorophyll <i>a</i> ( $\mu\text{g L}^{-1}$ ), adapted from Brito <i>et al.</i> (2012) and Pereira Coutinho <i>et al.</i> (2012).	42
Table 2.9 - Periods of connection with the sea and water level in Salgados coastal lagoon.	44
Table 2.10 – Minimum, maximum and mean values of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) monitored during a 24 hours cycle in spring (May 2002) at the intermediate and downstream sampling stations from Salgados lagoon.	47
Table 2.11 – Annual mean values and standard deviation of water parameters in Salgados sampling stations.	50
Table 2.12 – Annual mean values and standard deviation of sediment parameters in Salgados sampling stations.	55
Table 2.13 - Significant correlations between water and sediment parameters from Salgados lagoon.	58
Table 2.14 – Water and sediment parameters in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.	62
Table 2.15 – Trophic state and water quality indexes mean values, standard deviation and classification in Salgados sampling stations, determined during all studied period (June 2001-July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).	65
Table 2.16 – Water quality in Salgados sampling stations, based on the 90 <sup>th</sup> percentile of chlorophyll <i>a</i> ( $\mu\text{g L}^{-1}$ ), adapted from Brito <i>et al.</i> (2012) and Pereira Coutinho <i>et al.</i> (2012).	67
Table 2.17 – Annual mean values and standard deviation of water parameters in Foz de Almargem and Salgados coastal lagoons.	74
Table 2.18 – Annual mean values and standard deviation of sediment parameters in Foz de	77

Almargem and Salgados coastal lagoons.

Table 2.19 – Water and sediment parameters in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).	83
Table 2.20 – Trophic state and water quality indexes mean values, standard deviation and classification in Foz de Almargem and Salgados lagoons, determined during all studied period (June 2001-July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).	86
Table 2.21 – Water quality in Foz de Almargem and Salgados lagoons, based on the 90 <sup>th</sup> percentile of chlorophyll <i>a</i> ( $\mu\text{g L}^{-1}$ ), adapted from Brito <i>et al.</i> (2012) and Pereira Coutinho <i>et al.</i> (2012).	88

### 3. PHYTOPLANKTON COMMUNITIES

Table 3.1 – Annual mean annual and standard deviation of phytoplankton parameters in Foz de Almargem sampling stations.	99
Table 3.2 - Significant correlations between phytoplankton and environmental parameters in Foz de Almargem lagoon.	101
Table 3.3 – Phytoplankton communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Foz de Almargem sampling stations.	103
Table 3.4 – Annual mean values and standard deviation of phytoplankton parameters in Salgados sampling stations.	116
Table 3.5 - Significant correlations between phytoplankton and environmental parameters in Salgados lagoon.	118
Table 3.6 – Phytoplankton communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.	121
Table 3.7 – Annual mean values and standard deviation of phytoplankton parameters in Foz de Almargem and Salgados lagoons.	128
Table 3.8- Resume of the significant correlations between phytoplankton communities and environmental parameters in Foz de Almargem and Salgados lagoons.	129
Table 3.9 - Significant correlations between environmental parameters and the abundances of phytoplankton <i>taxa</i> that occurred in both lagoons.	131
Table 3.10 – Phytoplankton parameters in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).	133
Table 3.11 - Significant correlations between environmental parameters and potentially harmful <i>taxa</i> from Foz de Almargem and Salgados lagoons.	143

### 4. BENTHIC MACROINVERTEBRATE COMMUNITIES

Table 4.1 – Annual mean values and standard deviation of benthic macroinvertebrate parameters in Foz de Almargem sampling stations.	159
Table 4.2 - Significant correlations between benthic macroinvertebrate and environmental parameters (water and sediment) from Foz de Almargem lagoon.	165
Table 4.3 – Benthic macroinvertebrate communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Foz de Almargem sampling stations.	169
Table 4.4 – Annual mean values and standard deviation of benthic macro invertebrate parameters in Salgados sampling stations.	177
Table 4.6 – Benthic macroinvertebrate communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.	184

Table 4.7 – Annual mean values and standard deviation of benthic macro invertebrate parameters in Foz de Almargem and Salgados lagoons.	190
Table 4.8 - Resume of the significant correlations between benthic macroinvertebrate communities and environmental parameters in Foz de Almargem and Salgados lagoons.	194
Table 4.9 – Benthic macroinvertebrate communities in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).	198
Table 4.10 - Classification of benthic macroinvertebrate <i>taxa</i> based on the salinity preference/tolerance in Foz de Almargem and Salgados lagoons.	199

## 5. GENERAL DISCUSSION AND CONCLUSIONS

Table 5.1 – Trophic state and water quality in Foz de Almargem and Salgados lagoons, based on different authors and approaches	222
--	-----

## LIST OF APPENDICES

Appendix I.A - Results of One-Way ANOVA, Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for water parameters (Ln (x+1)) in Foz de Almargem coastal lagoon.	235
Appendix I.B - Results of One-Way ANOVA, Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for sediment parameters (Ln x+1) in Foz de Almargem coastal lagoon.	236
Appendix I.C - Results of One-Way ANOVA, Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for water parameters (Ln (x+1)) in Salgados coastal lagoon.	237
Appendix I.D - Results of Mann-Whitney U test and Student T test for sediment parameters (Ln (x+1)) comparison between sites from Salgados lagoon.	238
Appendix I.E - Results from Mann-Whitney U test and Student T test for water parameters (Ln (x+1)) comparison between lagoons.	239
Appendix I.F - Results of Mann-Whitney U test and Student T test for sediment parameters (Ln (x+1)) comparison between lagoons.	240
Appendix I.G - Results of One-Way ANOVA and Kruskal-Wallis test among sites for phytoplankton parameters (Ln (x+1)) in Foz de Almargem coastal lagoon.	240
Appendix I.H - Results of One-Way ANOVA and Kruskal-Wallis test among sites for phytoplankton parameters (Ln (x+1)) in Salgados coastal lagoon.	240
Appendix I.I - Results of Mann-Whitney U test and Student T test for phytoplankton parameters (Ln (x+1)) comparison between lagoons.	241
Appendix I.K - Results of One-Way ANOVA and Kruskal-Wallis and multiple comparisons LSD Fisher test among sites for benthic macroinvertebrate parameters (Ln (x+1)) in Foz de Almargem coastal lagoon.	242
Appendix I.K - Results of Mann-Whitney U test and Student T test for benthic macroinvertebrate parameters (Ln (x+1)) in Salgados coastal lagoon.	243
Appendix I.L - Results of Mann-Whitney U test and Student T test for benthic macro invertebrate parameters (Ln (x+1)) comparison between lagoons.	243
Appendix II.A – List of phytoplankton <i>taxa</i> identified in Foz de Almargem coastal lagoon.	244
Appendix II.B – List of phytoplankton <i>taxa</i> identified in Salgados coastal lagoon.	245
Appendix II.C – List of benthic macroinvertebrate <i>taxa</i> identified in Foz de Almargem coastal lagoon.	246
Appendix II.D – List of benthic macroinvertebrate <i>taxa</i> identified in Salgados coastal lagoon.	246

# 1. INTRODUCTION, GENERAL AIMS AND STUDY AREAS

## 1.1. Introduction

Coastal lagoons were defined by Kjerfve (1994) as shallow coastal water bodies separated from the ocean by a barrier, connected at least intermittently to the ocean by one or more restricted inlets and usually oriented shore-parallel. Lagoons formed as a result of rising sea level during the Holocene or Pleistocene and the building of coastal barriers by marine processes. On a geologic time scale, they are short-lived landscape features, with an existence intrinsically linked to their filtering efficiency and the rate of relative sea-level change in response to global climatic change, local tectonic activity, and anthropogenic activities (Kjerfve, 1994).

The size of coastal lagoons varies substantially, having depths which seldom exceed a couple of meters. It may or may not be subject to tidal mixing and salinity can vary from that of a coastal fresh-water lake to a hypersaline lagoon, depending on the hydrologic balance. Hydrological features are moulded to a certain extent by the morphology of the lagoon and by the dimension of the canals through which exchange of water with the sea occurs (Bird, 1994; Kjerfve, 1994).

Kjerfve and Magill (1989) sub-divided coastal lagoons into three geomorphic types based on the water exchange with the ocean: choked lagoons, restricted lagoons and leaky lagoons. According to these authors:

- *Choked lagoons* are connected to the sea by a single long narrow entrance channel, along coasts with high wave energy and significant littoral drift. Tidal oscillations are often reduced to 5% or less as compared to the adjacent coastal tide and lagoons are characterized by long flushing times, dominant wind forcing and intermittent stratification events due to intense solar radiation or runoff events. Lagoons are mostly oriented shore-parallel but some are associated with river deltas and then occasionally oriented shore-normal.
- *Restricted lagoons* consist of a large and wide water body, usually oriented shore-parallel and exhibit two or more entrance channels or inlets. As a result, restricted coastal lagoons have a well-defined tidal circulation, are influenced by winds, mostly vertically well mixed and exhibit salinities from brackish water to

oceanic salinities. Flushing times are usually considerably shorter than for choked coastal lagoons.

- *Leaky lagoons* are elongated shore-parallel water bodies with many ocean entrance channels along coasts where tidal currents are sufficiently strong to overcome the tendencies by wave action and littoral drift to close the channel entrances. These lagoons occupy the opposite end of the spectrum from choked lagoons. Leaky lagoons are characterized by numerous wide tidal passes, unimpaired water exchange with the ocean on wave, tidal and longer time scales, strong tidal currents, and salinities close to that of the coastal ocean.

The choked lagoon type is characteristic of physically controlled ecosystems with strong fluctuations in environmental parameters depending on weather conditions (Gamito *et al.*, 2005). Some examples of portuguese choked lagoons are Foz de Almargem, Salgados, Santo André and Albufeira lagoons. In South Portugal, Ria de Alvor can be considered a restricted lagoon, although it has only one entrance channel, and Ria Formosa belongs to the leaky lagoon type.

These coastal ecosystems are highly dynamic and strongly influenced by river input, wind stress, tides, precipitation-evaporation balance, responding differently to these forcing functions (Kjerfve, 1994). Water quality and eutrophication depend critically on lagoon circulation, salt and material dispersion, water exchange with the ocean and turnover, residence or flushing times (Bird, 1994; Kjerfve, 1994).

The generated environmental stress regulates the structure of biological assemblages and leads to complex interactions among physical (light, temperature, mixing, flow), chemical (organic and inorganic carbon, oxygen, nutrients) and biological parameters and processes (nutrients uptake, predation, competition). Changes in the primary producers' structure affect secondary producers, as they are the basis of the trophic food web (Pérez-Ruzafa *et al.*, 2002; Gamito *et al.*, 2005; Viaroli *et al.*, 2008).

Coastal lagoons are commonly characterised by high productivity as they accumulate nutrients supplied by the surrounding watershed and therefore are particularly vulnerable to water quality deterioration, namely eutrophication (Taylor *et al.*, 1999; Cloern, 2001). Eutrophication is often caused by a rapid enrichment of nutrients (phosphorus and nitrogen) as a consequence of some anthropogenic activities such as

the introduction of sewage effluents and agriculture runoff. High concentrations of nutrients promote the growth of phytoplankton leading to the potential occurrence of algal blooms, generally dominated by single species some of which can be harmful due to the production of biotoxins (Tomàs- Vives, 1996; Glibert *et al.*, 2005). Algal blooms in the water column also affect benthic primary producers through shading, causing their decline. Dead material decomposition in the sediments promotes the decay of dissolved oxygen and the efflux of nutrients from the sediments to the water column, contributing to the intensification of eutrophication (Brito *et al.*, 2010, 2012).

The eutrophication process comprises four major successional stages: oligotrophic, mesotrophic, eutrophic and hypertrophic. A brief description of each state defines that (Gamito *et al.*, 2005 and references therein):

- *Oligotrophic lagoons* have low levels of nutrient concentrations in the water column and consequently there is a restriction of phytoplankton growth, keeping water at high transparency levels. Light can easily reach the bottom and is not a limiting factor for benthic vegetation.
- *Mesotrophic lagoons* are characterized by a medium level nutrient concentration in the water high enough to allow the growth of macroalgae, together with phytoplankton, as the major primary producers. Nutrients at this stage can still be assimilated by organisms, hence introducing major changes in the community structure, but keeping the water at relatively high levels of transparency. Mesotrophy can be induced by agriculture run-off and urban or industrial sewage. Nevertheless, river and groundwater inputs with high nutrients concentration, atmospheric deposition or the exchange of nutrient-enriched seawater from upwelling areas outside the lagoon can provide significant loads of nutrients. Both the planktonic and the benthic systems are affected by nutrient enrichment in the water column, increasing the competition between seagrass and macroalgae.
- *Eutrophic lagoons* present high levels of nutrients in water and sustain large assemblages of phytoplankton as dominant primary producers.
- *Hyper-eutrophic lagoons* correspond to the state in which phytoplankton assemblages increase up to the self-shadow level, preventing light from reaching the bottom and not allowing macroalgae to grow.

Water quality deterioration and eutrophication have become a major problem affecting coastal lagoons worldwide and in 2000, the European Community established as one of the main goals of the European Water Framework Directive (C.E.C., 2000), the achievement of good ecological status in coastal and transitional waters by the year 2015.

In Portugal, Bettencourt *et al.* (2004) developed guidelines for the application of the WFD concerning the definition of the typology and reference conditions for Portuguese transitional and coastal waters. Ferreira *et al.* (2007) defined a monitoring plan for water quality and ecology in Portuguese coastal waters. For restricted coastal water bodies, which include coastal lagoons, the monitoring plan proposes the surveillance of hydro-morphological elements; physic and chemical water parameters, namely thermal conditions, dissolved oxygen, salinity and nutrients; special pollutants in suspended particulate matter, sediments and tissues of fish and shellfish; and biological quality elements. The biological quality elements to monitor are focused on phytoplankton, other aquatic flora, macroinvertebrates and fish. Sampling frequency depends on the quality elements monitored. For physic and chemical water parameters, phytoplankton biomass and abundance is suggested a monthly frequency, while for phytoplankton species composition and macroinvertebrates it could be every six months (Ferreira *et al.*, 2007).

Phytoplankton communities have been used as biological indicators of water quality, once respond to changes in nutrients concentrations, water renewal, physical, chemical and biological parameters (CEMAGREF-IARE, 1994; Ferreira *et al.*, 2007; Gamito *et al.*, 2005; Brito *et al.*, 2012; Pereira Coutinho *et al.*, 2012). Phytoplankton dynamics is influenced by bottom-up and/or top-down factors (Krebs, 1994). Bottom-up factors control species growth (*e.g.* light intensity, temperature, salinity, availability of nutrients, nutrients ratio and chemical form), while top-down factors control its biomass (*e.g.* predation, competition) (Wehr and Descy, 1998).

Benthic macroinvertebrates also represent a relevant component of coastal ecosystems, playing a vital role in nutrient cycling, detrital decomposition and as a food source for higher trophic levels (Pearson and Rosenberg, 1978). Due to the relatively sessile habit and thus, the incapability to avoid unfavourable conditions, macrobenthic species are sensitive indicators of changes in the environment caused by natural or anthropogenic disturbances (Salas *et al.*, 2006). Since benthic species are relatively long-lived they

integrate water and sediment quality conditions with time and thereby, indicate temporal and chronic disturbances (Warwick *et al.*, 1990). Effects of these disturbances include changes in diversity, biomass, abundance of stress tolerant or sensitive benthic species, and changes in the trophic or functional structure of the benthic community (Pearson and Rosenberg, 1978; Reiss and Kröncke, 2005; Gamito, 2006, 2008).

## **1.2. General Aims**

The present study intended to compare environmental conditions and biotic communities of two choked coastal lagoons, with the purpose of evaluating the effects of organic pollution from wastewater discharges in water quality and biotic communities from different levels of the food chain. Thereby, three general aims were defined:

1. Characterize the environmental conditions (hydrological aspects, water parameters and sediment parameters) and evaluate the water quality and trophic state of the coastal lagoons;
2. Study the phytoplankton communities' dynamics and determine its relation with environmental parameters and the trophic state of the coastal lagoons;
3. Study the dynamics of benthic macroinvertebrate communities and determine its relation with environmental parameters and the trophic state of the coastal lagoons.

A few studies had been done before 2001 in Salgados lagoon, mainly focused on the landscape, fauna and flora characterization and management (*e.g.* Fernandes, 2001; Ministro, 2002), water quality (*e.g.* Neves, 1999) and also on the implications of artificial opening of the lagoon to the sea (*e.g.* Pinto *et al.*, 2001). For Foz de Almargem lagoon, no previous studies were available.

### 1.3. Study Areas

The selection of the study areas was based on the presence and absence of discharges from wastewater treatment plants in Salgados and Foz de Almagem, respectively.

Both lagoons are located in the south coast of Algarve and belong to the Algarve's rivers hydrographical region (Figure 1.1).

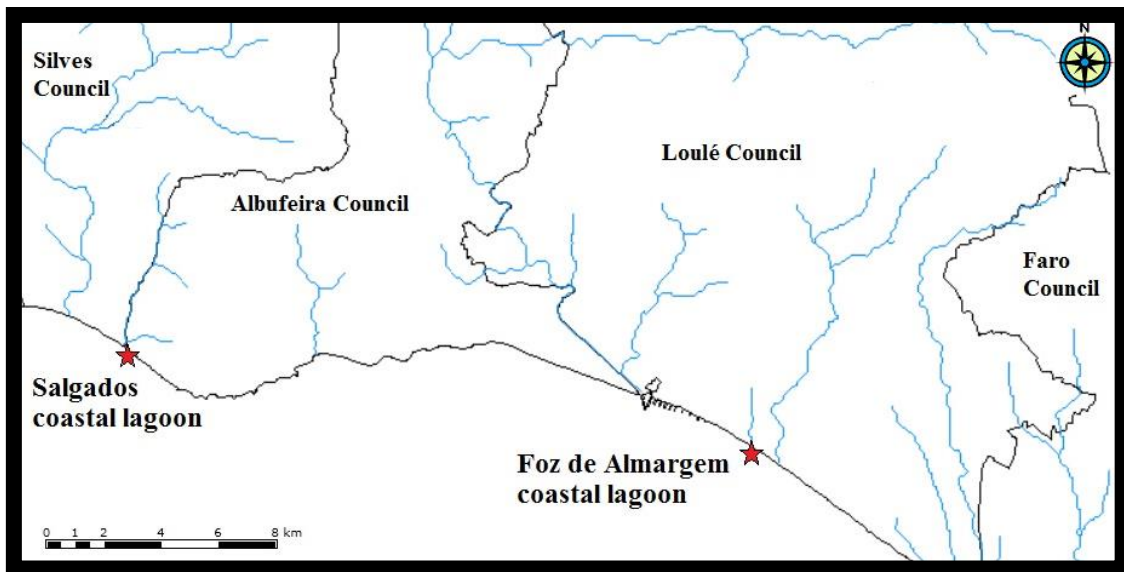


Figure 1.1 – Location of Foz de Almagem and Salgados lagoons in the south coast of Algarve. (adapted from <http://geo.snirh.pt/AtlasAgua/>)

Foz de Almagem coastal lagoon is located in the Loulé council (Lat. 37° 03` 39`` N; Long. 8° 04` 58`` W). The drainage basin comprises two small rivers, Almagem and Fonte Santa, with a seasonal dynamic strongly influenced by meteorological conditions. The lagoon is a small brackish wetland, which occupies around 20 ha, most of it shallow and only with a deeper main channel (Fig. 1.2). When all the lagoon area is flooded, the mean depth is approximately 1m and the maximum depth is about 2.5 m. Most of the year, the lagoon is isolated from the sea by a thin sand barrier, but from autumn until spring sometimes this barrier is naturally destroyed by waves or artificially by fishermen forming a channel between the lagoon and the sea. The frequency and duration of natural channel opening depends mainly on the wave height and tide amplitude. When the lagoon is opened there is tidal influence but only in the main channel, where the maximum depth is around 1.5 m; the remaining area is not flooded and the sediment becomes exposed.

No sources of organic pollution were known, besides the runoff from agriculture lands.

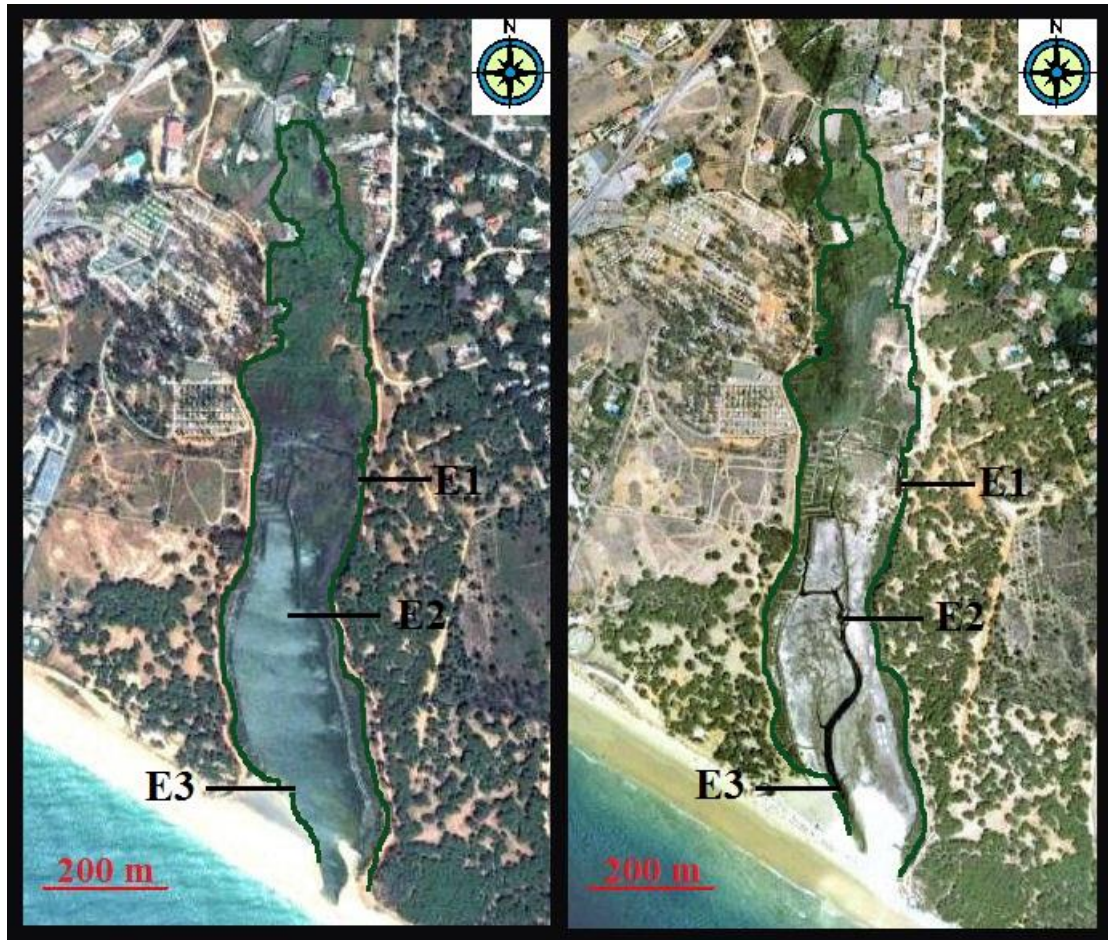


Figure 1.2 – Foz de Almagem coastal lagoon with high (left side) and low (right side) water level. *Sampling stations:* E1 – Upstream; E2 – Intermediate; E3 – Downstream (adapted from Google Earth).

Salgados coastal lagoon is a semi-enclosed brackish wetland, located in the Albufeira and Silves councils (Lat. 37° 05` 25`` N; Long. 8° 19`44`` W). The drainage basin comprises 39 Km<sup>2</sup> from two small rivers, Espiche and Vale Rabelo (Neves, 1999). The lagoon occupies a total surface of 40 ha, most of it shallow and only with a deeper area downstream. The mean depth is approximately 1 m and the maximum depth is about 5 m (Figure 1.3).

A thin sand barrier separates the lagoon from the sea and in natural conditions, when the water level in the lagoon would reach 5.7 m above the sea level, a channel was formed connecting the lagoon to the sea (Pinto *et al.*, 2001). Nowadays, a channel is artificially opened every time the water level in the lagoon rises from 4.5 to 5.0 m above the sea level. The artificial openings of the lagoon are managed by the regional environmental

services, former CCDR Algarve, with the purpose of renewing the lagoon water and preventing the flooding of a golf course located nearby. Usually, the period of connection between the lagoon and the sea does not exceed a week, depending on the freshwater inputs, the wave height and tide amplitude. When rivers runoff is high and the sea is calm, the lagoon stays open for longer time.

Besides the freshwater inputs from the small rivers, Salgados lagoon also used to receive the discharges of two wastewater treatment plants, which accounted approximately 27% of the total freshwater input in summer (Neves, 1999) and maintained the water level high in this season. These wastewater treatment plants worked inefficiently most of the time, due to the high affluence of tourists to this region all over the year. Consequently, effluents were discharged into Espiche river with concentrations of organic matter, phosphorous and nitrogen higher than those allowed by national legislation. Wastewater was therefore, the main direct source of organic pollution in the lagoon (Neves, 1999). Nevertheless, in the drainage basin there were also some sources of diffuse organic pollution namely, the runoff from agriculture lands and from a golf course implanted in one of the lagoon margins.

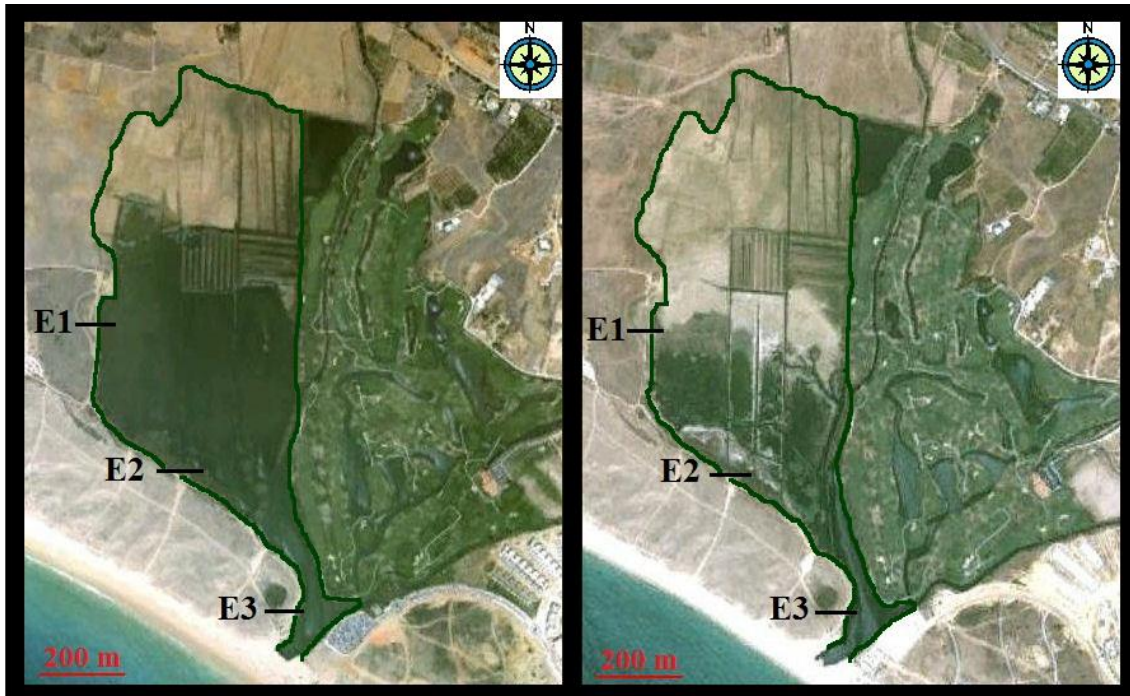


Figure 1.3 – Salgados coastal lagoon with high (left side) and low (right side) water level. Sampling stations: E1 – Upstream; E2 – Intermediate; E3 – Downstream (adapted from Google Earth).

According to the report that defined the typology and reference conditions for Portuguese transitional and coastal waters (Bettencourt *et al.*, 2004), coastal lagoons in Portugal were considered coastal waters and were divided in two typologies (Figure 1.4):

**(A3) – Mesotidal semi-enclosed lagoons.** These types of lagoons have a direct but intermittent connection with the ocean, which is frequently closed by a sand bar. Artificial opening occurs mainly in the summer months. These systems are shallow, with a mean water depth less than 2 m. Salinity varies widely and is strongly influenced by evaporation, occasional freshwater inputs (precipitation and runoff) and by cycles of temporary communication with the sea. The tidal influence on the lagoons is moderate and only occurs during periods of free connection with the ocean. Sand dunes cover the coastal and lagoon shores and extensive reed beds colonize wetland areas. Santo André, Albufeira and Óbidos lagoons were included in this typology.

**(A4) – Mesotidal shallow lagoon.** The communication between the lagoon and the sea is permanent and occurs through several inlets located along the system. The shallow depth, strong tidal currents and high water renewal make this type of lagoon vertically well-mixed. The mean water depth is about 2 m and salinity values are always above 30 since the freshwater input can be considered negligible – in summer conditions this type of system may become an inverse estuary. This type encompasses a complex of coastal seawater lagoons on sandy or muddy soils, extensive mudflats, sandbanks, sand dune systems, salt marshes, wetlands and subtidal seagrass beds. Ria Formosa and Ria de Alvor are the most significant examples in Portugal of this type of lagoon.

This classification associated the systems in southern Portugal with most Mediterranean systems, which present a hot summer Mediterranean climate with dry season and precipitation during the winter (Brito *et al.*, 2012).

Although Foz de Almarem and Salgados lagoons are located in southern Portugal (between Ria Formosa and Ria de Alvor), these lagoons are not permanently open to the sea and present characteristics of semi-enclosed lagoons.

Table 1 resumes the general characteristics of Foz de Almarem and Salgados lagoons on the perspective of the Water Framework Directive.



Proposed typology and classification of systems larger than 1 km <sup>2</sup>		
Type	Descriptor	Systems larger than 1 km <sup>2</sup>
A1	Mesotidal stratified estuary	Minho estuary Lima estuary Douro estuary Leça estuary
A2	Mesotidal well-mixed estuary with irregular river discharge	Ria de Aveiro Mondego estuary Tagus estuary Sado estuary Mira estuary Arade estuary Guadiana estuary
A3	Mesotidal semi-enclosed lagoon	Óbidos lagoon Albufeira lagoon St. André lagoon
A4	Mesotidal shallow lagoon	Ria de Alvor Ria Formosa

TICOR systems shown in blue.

Figure 1.4 – Typology of transitional waters and sheltered coastal waters (Bettencourt *et al.*, 2004).

Table 1.1 – Resume of the general characteristics of Foz de Almarem and Salgados lagoons.

	<i>Foz de Almarem lagoon</i>	<i>Salgados lagoon</i>
Latitude	37° 03' 39" N	37° 05' 25" N
Longitude	8° 04' 58" W	8° 19' 44" W
Type	Coastal Water	
Descriptor	A3 – Mesotidal semi-enclosed lagoon	
Tidal range	Mesotidal – 2 m during periods of free connection to the ocean	
Salinity	Mesohaline (5-6 to 18-20 ‰)	
	Strongly influenced by occasional freshwater inputs and by cycles of temporary communication with the ocean	
Shape	Semi-enclosed	
Depth	Mean depth < 2 m	

## **2. ENVIRONMENTAL CONDITIONS: HYDROLOGY, WATER, SEDIMENT AND TROPHIC STATE**

### **2.1. Specific Aims**

A general aim of this study was to characterize environmental conditions and to evaluate the water quality and trophic state of the two coastal lagoons. Specifically this aim included:

1. Description of seasonal changes in the hydrological regimen of the lagoons, associated to rainfall and connection with the sea.
2. Characterization of seasonal variation in water parameters, comparison along the gradient of distance to the sea and between lagoons.
3. Characterization of seasonal variation in sediment parameters, comparison along the gradient of distance to the sea and between lagoons.
4. Study the relations among environmental parameters in the two lagoons.
5. Comparison of environmental parameters, when the lagoons were isolated and in connection with the sea.
6. Evaluation of the trophic state and water quality along the gradient of distance to the sea and comparison between lagoons.

### **2.2. Material and Methods**

#### **2.2.1. Hydrological aspects**

Hydrological characterization consisted on basic description of water level variation in the lagoons. Water level data and information about Salgados lagoon connection to the sea was provided by the regional environmental services, CCDR-Algarve, former DRAOT. Water level measurements were daily collected in the deepest part of the lagoon.

No quantitative data was available for the water level in Foz de Almargem lagoon and an ordinal scale was adopted, based on reference points in the lagoon margins.

Information about the lagoon openings to the sea was given by local fishermen and by observation *in situ*.

Meteorological data were obtained from the INAG - Instituto Nacional da Água website (<http://snirh.pt>). The meteorological field station nearest to Foz de Almargem coastal lagoon is located in Loulé (31I/01UG) and the only data available for the studied period concerned daily rainfall, wind speed and wind direction. For Salgados coastal lagoon, daily rainfall and temperature data were gathered from the Algoz meteorological field station (31H/02C).

## **2.2.2. Water parameters**

### **2.2.2.1. Field procedures and laboratory analyses**

Field work was done from June 2001 to July 2002, with an interval of approximately 45 days.

In each lagoon, water sampling took place in three stations along a gradient of distance from the sea (E1- Upstream; E2- Intermediate; E3- Downstream).

Physical and chemical water parameters were analysed *in situ* and in laboratory. Temperature, salinity, dissolved oxygen concentration, oxygen saturation and pH were measured in the superficial water layer (50 cm depth) with a multi-parameter probe (YSI 556 MPS). Water samples with 1 L were collected at the same depth and preserved in cold, dark conditions for laboratory analysis of suspended matter (total solids in suspension) and dissolved nutrients (ammonia, nitrites, nitrates, orthophosphates and total phosphorus), as described in Greenberg *et al.* (1992). Dissolved inorganic nitrogen (DIN) was obtained as the sum of  $\text{N-NH}_4^+$ ,  $\text{N-NO}_2^-$  and  $\text{N-NO}_3^-$ .

In spring (May 2002), temperature, salinity, dissolved oxygen concentration, oxygen saturation and pH were monitored during a 24 hours cycle in two sampling stations (upstream and downstream in Foz de Almargem; intermediate and downstream in Salgados).

Besides physical and chemical water parameters, chlorophyll *a* was also quantified as an indicator of phytoplankton biomass that is required by many indexes of water quality and trophic state (Carlson, 1977; Vollenweider *et al.* 1998; Brito *et al.* 2012; Pereira

Coutinho *et al.* 2012). Water samples for chlorophyll *a* determinations were filtered through Whatman GF/C glass fibre filters and pigment extraction was performed with 90 % acetone. Pigment concentration was measured by spectrophotometry (Parsons *et al.*, 1984; Greenberg *et al.*, 1992) and calculations were done according to Lorenzen (1967). Pigment diversity was determined based on the absorbance at 430 and 665 nm (Margalef, 1960).

#### **2.2.2.2. Data analysis**

Data from the lagoons were first analysed separately, for sampling station comparison and then, mean values of each lagoon were compared.

Differences in water parameters from the sampling stations and differences between lagoons were tested through parametric tests (One-Way ANOVA: three stations; Student T test: two lagoons) or non-parametric tests (Kruskal-Wallis test: three stations; Mann-Whitney U test: two lagoons), depending on data normality distribution and homogeneity of variances, after logarithmic transformation ( $\ln x+1$ ). The LSD Fisher multiple comparison test was used to determine which of the three stations differed significantly (Maroco, 2010).

Principal component analyses (PCA) were performed on each sampling station data and on the mean values of the lagoons, to determine which variables were correlated and to summarize stations and lagoons characteristics in ordination diagrams. Water level in the lagoons and rainfall (cumulative values of rainfall from the 10 days prior to sampling) were also included, for seasonal comparison. Data were first centred and standardized, once variables had different units (Pielou, 1984).

#### **2.2.3. Sediment parameters**

##### **2.2.3.1. Field procedures and laboratory analyses**

Field work was done in the same periods as water monitoring.

In Foz de Almargem lagoon, sediment sampling was done in the three stations previously described, but in March 2002 no samples were collected as the water level was in its maximum and sampling stations were not reachable.

In Salgados lagoon, only two sampling stations were considered, upstream (E1) and downstream (E3). The intermediate station (E2) was difficult to access due to the channel depth and high density of emergent vegetation.

Sediment was characterized in terms of grain-size distribution, water and organic matter content and phytopigments concentration.

In each station and in every sampling occasion, a sediment sample was collected for grain-size analysis, with a 12 cm internal diameter corer at approximately 20 cm depth. Organic matter in samples was destroyed using hydrogen peroxide solution. Each sample was mixed and a sub-sample of about 150 g was taken and washed through a 63  $\mu\text{m}$  sieve, in order to separate the finer fractions (silt and clay) from the coarser fractions (gravel and sand). The fractions retained by the 63  $\mu\text{m}$  sieve were dried at 60°C for 72 hours and separated by a set of decreasing sieves into six different size classes (Berthois, 1956): gravel ( $\text{Ø} > 2 \text{ mm}$ ), very coarse sand ( $2 \text{ mm} > \text{Ø} > 1 \text{ mm}$ ), coarse sand ( $1 \text{ mm} > \text{Ø} > 500 \mu\text{m}$ ), medium sand ( $500 \mu\text{m} > \text{Ø} > 250 \mu\text{m}$ ), fine sand ( $250 \mu\text{m} > \text{Ø} > 125 \mu\text{m}$ ) and very fine sand ( $125 \mu\text{m} > \text{Ø} > 63 \mu\text{m}$ ). The silt ( $63 \mu\text{m} > \text{Ø} > 2 \mu\text{m}$ ) and clay ( $\text{Ø} < 2 \mu\text{m}$ ) fractions were separated and determined by sedimentation, using the pipette method described in Holme and McIntyre (1984). Samples were classified according to Flemming (2000), based on the clay, silt and sand ratios.

Samples for the determination of water content, organic matter content and phytopigments concentration were collected from the superficial sediment layer (1.5 cm depth), using a corer with 2 cm internal diameter. Three replicates were taken per site and sampling, for each analysis.

The water content in the sediment (% Water) was obtained by the difference between the wet weight (WW) and the dry weight (DW- 24 hours at 60°C in oven), as a percentage of the wet weight:  $\% \text{ Water} = (\text{WW}-\text{DW}) \cdot 100 / \text{WW}$ . The organic matter content (% OM) was determined by subtracting the ash weight (AW- 4 hours incineration at 500°C) from the dry weight, as a percentage of the dry weight:  $\% \text{ OM} = (\text{DW}-\text{AW}) \cdot 100 / \text{DW}$ .

Phytopigments (chlorophyll a and phaeo-pigments) were extracted from the sediment with acetone 90% for 24 hours. Concentrations were determined by spectrophotometry (Abs430 nm, Abs665 nm and Abs750 nm) according to the equations of Lorenzen (1967) modified by Plante-Cuny (1974). Pigments diversity (Abs430 nm/ Abs665 nm) (Margalef, 1960) and the percentage of chlorophyll a degradation [(Phaeo-pigments a) \*

100/ (Phaeo-pigments a + Chlorophyll a)] were calculated as indicators of microphytobenthos physiologic condition (Plante-Cuny, 1978).

### 2.2.3.2. Data analysis

Statistical analysis of sediment data was similar to the analysis performed on water parameters. In a first approach, sampling stations within each lagoon were compared and then, a comparison was made between the mean values of the lagoons.

Relations between environmental parameters (water and sediment), were evaluated by Pearson or Spearman correlation coefficients, according to data normality after transformation ( $\ln x+1$ ).

Principal Components Analyses (PCA) were also executed with the most relevant water and sediment parameters revealed by previous PCA, in order to be aware of relations between these parameters and to interpret the environmental characteristics of stations and lagoons.

The softwares used for data analysis were CANOCO (Ter Braak, version 4.54, 1988-2005 Biometris) and SPSS (version 19, IBM SPSS Statistics).

### 2.2.4. Trophic state and water quality

The evaluation of trophic state and water quality was based on different approaches: freshwater indexes (TSI of Carlson, 1977); the coastal water index TRIX (Vollenweider *et al.* 1998) and the 90<sup>th</sup> percentile of chlorophyll *a* in coastal lagoons (Brito *et al.* 2012; Pereira Coutinho *et al.* 2012).

TSI uses algal biomass as the basis for trophic state classification and three variables are used to independently estimate algal biomass, chlorophyll pigments, Secchi depth and total phosphorus. The range of the index is from approximately zero to 100, although theoretically it has no lower or upper bounds. As Secchi depth data were not collected in the lagoons, TSI was calculated just with chlorophyll ( $\mu\text{g L}^{-1}$ ) and total phosphorus ( $\mu\text{g L}^{-1}$  P) using the following equations (Carlson & Simpson, 1996):

$$\text{TSI (CHL)} = 9.81 \ln(\text{CHL}) + 30.6$$

$$\text{TSI (TP)} = 14.42 \ln(\text{TP}) + 4.15$$

The trophic state and water quality index TRIX (Vollenweider *et al.* 1998) is a combination of four state variables that directly express productivity, chlorophyll *a* ( $\text{mg m}^{-3}$ ) and oxygen as absolute deviation from saturation ( %), and nutritional factors available, dissolved inorganic nitrogen ( $\text{mg m}^{-3}$  N) and total phosphorus ( $\text{mg m}^{-3}$  P). It was calculated as follows:

$$\text{TRIX} = [\text{Log} (\text{Ch} \cdot \text{aD} \% \text{O} \cdot \text{N} \cdot \text{P}) - (-1.5)] / 1.2$$

Numerically, the index is scaled from zero to 10, corresponding to different categories of water quality, from high to poor water quality (Penna *et al.*, 2004).

Recently, Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012) proposed the use of the 90<sup>th</sup> percentile of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ) to assess the water quality in Portuguese coastal lagoons, as chlorophyll *a* is being applied in the implementation of the Water Framework Directive throughout Europe. Brito *et al.* (2012) established the classification of ecological quality based on reference values from two coastal lagoons in southern Portugal that are open throughout the year (Ria Formosa and Ria de Alvor), considering the 90<sup>th</sup> percentile of chlorophyll *a* during the growing season (February to October). Pereira Coutinho *et al.* (2012) defined an assessment of water quality for a different typology of coastal lagoons, semi-enclosed lagoons, which are not permanently open and are located in the western coast of Portugal (Óbidos, Albufeira and Santo André lagoons). As these lagoons can attain high phytoplankton abundances also in winter, the 90<sup>th</sup> percentile of chlorophyll *a* during the whole year was considered.

Although Foz de Almargem and Salgados lagoons are located in the same region as Ria Formosa and Ria de Alvor, they present characteristics that are more similar to the semi-enclosed lagoons of the western coast. Thereby, both assessments were used and compared in the studied lagoons.

## 2.3. Results and discussion

### 2.3.1. Foz de Almagem coastal lagoon

#### 2.3.1.1. Hydrological aspects

The raining season started in September 2001 and lasted until June 2002 (Figure 2.1). October, December, January and March were the months with a greater number of raining days (19 days in October and 23 days in March). During most of the studied period daily rainfall was under 60 mm and just in November and December values reached 72.5 mm and 111.5 mm, respectively.

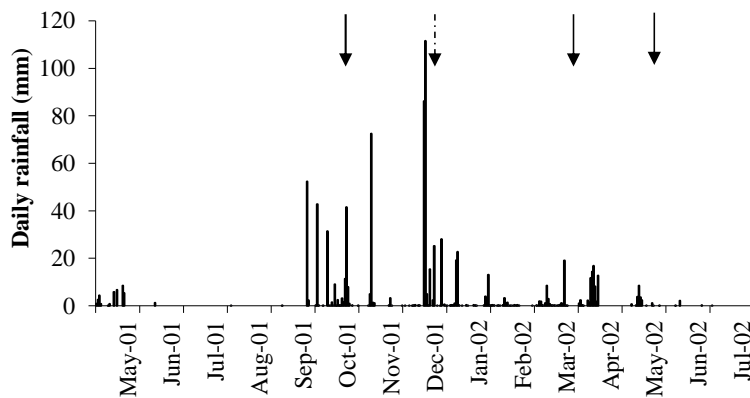


Figure 2.1 - Variation of daily rainfall registered in Loulé meteorological station and Foz de Almagem coastal lagoon openings to the sea. Full arrow indicates artificial opening and dashed arrow represent natural opening.

Water level in the lagoon varied greatly (Table 2.1) being mostly influenced by freshwater input (rainfall runoff), water evaporation and tides (when the lagoon was opened). During summer 2001, the lagoon was isolated from the sea and in August only the main channel had water. With the first autumn rains and the spread of seawater from the waves through the sand barrier, the water level increased slightly. From autumn until spring, the connection between the lagoon and the sea opened four times and seawater frequently entered the lagoon through wave spreading. In October 2001, March 2002 and May 2002, fisherman intentionally opened a channel from the lagoon to the sea. During December 2001, there was a natural opening of the lagoon due to rainfall and sea storms. When the lagoon was connected to the sea, water level was influenced by tides and only the deepest areas of the lagoon were submerged. In March

2002, the lagoon reached the maximum water level and even the adjacent areas were flooded. During the summer of 2002, water level was higher than in the summer of 2001.

Table 2.1- Water level in the lagoon and periods of isolation, semi-isolation and connection between Foz de Almargem coastal lagoon and the sea.

<i>Date</i>	<i>Sampling</i>	<i>Water level in the lagoon</i>	<i>Lagoon connected to the sea by channel</i>	<i>Lagoon semi-isolated by sand barrier</i>	<i>Lagoon isolated by sand barrier</i>
10.06.01	Jun-01	Middle			X
17.08.01	Aug-01	Low			X
2.10.01				X	
6.10.01	Oct-01	Middle			X
18.10.01		Tidal influence	X		
2.12.01	Dec-01	Middle-Low			X
15.12.01		Tidal influence	X		
16.01.02	Jan-02	Tidal influence	X		
23.01.02		High		X	
4.03.02		High		X	
12.03.02	Mar-02	High			X
25.03.02		Tidal influence	X		
9.05.02	May-02	High			X
14.05.02		Tidal influence	X		
15.07.02	Jul-02	High			X

### 2.3.1.2. Water parameters

Water temperature oscillated between 14.35 °C (December 2001) and 28.48 °C (August 2001), and was similar in the three sampling stations (Figure 2.2). Maximum salinity was reached in August 2001 (34.40 ‰) when water level in the lagoon was very low and the minimum was in January 2002 (3.54 ‰). The seasonal pattern of salinity resulted mainly from the hydrological balance between the freshwater inputs (*e.g.* rivers and rainfall), the evaporative water loss and the seawater inputs.

Salinity variation between sampling stations was not always the same. The higher values found in the intermediate and upstream stations during August and October 2001 were probably due to water depths of less than 1 m at these stations causing lack of circulation, and more intense evaporative effects. The downstream station was located nearer to the deepest part of the lagoon and had water depths fewer than 2 m. In January 2002, when the lagoon was in connection to the sea, a salinity gradient was observed according to the distance of sampling stations to the sea. March, May and July samplings were done when the lagoon was isolated from the sea and water level was high. Consequently, the upstream station had lower salinity and the two other stations showed small differences between them.

Dissolved oxygen concentration varied from 5.49 mg L<sup>-1</sup> (69.88 % oxygen saturation) in October 2001 to 12.65 mg L<sup>-1</sup> (121.8 % oxygen saturation) in December 2001. Until December 2001, the downstream station registered higher concentrations and saturation than the other stations but after that there was a decrease, probably due to seawater influence. Oxygen saturation (%) and dissolved oxygen (mg L<sup>-1</sup>) were significantly correlated ( $r = 0.739$ ;  $p = 0.01$ ).

Minimum pH 7.64 was registered in June 2001 and reached 9.49 in August 2001. Sampling stations had similar pH values, except in June and August 2001.

Total solids in suspension (TSS) increased during the summer months, when the lagoon water level was low and the effect of evaporation was greater. The maximum value occurred in October (127.78 mg L<sup>-1</sup>), after the first rains of the season. The TSS decrease registered in December 2001 might have been caused by the previous opening of the lagoon in the end of October 2001. During subsequent sampling periods the water level in the lagoon was high and TSS concentration was below 20 mg L<sup>-1</sup>.

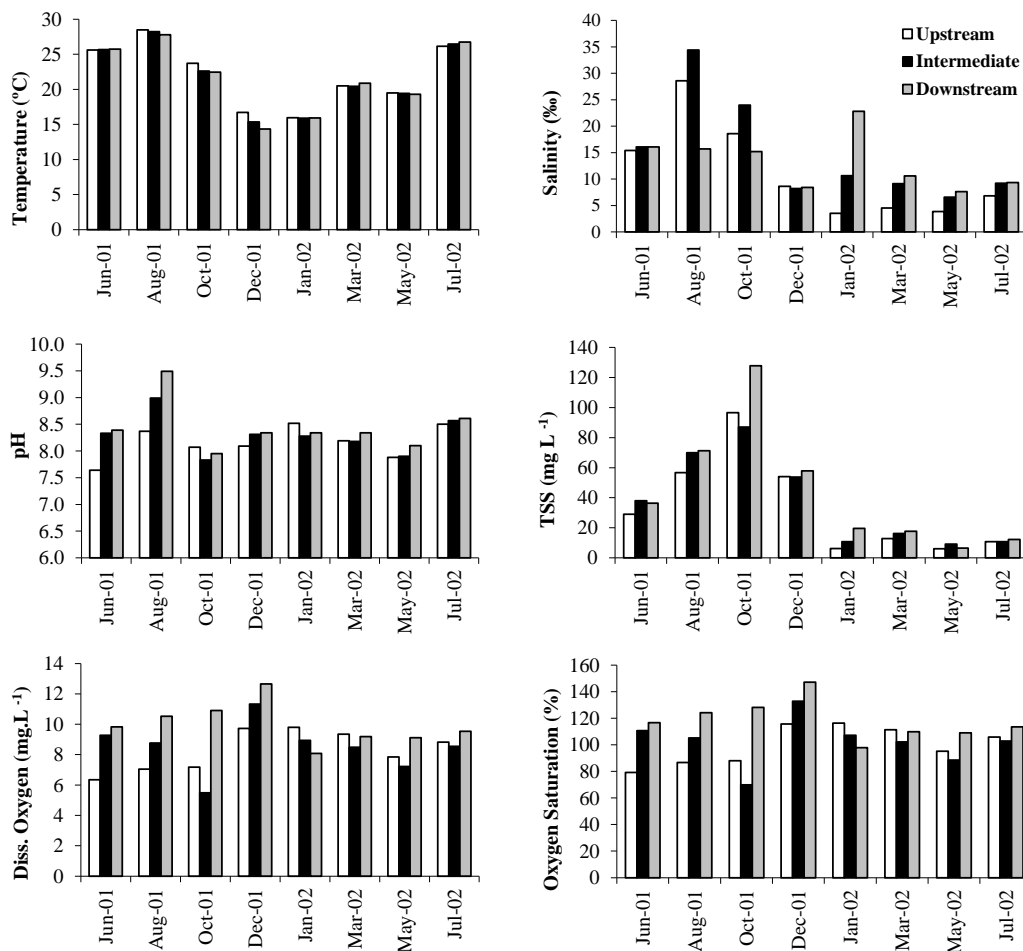


Figure 2.2 - Seasonal variation of physical and chemical water parameters (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Foz de Almagem sampling stations.

During the 24 hours cycle made in May 2002, all parameters monitored presented higher variations in the upstream sampling station (Figure 2.3). However, the minimum, the maximum and the mean values were superior in the downstream sampling station, except for water temperature (Table 2.2).

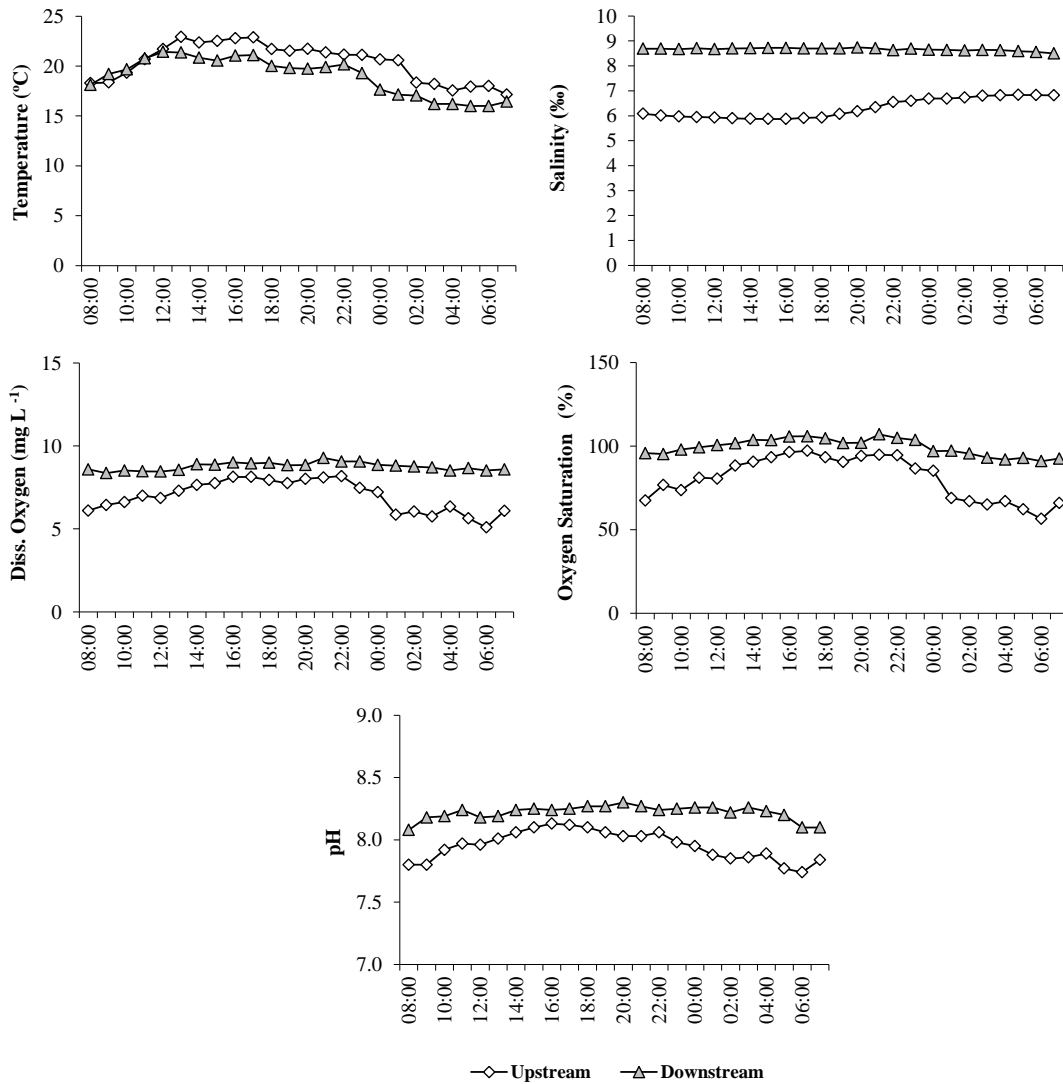


Figure 2.3 - Daily variation of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) during spring (May 2002) in the upstream and downstream sampling stations from Foz de Almagem coastal lagoon.

Water temperature variation was similar in the two sampling stations ( $5.77^{\circ}\text{C}$  and  $5.43^{\circ}\text{C}$ ), increasing during the day and decreasing during the night.

Salinity varied  $0.97\text{‰}$  in the upstream station and  $0.24\text{‰}$  in the downstream station.

Dissolved oxygen concentration oscillated  $3.09\text{ mg L}^{-1}$  ( $56.5\%$  oxygen saturation) in the upstream station and  $0.92\text{ mg L}^{-1}$  ( $16.1\%$  oxygen saturation) downstream.

The pH amplitude in the upstream station was 0.39 and 0.22 downstream.

During the day, photosynthesis consumes carbon dioxide and produces more oxygen than is consumed by bacteria and other organisms, causing an increase in pH, oxygen concentration and oxygen saturation. The decline in these parameters during the night is explained by the consumption of oxygen and the production of carbon dioxide by respiration and decomposition processes (Cravo, 2003). Greater fluctuations are usually associated to higher abundance of autotrophic organisms and bacteria (Conte de Barros, 1996).

Table 2.2 – Minimum, maximum and mean values of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) monitored during a 24 hours cycle in spring (May 2002) at the upstream and downstream sampling stations from Foz de Almargem lagoon.

	<i>Upstream Station</i>			<i>Downstream Station</i>		
	<b>Minimum</b>	<b>Maximum</b>	<b>Mean</b>	<b>Minimum</b>	<b>Maximum</b>	<b>Mean</b>
Temperature (°C)	17.15	22.92	20.37	16.00	21.43	18.99
Salinity (‰)	5.87	6.84	6.30	8.50	8.74	8.67
pH	7.74	8.13	7.95	8.08	8.30	8.22
Dissolved oxygen concentration (mg L <sup>-1</sup> )	5.09	8.18	6.98	8.36	9.28	8.76
Oxygen saturation (%)	56.50	97.20	80.73	91.00	107.10	99.40

Dissolved inorganic nitrogen (DIN) concentrations were low until December 2001 (1.89 – 13.43  $\mu\text{M N}$ ) and increased after a period of more intense rainfall (maximum: 157.74  $\mu\text{M N}$ ) (Figure 2.4). Nitrate was the most abundant nitrogen compound (0.32 – 156.45  $\mu\text{M N}$ ) and DIN seasonal pattern was similar to nitrate variation. Ammonia was higher in the summer (October 2001: 10.72  $\mu\text{M N}$ ; July 2002: 10.89  $\mu\text{M N}$ ) and decreased during winter (January 2002: 0.22  $\mu\text{M N}$ ). Nitrite contribution to DIN was negligible (0.09 – 2.78  $\mu\text{M N}$ ). The upstream station had the highest concentrations of DIN, nitrate and ammonia.

Orthophosphates concentration was greatest during the summer months (August 2001: 2.55  $\mu\text{M P}$ ) and lowest in winter (December 2001: 0.13  $\mu\text{M P}$ ). Differences between stations were more obvious in the summer. Total phosphorus concentration was similar to orthophosphates concentration, except in July 2002.

The N: P ratio varied between 1.24 and 129.28. From June 2001 to December 2001 (except in the upstream station), the N: P ratio was close or lower than the normal Redfield ratio (16:1), increasing in all stations during winter.

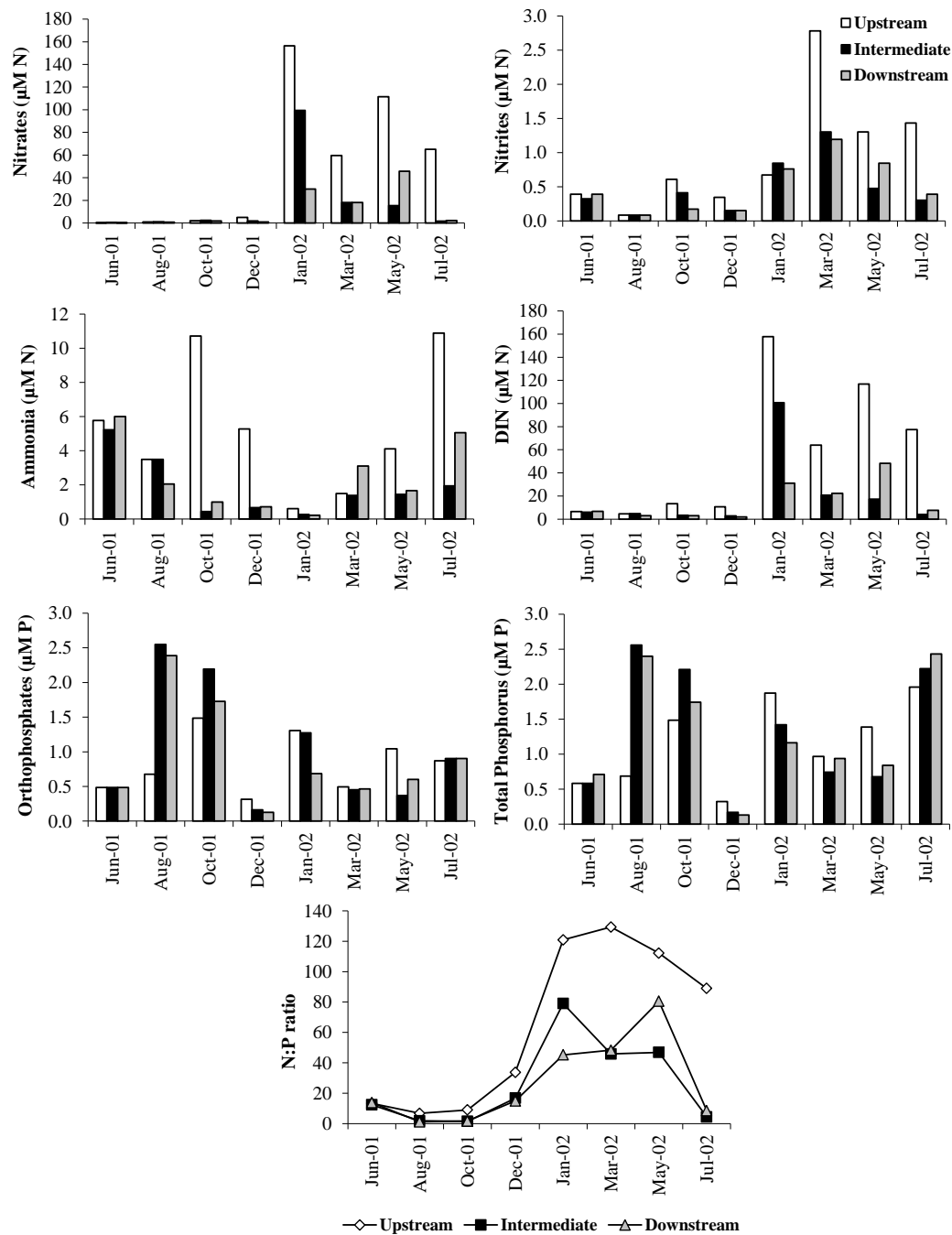


Figure 2.4 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration and N: P ratio (DIN: orthophosphate) in Foz de Almagrem sampling stations.

Chlorophyll *a* concentration in the lagoon was under  $10 \mu\text{g.L}^{-1}$  most of the year, except in March 2002 (Figure 2.5). The intermediate station presented the greatest seasonal variation ( $13.05 \mu\text{g L}^{-1}$ ) and maximum chlorophyll *a* concentration ( $13.48 \mu\text{g L}^{-1}$ ).

Phaeo-pigments concentration was lower than chlorophyll *a*, except in May 2002.

Pigment diversity index was similar in the three stations, presenting higher values in August 2001 (2.71) and March 2002 (2.74).

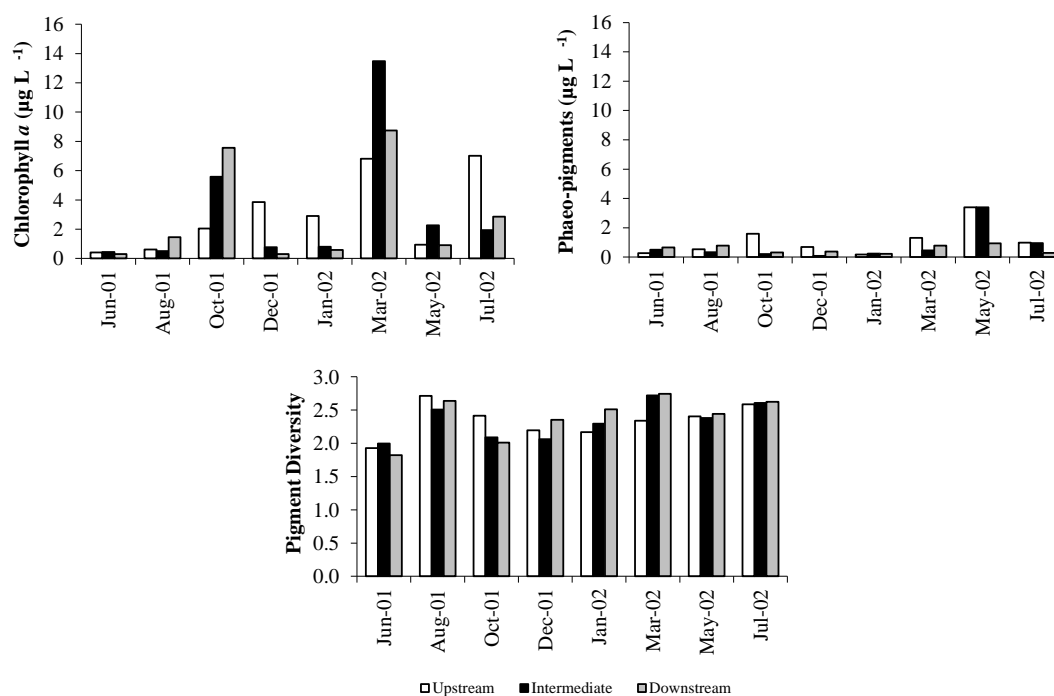


Figure 2.5 - Seasonal variation of photosynthetic pigments concentration (chlorophyll *a* and phaeo-pigments) and Margalef's pigment diversity index in Foz de Almargem sampling stations.

The upstream station presented higher means of temperature, nitrogen compounds (nitrates, nitrites, ammonia and total dissolved inorganic nitrogen), N:P ratio, phaeo-pigments concentration and lower means of salinity, pH, total solids in suspension, dissolved oxygen concentration (Table 2.3).

In the intermediate station, salinity, orthophosphates and total phosphorus concentrations registered the greatest mean values, while nitrites and ammonia concentrations, N:P ratio and pigment diversity showed the lowest means.

The highest means for pH, total solids in suspension, dissolved oxygen concentration and pigment diversity were determined in the downstream station. This station also had the lowest means for temperature, nitrates and total dissolved inorganic nitrogen concentrations, chlorophyll *a* and phaeo-pigments concentrations.

Despite the variation of water parameters means in the sampling stations, statistical analyses showed that there were no significant differences among stations ( $p > 0.05$ ), except for dissolved oxygen concentration and ammonia concentration ( $p \leq 0.05$ )

(Appendix I.A). Mean dissolved oxygen concentration was significantly higher downstream compared to the upstream station and mean ammonia concentration in the upstream station was significantly higher than in the intermediate station.

Table 2.3 – Annual mean values and standard deviation of water parameters in Foz de Almagem sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Temperature (°C)	22.07±4.60	21.75±4.81	21.64±4.97
Salinity (‰)	11.25±8.93	14.80±9.72	13.23±5.17
pH	8.16±0.31	8.30±0.37	8.44±0.47
Total Solids in Suspension	34.03±32.52	36.95±30.39	43.68±40.90
Dissolved oxygen concentration (mg L <sup>-1</sup> )	8.26±1.34	8.51±1.67	9.98±1.39
Nitrates concentration (µM N)	50.15±59.30	17.59±33.81	12.52±17.24
Nitrites concentration (µM N)	0.95±0.87	0.49±0.40	0.50±0.40
Ammonia concentration (µM N)	5.30±3.82	1.86±1.70	2.48±2.09
Total dissolved inorganic nitrogen concentration (µM N)	56.40±57.98	19.95±33.32	15.50±16.90
Orthophosphates concentration (µM P)	0.83±0.42	1.05±0.89	0.92±0.75
Total Phosphorus concentration (µM P)	1.16±0.61	1.32±0.91	1.29±0.83
N:P ratio	64.28±53.72	26.10±28.16	26.81±28.32
Chlorophyll a concentration (µg L <sup>-1</sup> )	3.07±2.64	3.22±4.48	2.84±3.40
Phaeo-pigments concentration (µg L <sup>-1</sup> )	1.11±1.04	0.77±1.10	0.55±0.28
Pigment diversity index (bits)	2.34±0.25	2.33±0.27	2.39±0.32

PCA ordination of environmental variables (hydrological and water parameters) and sampling stations is presented in figure 2.6. Samples displayed on the left side of axis I (*e.g.* upstream May and March 2002) were characterised by higher water level (component loading = -0.739), total dissolved inorganic nitrogen (component loading = -0.917), nitrates (component loading = -0.913), N: P ratio (component loading = -0.915) and lower values of total solids in suspension (component loading = 0.812), salinity (component loading = 0.757) and temperature (component loading = 0.562). On the right side of the axis I were located the samples with the opposite variation of the same parameters (*e.g.* upstream August 2001, downstream October 2001). The first axis accounted for 30.4 % of the total variance.

The second axis explained 16.7 % of the total variance and was related to the total phosphorus (component loading = 0.874), orthophosphates (component loading = 0.695), pH (component loading = 0.739) and pigments diversity (component loading = 0.497), projecting in the upper side of the axis the samples with higher values (*e.g.* intermediate and downstream August 2001) and in the bottom, the ones with lower

values (e.g. June and December 2001). Cumulative rainfall, dissolved oxygen and ammonia concentrations did not contribute much for the samples ordination in the first two axes.

Some environmental variables were close to each other on the ordination diagram, due to high positive correlation coefficients between them. This was the case of nitrate, total dissolved inorganic nitrogen, N: P ratio, nitrites and chlorophyll *a* concentrations; water level and phaeo-pigments concentration; temperature and salinity; TSS and ammonia concentration; total phosphorus, pH, pigment diversity and orthophosphates. Variables as total solids in suspension and total dissolved inorganic nitrogen or salinity and water level were negatively correlated, as arrows pointed at opposite directions.

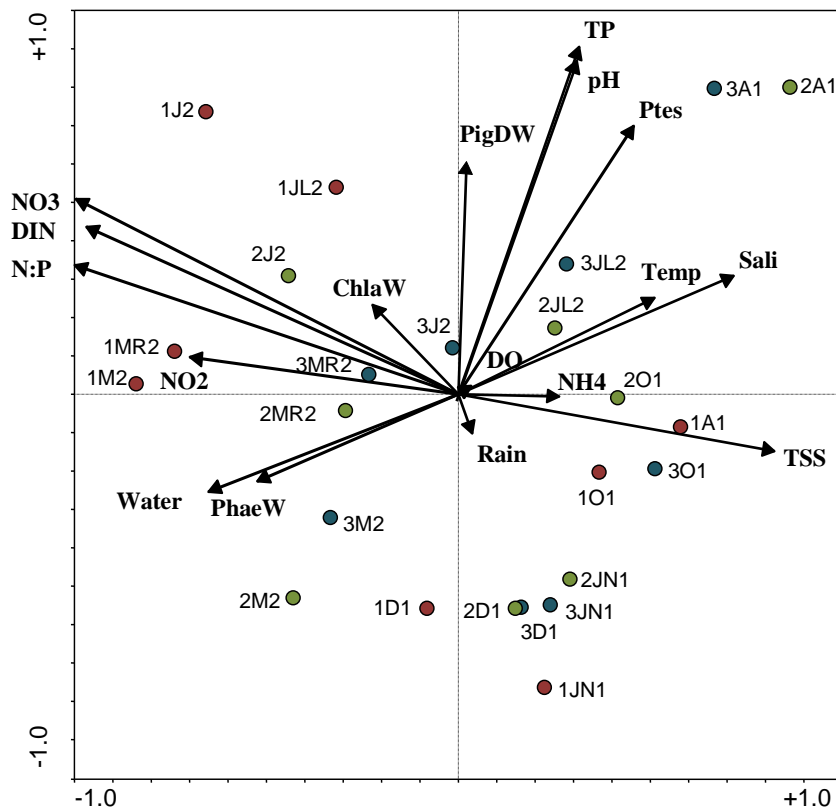


Figure 2.6 - Principal Component Analysis performed on the hydrological and water parameters from Foz de Almagem sampling stations. Cumulative percentage variance explained by axes: I – 30.4 %; I + II – 47.1 %.

*Station codes:* First character corresponds to the sampling station (1- upstream, 2- intermediate, 3- downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001, 2-2002).

*Environmental variables:* Rain- cumulative rainfall in 10 days previous to sampling; Water- water level in the lagoon; Temp- water temperature; Sali- salinity; pH; DO- dissolved oxygen concentration; TSS- total solids in suspension; NO<sub>3</sub>- nitrates concentration; NO<sub>2</sub>- nitrites concentration; NH<sub>4</sub>- ammonia concentration; DIN- total dissolved inorganic nitrogen concentration; Ptes- orthophosphates concentration; TP- total phosphorus concentration; N: P- DIN and TP ratio; ChlaW- Chlorophyll *a* concentration; PhaeW- Phaeo-pigments concentration; PigDW- pigments diversity index.

### 2.3.1.3. Sediment parameters

According to Flemming (2000), sediment samples from the upstream station were classified as slightly silty sand (A-I), except the ones collected in December 01 and July 02, which were included in the sand category. The intermediate station presented a greater variation of sand, silt and clay ratios along time and samples classification went from sand (October 01), to very silty sand (B-I: June 01 and July 02), very silty sandy mud (C-II: August and December 01) and extremely silty sandy mud (C-I: January and May 01). All downstream samples were classified as sand (Figure 2.7).

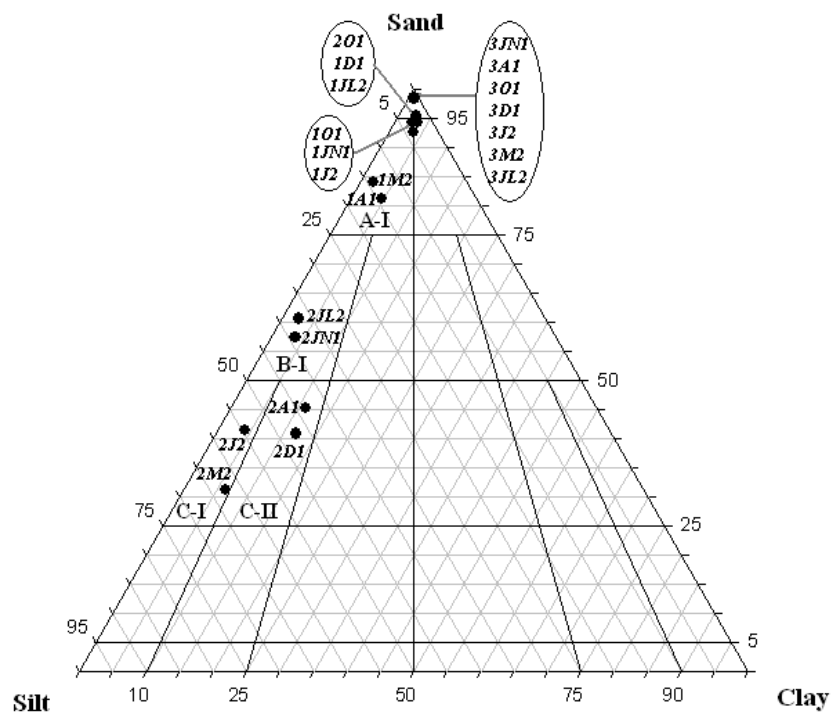


Figure 2.7 - Ternary diagram for textural classification of sediments from Foz de Almargem coastal lagoon, based on sand, silt and clay ratios (adapted from Flemming, 2000).

*Station codes:* First character corresponds to the sampling station (1- upstream; 2 – intermediate; 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; M- May; JL- July) and year of survey (1- 2001; 2- 2002).  
*Classification codes:* A-1 - Slightly silty sand; B-I - Very silty sand; C-I - Extremely silty sandy mud; C-II - Very silty sandy mud.

Although the upstream samples presented a high percentage of sand, grain size fractions were quite distinct from the ones found in the downstream station (Figure 2.8). Besides the higher amount of mud (silt+clay) (4.5 % - 18.8 %), the upstream station sediments were more heterogeneous than the sediments from the downstream station, which were better calibrated. The most common sand fractions upstream were

medium sand (17.4 % - 47.1 %), coarse sand (29.4 % - 43.7 %) and very coarse sand (5.8 % - 17.6 %), while downstream, medium sand (41.3 % - 71.1 %) and coarse sand (17.6 % - 46.1 %) were the dominant grain size fractions. The intermediate station was mainly dominated by silt (37.0 % - 62.6 %) and other grain size fractions inferior to 0,500 mm (30.8 % - 50.5 %). Coarse sand (2.9 % - 13.1 %) and very coarse sand (2.0 % - 3.3 %) represented a small proportion of sediments.

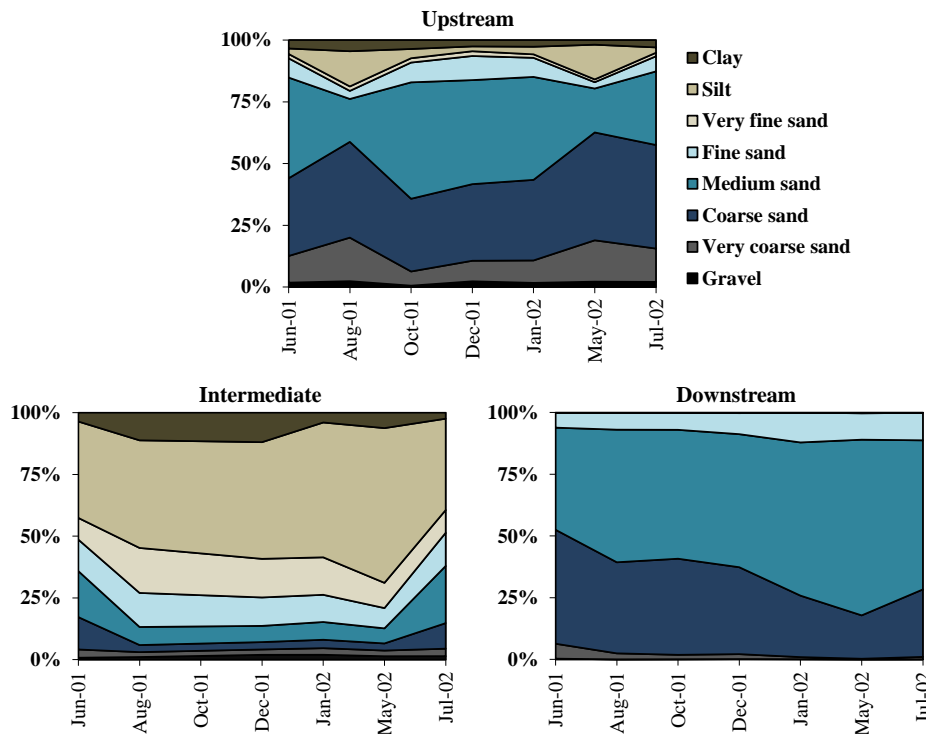


Figure 2.8 - Seasonal variation of individual grain size fractions in the tree sampling stations from Foz de Almargem coastal lagoon.

Differences and similarities in grain size fractions among stations reflected the major hydrological forces in each station. The dominance of sand (99.992 %  $\pm$  0.009) in the downstream station was due to wave spread and wind action, as this station was located near the sand barrier. The lower percentages of sand (53.273 %  $\pm$  21.505) and greater amounts of silt (40.839 %  $\pm$  19.359) and clay (5.888 %  $\pm$  4.134) from the intermediate station sediments were related to its location in the centre of the lagoon, where fine sediments deposition was favoured by lower river flow and wave action. The sediment from the upstream station presented a higher percentage of sand (90.985 %  $\pm$  5.814) and a lower amount of silt (5.927 %  $\pm$  5.654) and clay (3.088 %  $\pm$  0.849) than the intermediate station and its origin was mainly the river flow, just as the erosion and runoff from the surrounding sandy lands. The upstream station was located in a

confined area of the lagoon, near a sloped margin not far from the river entrance and therefore, its sediments were not directly exposed to marine influence.

Water content in the sediment varied from 20.58 % (Downstream: Jan-02) to 50.86 % (Intermediate: May-02) and through time, the intermediate station presented the highest values and the downstream station the lowest values (Figure 2.9).

To what concerns the organic matter content in the sediment, the minimum value (0.25 %) was determined for the downstream station during December 2001 and the maximum (9.13 %) occurred in the intermediate station during January 2002. The higher values in each sampling period were found in the intermediate station (except in December 2001) and the lower values in the downstream station.

Chlorophyll *a* concentration in the sediment ranged from 0.42  $\mu\text{g g}^{-1}$  in January 2002 (Downstream) to 26.25  $\mu\text{g g}^{-1}$  in May 2002 (Upstream). The upstream station registered the higher values and the downstream station the lower ones.

Phaeo-pigments concentration oscillated from 0.19  $\mu\text{g g}^{-1}$  in August 2001 (Downstream) to 30.50  $\mu\text{g g}^{-1}$  in May 2002 (Upstream) and through time, variation in the stations followed a similar trend as chlorophyll *a* concentration.

Chlorophyll *a* degradation, accounted as phaeo-pigments percentage, had its minimum (9.93 %) downstream in July 2002 and its maximum (67.67 %) in the intermediate station, during August 2001. The lower values were observed in the downstream station, except for the months of December 2001 and January 2002, when the tendency inverted and the station presented the highest values of all three stations. From June to October 2001 and in May 2002, the higher values were found in the intermediate station.

Pigment diversity range went from 2.23 (Downstream) to 4.28 (Intermediate) in May 2002. The intermediate station revealed higher values in October 2001, May and July 2002, while the upstream station showed the highest values during the remaining samplings.

Seasonal variations within each sampling station resulted from the interaction of the river flow, erosion and runoff from the margins (influenced by rainfall) and the entrance of seawater through tides (when the lagoon was connected to the sea) and wave spreading. Comparison of sediment parameters between periods of isolation and periods of connection to the sea was not performed, as most of the sampling was done when the lagoon was isolated (except January 2002).

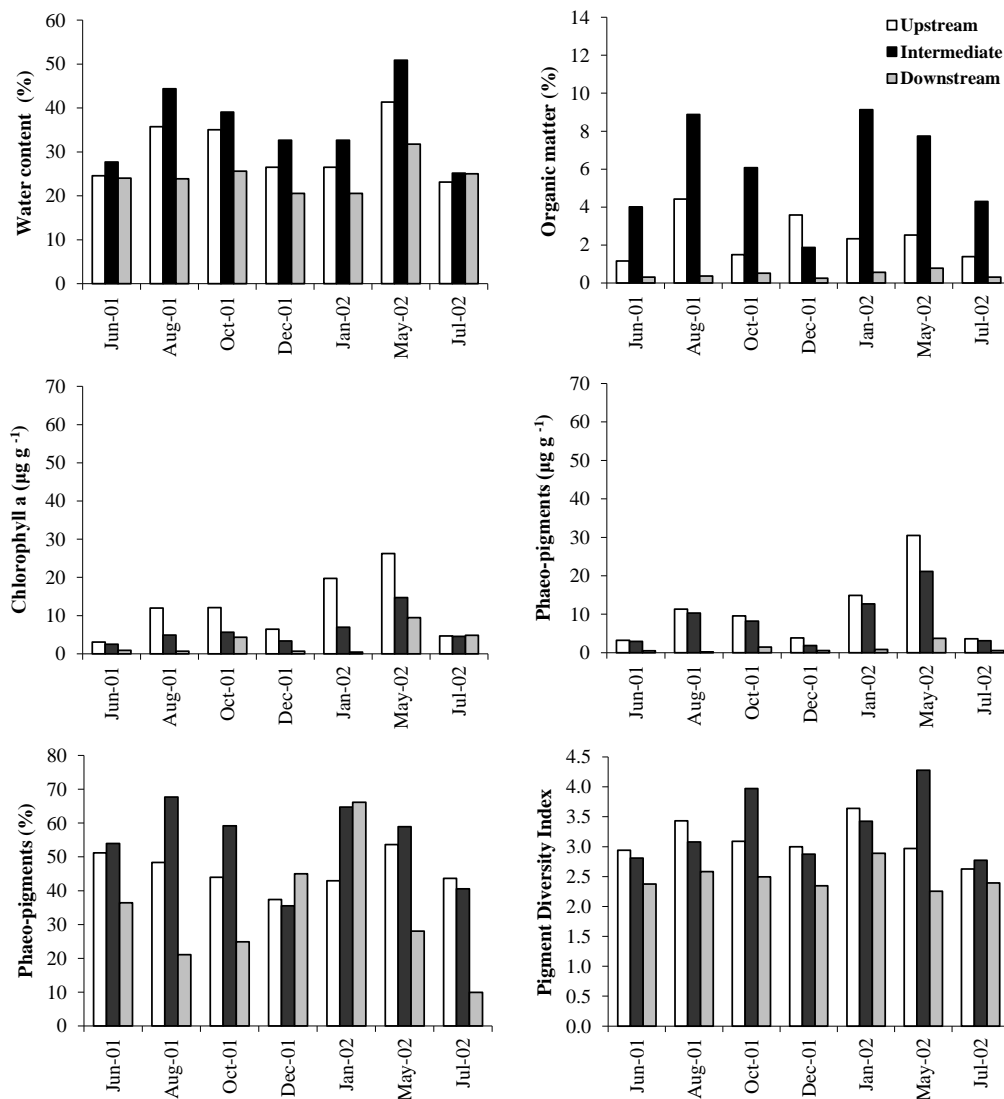


Figure 2.9 - Seasonal variation of water content, organic matter content, photosynthetic pigments concentration (chlorophyll *a* and phaeo-pigments), chlorophyll *a* degradation index (% phaeo-pigments) and Margalef's pigment diversity index in the sediment of the three sampling stations from Foz de Almargem coastal lagoon.

The highest annual mean concentrations of chlorophyll *a* and phaeo-pigments were found in the upstream station (Table 2.4). The intermediate station showed the lowest sand content and the greatest means of clay content, silt content, water content, organic matter content, chlorophyll *a* degradation index and pigments diversity. In the downstream station was determined the highest mean of sand content, but all other sediment parameters presented the lowest mean values.

Table 2.4 – Annual mean values and standard deviation of sediment parameters in Foz de Almagem sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Clay content (%)	3.09±0.85	7.28±4.17	0.00±0.00
Silt content (%)	5.93±5.65	47.06±8.95	0.00±0.00
Sand content (%)	90.99±5.81	45.66±10.17	99.99±0.01
Water content (%)	30.42±6.91	36.08±9.21	24.51±3.79
Organic matter content (%)	2.42±1.22	6.00±2.74	0.45±0.19
Chlorophyll <i>a</i> concentration ( $\mu\text{g g}^{-1}$ )	12.03±8.44	6.09±4.07	3.04±3.38
Phaeo-pigments concentration ( $\mu\text{g g}^{-1}$ )	10.98±9.70	8.60±6.90	1.10±1.22
Chlorophyll <i>a</i> degradation index (%)	45.87±5.55	54.36±12.07	33.07±18.34
Pigment diversity index (bits)	3.10±0.34	3.32±0.60	2.48±0.21

The statistical analyses used to compare sediment parameters among stations (Appendix I.B) showed that the downstream station was significantly higher in sand content than the intermediate station and lower in clay content, silt content, water content, organic matter content, phaeo-pigment percentage and pigment diversity. From the intermediate to the upstream station there was a significant decrease in silt content and organic matter content, just as an increase in sand content. When compared with the upstream station, the downstream station presented higher sand content and lower chlorophyll *a* concentration, phaeo-pigments concentration and pigment diversity, besides clay content, silt content and organic matter content.

The PCA ordination biplot (Figure 2.10) showed a separation of downstream samples and a few upstream samples (June 2001, December 2001, July 2002) (left side) from the intermediate samples and remaining upstream samples (right side), according to axis I. The first axis accounted for 54.4% of the total variance and the sediment parameters which contributed most for samples distribution along the axis were water content (component loading = 0.878), organic matter content (component loading = 0.858), pigment diversity index (component loading = 0.812), phaeo-pigment concentration (component loading = 0.791) and silt content (component loading = 0.740). The samples plotted in the left side of axis I had lower values of water content, organic matter content, pigment diversity index, phaeo-pigments concentration and silt content while the samples in the right side presented higher values for these same parameters. The second axis explained 17.3 % of the total variance and chlorophyll *a* concentration was the parameter with greater contribution to this axis (component loading = 0.778).

Samples displayed in the upper side of the ordination diagram showed higher values of chlorophyll *a* concentration (*e.g.* upstream May and January 2002) and those samples in the bottom had lower values (*e.g.* intermediate June and December 2001). Some of the parameters were highly and positively correlated and were represented close to each other, namely water content and pigment diversity index, organic matter content and chlorophyll *a* degradation index, silt content and clay content. Those parameters pointing in opposite directions were negatively correlated, as the case of Sand: Mud ratio and water content or Sand: Mud ratio and pigments diversity index.

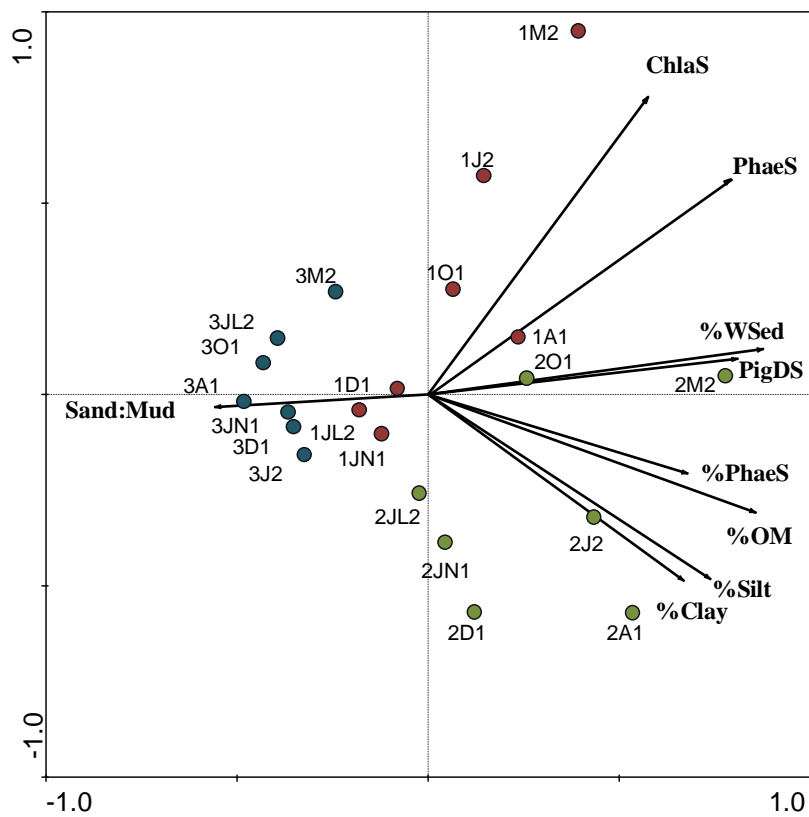


Figure 2.10 - Principal component analysis performed on sediment parameters from Foz de Almagem coastal lagoon. Cumulative percentage variance explained by axes: I – 54.4%; I + II – 71.7%.

*Station codes:* First character corresponds to the sampling station (1 – upstream, 2 - intermediate, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002). *Sediment parameters:* Sand:Mud – Sand mud ratio; %Clay – Clay content; %Silt – Silt content; %OM – Organic matter content; %WSed – Water content; ChlaS - Chlorophyll *a* concentration; PhaeS - Phaeo-pigments concentration; PigDS - Margalef's pigment diversity index; %PhaeS - Chlorophyll *a* degradation index.

### 2.3.1.4. Relations between environmental parameters

Some of the water and sediment parameters presented significant ( $0.01 < p \leq 0.05$ ) and highly significant ( $p \leq 0.01$ ) correlations (Table 2.5).

Water content in the sediment was negatively correlated to dissolved oxygen in water and positively correlated to phaeo-pigments concentration in water, meaning that the samples with higher water content in the sediment (intermediate and upstream) tended to have lower values of dissolved oxygen in water and higher values of phaeo-pigments concentration in water and *vice-versa*. Chlorophyll *a* and phaeo-pigments concentrations in the sediment showed a positive linear association with nitrates, dissolved inorganic nitrogen and phaeo-pigments concentrations in water. Samples with higher concentrations of chlorophyll *a* and phaeo-pigments in the sediment (upstream and intermediate) were the ones with higher values of nitrates, dissolved inorganic nitrogen and phaeo-pigments concentrations in water and *vice-versa*. A negative correlation was also found between phaeo-pigments in the sediment and dissolved oxygen in water. Organic matter content, chlorophyll *a* degradation index and pigment diversity index in the sediment were negatively associated to dissolved oxygen in water, which indicated that the samples with higher dissolved oxygen in water (downstream) presented lower values of organic matter content, chlorophyll *a* degradation and pigment diversity in the sediment and *vice-versa*.

Table 2.5 - Significant correlations between water and sediment parameters from Foz de Almagem coastal lagoon. \* Correlation significant at the 0.05 level; \*\* Correlation significant at the 0.01 level.

<i>Sediment parameters</i>	<i>Water parameters</i>	<i>Results</i>
<b>Water content (%WSed)</b>	Dissolved Oxygen (DO)	R = -0.490; $p = 0.024$ *
	Phaeo-pigments concentration (PhaeW)	R = 0.473; $p = 0.030$ *
<b>Organic matter content (%OM)</b>	Dissolved Oxygen (DO)	R = -0.453; $p = 0.039$ *
<b>Chlorophyll <i>a</i> concentration (ChlaS)</b>	Nitrates concentration (NO <sub>3</sub> )	R = 0.510; $p = 0.018$ *
	Dissolved inorganic nitrogen (DIN)	R = 0.513; $p = 0.013$ *
	Phaeo-pigments concentration (PhaeW)	R = 0.484; $p = 0.026$ *
<b>Phaeo-pigments concentration (PhaeS)</b>	Dissolved Oxygen (DO)	R = -0.544; $p = 0.011$ *
	Nitrates concentration (NO <sub>3</sub> )	R = 0.503; $p = 0.020$ *
	Dissolved inorganic nitrogen (DIN)	R = 0.525; $p = 0.014$ *
	Phaeo-pigments concentration (PhaeW)	R = 0.471; $p = 0.031$ *
<b>Chlorophyll <i>a</i> degradation index (%PhaeS)</b>	Dissolved Oxygen (DO)	R = -0.498; $p = 0.022$ *
<b>Pigment diversity index (PigDS)</b>	Dissolved Oxygen (DO)	R = -0.615; $p = 0.003$ **

Figure 2.11 presents the ordination of samples in a PCA biplot, according to the most relevant water and sediment parameters determined in previous PCA, performed separately for water parameters and for sediment parameters.

Downstream samples (except May 2002) and a few upstream and intermediate samples were positioned in the left side of axis I, characterized by lower values of phaeo-pigments (component loading = 0.909) and chlorophyll *a* in the sediment (component loading = 0.846) and total inorganic dissolved nitrogen in the water (component loading = 0.680). The remaining samples, in the right side of the first axis, presented higher values of phaeo-pigments and chlorophyll *a* in the sediment and total inorganic dissolved nitrogen in the water (e.g. upstream and intermediate May 2002). The first axis accounted for 36.7% of the total variance.

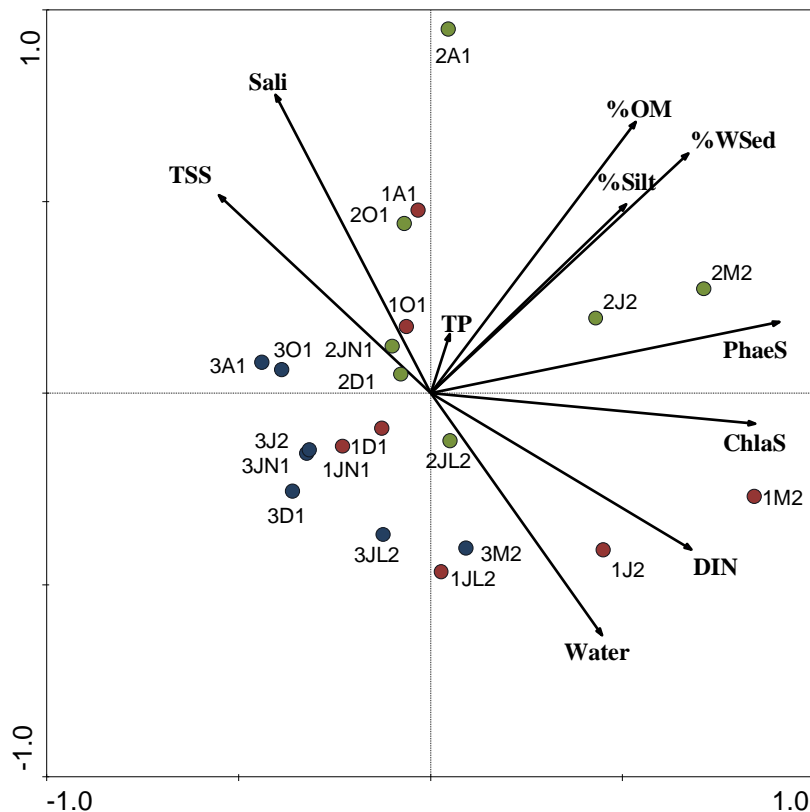


Figure 2.11 - Principal component analysis performed on water and sediment parameters from Foz de Almagem coastal lagoon. Cumulative percentage variance explained by axes: I – 36.7%; I + II – 63.0%.

*Station codes:* First character corresponds to the sampling station (1 – upstream, 2 - intermediate, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002). *Water parameters:* Water - Water level in the lagoon; Sali – Salinity; TSS – Total solids in suspension; TP- Total phosphorus concentration; DIN –Total dissolved inorganic nitrogen. *Sediment parameters:* %Silt – Percentage of silt; %OM – Percentage of organic matter; %WSed – Water content; ChlaS - Chlorophyll *a* concentration; PhaeS - Phaeo-pigments concentration.

The second axis explained 26.3 % of the total variance and the parameters with greater contribution to this axis were salinity (component loading = 0.778), organic matter content in the sediment (component loading = 0.778) and water level (component loading = -0.631). Samples displayed in the upper side of the ordination diagram showed higher values of salinity, organic matter content in the sediment and lower water level (*e.g.* upstream and intermediate August 2001). Samples in the bottom part of the diagram had lower values of salinity, organic matter content in the sediment and higher water level (*e.g.* upstream July 2002 and downstream May 2002). Water content in the sediment and total solids in suspension contributed to both axes (Water content: first component loading = 0.671, second component loading = 0.626; TSS: first component loading = -0.551, second component loading = 0.516). Silt content in sediment and total phosphorus in water had a smaller contribution to samples ordination.

In order to evaluate the importance of microphytobenthos biomass and phytoplankton biomass in the water column, concentrations of chlorophyll *a* in the water and chlorophyll *a* in the sediment were converted to  $\text{mg m}^{-2}$  (Brito *et al.* 2010).

All stations presented a greater amount of chlorophyll *a* in the sediment than in the water column and the highest values were observed in May 2002 (Figure 2.12).

Chlorophyll *a* in the upstream station sediment ranged from  $8.30 \text{ mg m}^{-2}$  (95.3%) in June 2001 to  $70.52 \text{ mg m}^{-2}$  (98.7%) in May 2002, accounting a minimum of 64.0% (July 2002) and a maximum of 98.7% of total chlorophyll *a*. The annual mean value of  $32.33 \text{ mg m}^{-2}$  represented 89.5% of total chlorophyll *a* upstream.

In the intermediate and downstream stations, the values were lower than in the upstream station and the annual means were  $14.77 \text{ mg m}^{-2}$  (89.4%) and  $8.98 \text{ mg m}^{-2}$  (78.0%) respectively. The intermediate station presented values from  $6.11 \text{ mg m}^{-2}$  (93.34%) in June 2001 to  $35.72 \text{ mg m}^{-2}$  (94.0%) in May 2002, while the downstream station varied between  $1.23 \text{ mg m}^{-2}$  (95.4%) in January 2002 and  $27.92 \text{ mg m}^{-2}$  (94.0%) in May 2002. The variation of chlorophyll *a* in the sediment accounted 71.1% to 95.9 % of total chlorophyll *a* in the intermediate station and 58.6% to 96.9 % of total chlorophyll *a* downstream.

Chlorophyll *a* in the water column oscillated between 1.3% ( $0.94 \text{ mg m}^{-2}$ ) and 36.0% ( $7.01 \text{ mg m}^{-2}$ ) of total chlorophyll *a* in the upstream station; 4.1% ( $0.51 \text{ mg m}^{-2}$ ) and 28.9% ( $5.58 \text{ mg m}^{-2}$ ) of total chlorophyll *a* in the intermediate station; 3.1% ( $0.90 \text{ mg}$

m<sup>-2</sup>) and 41.4% (1.45 mg m<sup>-2</sup>) of total chlorophyll *a* in the downstream station. The annual mean values in each station (without March 2002 data) were 2.54 mg m<sup>-2</sup> (upstream: 10.5%), 1.76 mg m<sup>-2</sup> (intermediate: 10.6%) and 1.99 mg m<sup>-2</sup> (downstream: 22.0%).

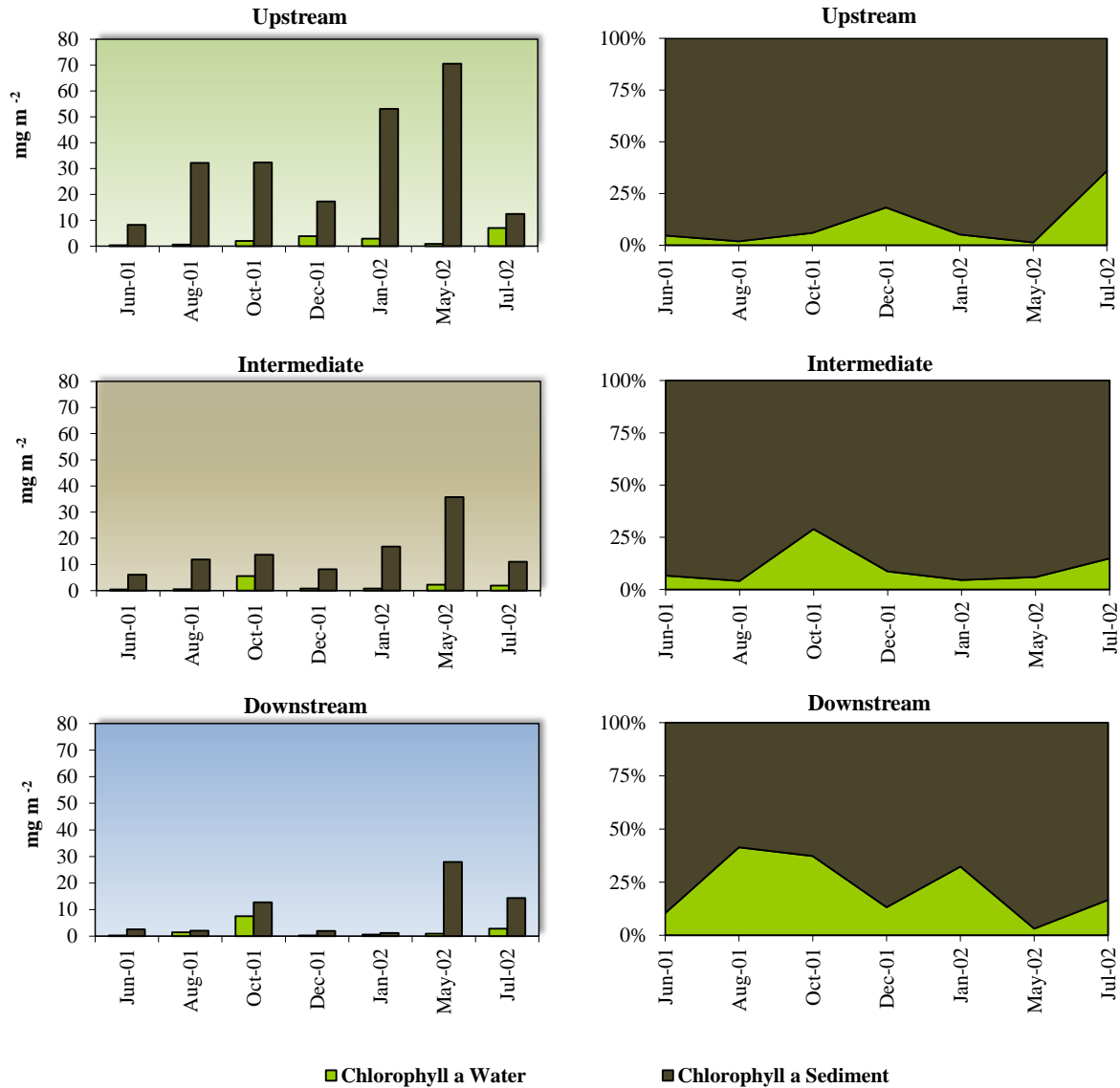


Figure 2.12 - Seasonal variation of chlorophyll *a* in the sediment, chlorophyll *a* in the water and percentage of each relative to total chlorophyll *a* in Foz de Almargem sampling stations.

### 2.3.1.5. Comparison of environmental parameters during isolation and connection of the lagoon with the sea

During the studied period, only in January 2002 sampling took place when the lagoon was opened and with direct influence from the sea. Data from January 2002 were

therefore compared with data from the previous sampling done in December 2001, when the lagoon was isolated from the sea. Before December, there was a first opening of the lagoon in the end of October and between December and January samplings, the lagoon opened for a second time. Table 2.6 resumes the information from the comparison of December and January environmental parameters at each station.

All stations presented a decrease in total solids in suspension and ammonia concentrations and registered an increase in nitrates, nitrites, total dissolved inorganic nitrogen, orthophosphates, total phosphorus and the N:P ratio. Temperature, salinity and chlorophyll *a* decreased in the upstream station and increased in the other stations. Dissolved oxygen concentration and pH showed an increase in the upstream station. In the intermediate and downstream stations was noticed a decrease in dissolved oxygen concentration. Phaeo-pigments concentration increased in the intermediate station and decreased in the upstream and downstream stations.

The water parameters with major variations were nitrates, total dissolved inorganic nitrogen, N:P ratio, total solids in suspension, ammonia and total phosphorus concentrations.

The upstream station presented the minor variations for dissolved oxygen ( $0.07 \text{ mg L}^{-1}$ ) and nitrites concentrations ( $0.33 \text{ }\mu\text{M N}$ ), and also the greatest variations of nitrates ( $151.42 \text{ }\mu\text{M N}$ ), total dissolved inorganic nitrogen ( $147.08 \text{ }\mu\text{M N}$ ), N:P ratio (87.10), total solids in suspension ( $-47.79 \text{ mg L}^{-1}$ ), ammonia ( $-4.67 \text{ }\mu\text{M N}$ ), total phosphorus ( $1.15 \text{ }\mu\text{M P}$ ), chlorophyll *a* ( $-0.96 \text{ }\mu\text{g L}^{-1}$ ), phaeo-pigments ( $-0.51 \text{ }\mu\text{g L}^{-1}$ ) and pH (0.43). The smallest variations of temperature ( $0.58^\circ\text{C}$ ), salinity (2.40 ‰), ammonia ( $-0.39 \text{ }\mu\text{M N}$ ) and chlorophyll *a* ( $0.03 \text{ }\mu\text{g L}^{-1}$ ) occurred at the intermediate station, just as the major variations in nitrites ( $0.70 \text{ }\mu\text{M N}$ ), orthophosphates ( $1.11 \text{ }\mu\text{M P}$ ) and pigment diversity (0.23).

Downstream was the station with the greatest variation in temperature ( $1.58^\circ\text{C}$ ), salinity (14.41 ‰) and dissolved oxygen concentration ( $-4.57 \text{ mg L}^{-1}$ ). It also registered the minimum variation for nitrates ( $28.98 \text{ }\mu\text{M N}$ ), total dissolved inorganic nitrogen ( $29.09 \text{ }\mu\text{M N}$ ), N:P ratio (30.32), total solids in suspension ( $-38.18 \text{ mg L}^{-1}$ ), orthophosphates ( $0.56 \text{ }\mu\text{M P}$ ) and total phosphorus concentrations ( $1.03 \text{ }\mu\text{M P}$ ).

Table 2.6 – Water and sediment parameters in January 2002 and variation between values when the lagoon was closed (December 2001) and opened to the sea (January 2002) in Foz de Almagem sampling stations.

	<i>Upstream station</i>		<i>Intermediate station</i>		<i>Downstream station</i>	
	<b>Jan-02</b>	<b>Variation</b>	<b>Jan-02</b>	<b>Variation</b>	<b>Jan-02</b>	<b>Variation</b>
<b>Water parameters</b>						
Temperature (°C)	15.94	<b>-0.76</b>	15.89	<b>0.54</b>	15.93	<b>1.58</b>
Salinity (‰)	3.54	<b>-5.09</b>	10.64	<b>2.40</b>	22.60	<b>14.41</b>
pH	8.52	<b>0.43</b>	8.28	<b>-0.03</b>	8.34	<b>0.00</b>
Total Solids in Suspension (mg L <sup>-1</sup> )	6.21	<b>-47.79</b>	10.75	<b>-43.00</b>	19.65	<b>-38.18</b>
Dissolved oxygen (mg L <sup>-1</sup> )	9.80	<b>0.07</b>	8.94	<b>-2.39</b>	8.08	<b>-4.57</b>
Nitrates concentration (µM N)	156.45	<b>151.42</b>	99.52	<b>97.63</b>	30.00	<b>28.98</b>
Nitrites concentration (µM N)	0.67	<b>0.33</b>	0.85	<b>0.70</b>	0.76	<b>0.61</b>
Ammonia concentration (µM N)	0.61	<b>-4.67</b>	0.28	<b>-0.39</b>	0.22	<b>-0.50</b>
Total diss. Inorg. nitrogen (µM N)	157.74	<b>147.08</b>	100.64	<b>97.94</b>	30.98	<b>29.09</b>
Orthophosphates concentration (µM P)	1.31	<b>0.99</b>	1.27	<b>1.11</b>	0.68	<b>0.56</b>
Total Phosphorus concentration (µM P)	1.87	<b>1.55</b>	1.42	<b>1.25</b>	1.16	<b>1.03</b>
N:P ratio	120.85	<b>87.10</b>	79.02	<b>62.24</b>	45.28	<b>30.32</b>
Chlorophyll <i>a</i> concentration (µg L <sup>-1</sup> )	2.90	<b>-0.96</b>	0.80	<b>0.03</b>	0.59	<b>0.29</b>
Phaeo-pigments concentration (µg L <sup>-1</sup> )	0.17	<b>-0.51</b>	0.23	<b>0.15</b>	0.23	<b>-0.15</b>
Pigment diversity index	2.17	<b>-0.03</b>	2.29	<b>0.23</b>	2.51	<b>0.16</b>
<b>Sediment parameters</b>						
Clay content (%)	2.69	<b>0.13</b>	3.98	<b>-7.96</b>	0.00	<b>0.00</b>
Silt content (%)	3.08	<b>1.12</b>	54.58	<b>7.35</b>	0.00	<b>0.00</b>
Sand content (%)	94.23	<b>-1.25</b>	41.44	<b>0.62</b>	100.00	<b>0.01</b>
Organic matter content (%)	2.34	<b>-1.25</b>	9.13	<b>7.26</b>	0.57	<b>0.31</b>
Chlorophyll <i>a</i> concentration (µg g <sup>-1</sup> )	19.74	<b>13.30</b>	6.93	<b>3.59</b>	0.42	<b>-0.25</b>
Phaeo-pigments concentration (µg g <sup>-1</sup> )	14.89	<b>11.08</b>	12.70	<b>10.86</b>	0.81	<b>0.26</b>
Chlorophyll <i>a</i> degradation index (%)	42.90	<b>5.51</b>	64.70	<b>29.18</b>	66.14	<b>21.12</b>
Pigment diversity index	3.64	<b>0.64</b>	3.43	<b>0.55</b>	2.89	<b>0.55</b>

To what concerns sediment parameters, chlorophyll *a*, phaeo-pigments concentrations, chlorophyll *a* degradation index, clay and silt contents were the parameters with greater variations.

At all stations, there was an increase of phaeo-pigments concentration, chlorophyll *a* degradation index and pigment diversity.

The upstream station presented the smallest variations for chlorophyll *a* degradation index (5.51%) and the greatest variations of chlorophyll *a* (13.30 µg g<sup>-1</sup>), phaeo-pigments concentration (11.08 µg g<sup>-1</sup>), pigment diversity (0.64) and sand content (-1.25%).

The major variations of chlorophyll *a* degradation index (29.18%), clay content (-7.96%), silt content (7.35%) and organic matter content (7.26%) occurred at the intermediate station. The minor variation of pigment diversity (0.55) was observed in the intermediate and in the downstream stations. Downstream was also the station with

the smallest variation in organic matter content (0.31%), chlorophyll *a* (-0.25  $\mu\text{g g}^{-1}$ ) and phaeo-pigments concentrations (0.26  $\mu\text{g g}^{-1}$ ).

### 2.3.1.6. Trophic state and water quality

The trophic state index of Carlson determined with chlorophyll *a*, TSI (CHL), presented lower values than the one determined with total phosphorus concentrations, TSI (TP) (Figure 2.13). TSI (CHL) was minimum in June 2001 and December (Downstream: 18.79) and maximum in March 2002 (Intermediate: 56.12), corresponding to oligotrophic and eutrophic conditions respectively, but most of the samples presented values that indicate oligotrophy (46%) and mesotrophy (29%). Positive correlations were determined between TSI (CHL) values and nitrates ( $\rho = 0.541$ ;  $p = 0.006$ ), water level ( $\rho = 0.505$ ;  $p = 0.012$ ) and nitrites ( $r = 0.489$ ;  $p = 0.015$ ), besides chlorophyll *a*. The mean annual TSI (CHL) values determined for each station, showed a small decrease according to the distance from the sea (upstream:  $37.41 \pm 10.57$ ; intermediate:  $35.28 \pm 11.88$ ; downstream:  $33.75 \pm 13.06$ ), nevertheless all stations were in the category of oligomesotrophic conditions.

TSI (TP) lowest value was calculated in December 2001 (Downstream: 24.14) and the highest in August 2001 (Intermediate: 67.16). These values correspond to oligotrophic and eutrophic-hypereutrophic systems, nevertheless for most samples, TSI (TP) values suggest mesotrophic (25%) and eutrophic (42%) conditions. Apart from total phosphorus, TSI (TP) values were correlated with orthophosphates ( $r = 0.872$ ;  $p < 0.001$ ) and temperature ( $r = 0.412$ ;  $p = 0.045$ ). The mean annual TSI (TP) values determined for each station were quite similar (upstream:  $52.07 \pm 8.27$ ; intermediate:  $52.09 \pm 12.52$ ; downstream:  $51.19 \pm 12.58$ ) and thereby all stations were in the category of eutrophic conditions.

The trophic state index (TRIX) ranged from 3.62 (December 2001, downstream) to 6.61 (July 2002, upstream). Penna *et al.* (2004) made a correspondence between TRIX and water quality in coastal zones and according to their criteria, the water changed from high quality, characteristic of a system poorly productive with a low trophic level (TRIX: 2-4), to poor quality, highly productive and with the greatest trophic level (TRIX: 6-8). Along the year, the majority of the samples had a mediocre (42%) and

poor (29%) water quality, typical from moderate to high productive systems, with a high trophic level. TRIX values were positively correlated to chlorophyll *a* ( $r = 0.690$ ;  $p < 0.001$ ), total phosphorus ( $r = 0.656$ ;  $p = 0.001$ ), DIN ( $r = 0.513$ ;  $p = 0.010$ ) and also with nitrates ( $r = 0.546$ ;  $p = 0.006$ ), nitrites ( $r = 0.536$ ;  $p = 0.007$ ) and orthophosphates ( $r = 0.507$ ;  $p = 0.011$ ). Both Carlson's indexes were highly correlated ( $p < 0.001$ ) with TRIX (TSI CHL:  $r = 0.683$ ; TSI TP:  $r = 0.731$ ), but no significant correlation was found between them ( $r = 0.294$ ;  $p = 0.163$ ).

The mean annual TRIX values determined for each station, classified them all as mediocre water quality, although values slightly decreased with the proximity to the sea (upstream:  $5.58 \pm 0.85$ ; intermediate:  $5.32 \pm 0.67$ ; downstream:  $5.23 \pm 0.85$ ).

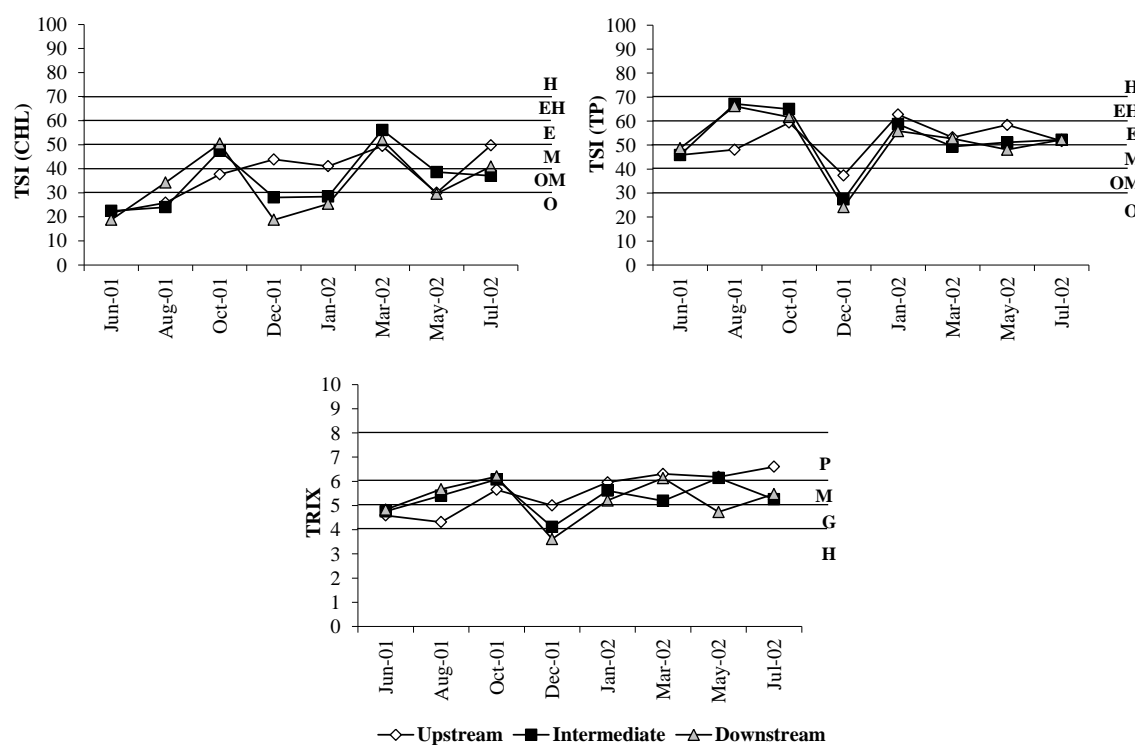


Figure 2.13 – Seasonal variation of trophic state and water quality indexes in Foz de Almargem coastal lagoon: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX.

TSI: O- Oligotrophic; OM- Oligomesotrophic; M- Mesotrophic; E- Eutrophic; EH- Eutrofic to Hypereutrophic; H- Hypereutrophic. TRIX: H- High water quality; G- Good water quality; M- Mediocre water quality; P- Poor water quality.

Considering only the data from sampling periods when the lagoon was closed (June-December 2001; March-July 2002), the trophic state and water quality mean values of the stations were similar or slightly lower than the values obtained with all data from the studied period (Table 2.7). Just TSI (CHL) means in the intermediate and downstream

stations were somewhat higher. So, there was no difference between the classification of the stations with data from the period the lagoon was closed and the classification of trophic state and water quality with all data.

Table 2.7 – Trophic state and water quality indexes mean values, standard deviation and classification in Foz de Almargem sampling stations, determined during all studied period (June 2001-July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
TSI (CHL): All data	37.41±10.57	35.28±11.88	33.75±13.06
	<b>Oligomesotrophic</b>	<b>Oligomesotrophic</b>	<b>Oligomesotrophic</b>
TSI (CHL): Closed period	36.90±11.30	36.26±12.48	34.95±13.63
	<b>Oligomesotrophic</b>	<b>Oligomesotrophic</b>	<b>Oligomesotrophic</b>
TSI (TP): All data	52.07±8.27	52.09±12.52	51.19±12.58
	<b>Eutrophic</b>	<b>Eutrophic</b>	<b>Eutrophic</b>
TSI (TP): Closed period	50.55±7.64	51.15±13.21	50.53±13.44
	<b>Eutrophic</b>	<b>Eutrophic</b>	<b>Eutrophic</b>
TRIX: All data	5.58±0.85	5.32±0.67	5.23±0.85
	<b>Mediocre water quality</b>	<b>Mediocre water quality</b>	<b>Mediocre water quality</b>
TRIX: Closed period	5.52±0.90	5.28±0.71	5.24±0.91
	<b>Mediocre water quality</b>	<b>Mediocre water quality</b>	<b>Mediocre water quality</b>

The comparison of the data from December 2001 (closed lagoon) and January 2002 (open lagoon), showed an increase in the three indexes values at all stations, except TSI (CHL) in the upstream station ( $\Delta = -2.81$ ). The smallest variation in TSI (CHL) was determined at the intermediate station ( $\Delta = 0.37$ ) and the greatest variation happened downstream ( $\Delta = 6.63$ ). Despite values variation, in January 2002 (TSI CHL<sub>upstream</sub> = 41.04; (TSI CHL<sub>intermediate</sub> = 28.41; TSI CHL<sub>downstream</sub> = 25.42) the classification of the stations was the same as in December 2001 (TSI CHL<sub>upstream</sub> = 43.85; TSI CHL<sub>intermediate</sub> = 28.04; TSI CHL<sub>downstream</sub> = 18.79), when the lagoon was isolated from the sea: upstream – mesotrophic; intermediate- oligotrophic; downstream- oligotrophic.

TSI (TP) and TRIX showed the lowest variations upstream ( $\Delta$ TSI TP = 25.35;  $\Delta$ TRIX = 0.96) and the highest variations downstream ( $\Delta$ TSI TP= 31.68;  $\Delta$ TRIX= 1.59). In the intermediate station, the variations of TSI (TP) ( $\Delta = 31.36$ ) and TRIX ( $\Delta = 1.50$ ) were closer to the ones in the downstream station.

According to TSI (TP) classification, all stations increased its trophic state. When the lagoon was closed, in December 2001, the upstream station was considered oligotrophic-mesotrophic (TSI TP = 37.35) and in January 2002 became eutrophic-

hypereutrophic (TSI TP = 62.70) . Both intermediate (TSI TP = 27.36) and downstream (TSI TP = 24.14) stations were classified as oligotrophic in December 2001 and when the lagoon was opened became eutrophic (TSI TP<sub>intermediate</sub> = 58.72; TSI TP<sub>downstream</sub> = 55.82),.

The TRIX evaluation indicated that in January 2002 all stations had a mediocre water quality (TRIX<sub>upstream</sub> = 5.96; TRIX<sub>intermediate</sub> = 5.62; TRIX<sub>downstream</sub> = 5.21). In December 2001, the upstream station already had this classification (TRIX<sub>upstream</sub> = 5.00), but the intermediate station showed a good water quality (TRIX<sub>intermediate</sub> = 4.12) and the downstream station had a high water quality (TRIX<sub>downstream</sub> = 3.62).

The 90<sup>th</sup> percentile of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ) was determined as proposed by Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012) to assess the water quality in Portuguese coastal lagoons. These authors consider different reference values according to coastal lagoons typology and periods of the year, so a comparison was made between the results obtained with each approach (Table 2.8).

Pereira Coutinho *et al.* (2012) defined two reference values for chlorophyll *a*, 20  $\mu\text{g L}^{-1}$  when the lagoons were closed and 6.7  $\mu\text{g L}^{-1}$  during the periods of connection with the sea.

Data from Foz de Almargem only included one sampling with the lagoon opened (January 2002), thereby the 90<sup>th</sup> percentile of chlorophyll *a* was calculated just for the periods when the lagoon was closed. The results found in all stations were under the reference value and the boundary of 30  $\mu\text{g L}^{-1}$ , that distinguishes High from Good water quality. So, during the period the lagoon was closed, water quality was high in all stations. Although there were no sufficient data to determine the 90<sup>th</sup> percentile of chlorophyll *a* for the opened period, in January 2002 all stations presented chlorophyll *a* values below the reference value of 6.7  $\mu\text{g L}^{-1}$  (upstream: 2.90  $\mu\text{g L}^{-1}$ ; intermediate: 0.80  $\mu\text{g L}^{-1}$ ; downstream: 0.59  $\mu\text{g L}^{-1}$ ).

When Foz de Almargem was closed, chlorophyll *a* was positively correlated with nitrites and nitrates concentrations and significant linear regressions were determined between chlorophyll *a* and nitrites concentrations ( $R^2= 0.313$ ;  $F= 8.638$ ;  $p = 0.008$ ) and between chlorophyll *a* and nitrates concentrations ( $R^2= 0.229$ ;  $F= 5.629$ ;  $p = 0.028$ ) (Figure 2.14).

The reference value defined by Brito *et al.* (2012), 5.3  $\mu\text{g L}^{-1}$ , considers the phytoplankton growing season in southern coastal water lagoons (February to October).

As data collected in Foz de Almargem do not cover the all growing season of 2001 nor 2002, the 90<sup>th</sup> percentile of chlorophyll *a* was calculated separately with the data from 2001 (June to October) and 2002 (March to July) and then, with data from the two years together (June to October 2001; March to July 2002).

The results from the growing season of 2001 (June to October), corresponded to high water quality in all stations, with values being lower than 8 µg L<sup>-1</sup>, which is the boundary value for High and Good water quality. Considering the data from the growing season of 2002 (March to July), the upstream and downstream stations were still classified with high water quality, although values were greater than those determined for the growing season of 2001. In the intermediate station, there was a change in water quality from high to good, as the 90<sup>th</sup> percentile of chlorophyll *a* was between 8 and 12 µg L<sup>-1</sup>. If data from the two growing season were analyzed together (June to October 2001; March to July 2002), then the upstream station had high water quality, but the intermediate and the downstream stations had good water quality.

Table 2.8 – Water quality in Foz de Almargem sampling stations, based on the 90<sup>th</sup> percentile of chlorophyll *a* (µg L<sup>-1</sup>), adapted from Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012).

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
<b>Pereira Coutinho <i>et al.</i> (2012): Semi-enclosed lagoons</b>	<i>Ref. value closed period (20 µg L<sup>-1</sup>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> during closed period	6.87 µg L <sup>-1</sup>	7.95 µg L <sup>-1</sup>	7.92 µg L <sup>-1</sup>
<b>Classification of Water Quality during closed period</b>	<b>High</b> (< 30 µg L <sup>-1</sup> )	<b>High</b> (< 30 µg L <sup>-1</sup> )	<b>High</b> (< 30 µg L <sup>-1</sup> )
<b>Brito <i>et al.</i> (2012): Coastal water lagoons</b>	<i>Ref. value growing season (February-October: 5.3 µg L<sup>-1</sup>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> (June-October 2001)	1.76 µg L <sup>-1</sup>	4.60 µg L <sup>-1</sup>	6.34 µg L <sup>-1</sup>
<b>Classification of Water Quality</b>	<b>High</b> (< 8 µg L <sup>-1</sup> )	<b>High</b> (< 8 µg L <sup>-1</sup> )	<b>High</b> (< 8 µg L <sup>-1</sup> )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (March-July 2002)	6.97 µg L <sup>-1</sup>	11.24 µg L <sup>-1</sup>	7.57 µg L <sup>-1</sup>
<b>Classification of Water Quality</b>	<b>High</b> (< 8 µg L <sup>-1</sup> )	<b>Good</b> (8-12 µg L <sup>-1</sup> )	<b>High</b> (< 8 µg L <sup>-1</sup> )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (Jun-Oct 2001; Mar-Jul 2002)	6.91 µg L <sup>-1</sup>	9.53 µg L <sup>-1</sup>	8.16 µg L <sup>-1</sup>
<b>Classification of Water Quality</b>	<b>High</b> (< 8 µg L <sup>-1</sup> )	<b>Good</b> (8-12 µg L <sup>-1</sup> )	<b>Good</b> (8-12 µg L <sup>-1</sup> )

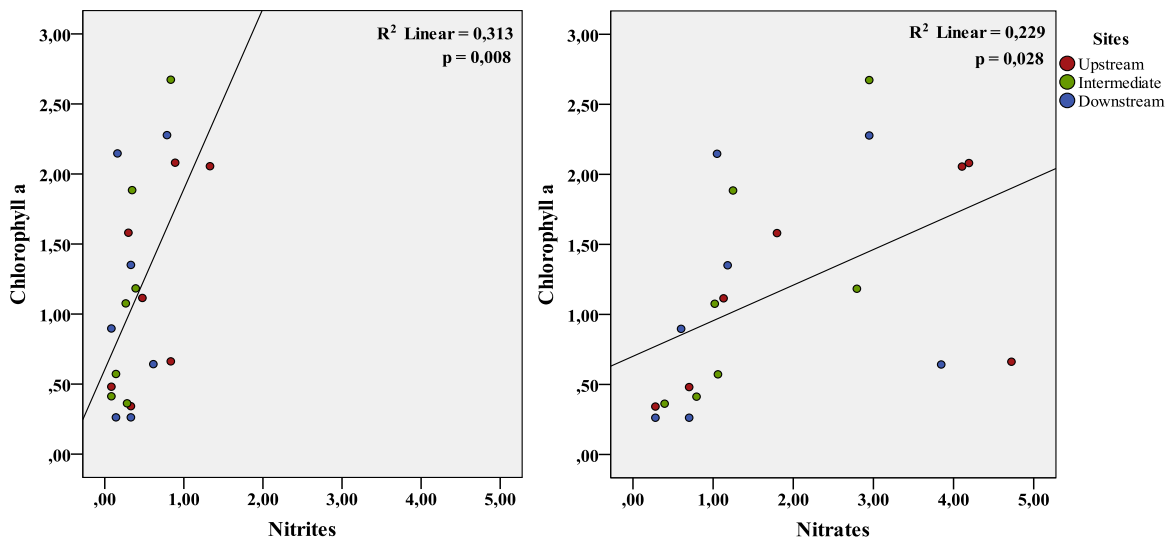


Figure 2.14 – Linear regression between chlorophyll *a* concentration ( $\mu\text{g L}^{-1}$ ) and the concentrations of nitrites ( $\mu\text{M}$ ) and nitrates ( $\mu\text{M}$ ), during the period Foz de Almagem lagoon was closed. Data transformed by  $\ln(x+1)$ .

## 2.3.2. Salgados coastal lagoon

### 2.3.2.1. Hydrological aspects

The raining season started in September 2001 and ended in May 2002 (Figure 2.15). December and March were the months with higher cumulative rainfall (183.5 mm and 103.5 mm) and number of raining days (19 and 22 days). In September, cumulative rainfall reached 95.7 mm but it was concentrated in just 7 days. Most of the time, daily rainfall was below 60 mm and only in September and December the maximum values of 63.7 mm and 66.0 mm were registered.

Water level in the deepest part of the lagoon varied between 3.00 m and 5.05 m. The minimum value occurred after the opening of the lagoon and the maximum was registered in April. During the raining season water level tended to increase, till the sand barrier was artificially or naturally destroyed causing a sudden decrease to around 3.00 m. In spring and summer, the greater evaporation of water and the reduction of rainfall runoff contributed to the lowering of water level. Nevertheless, the minimum value for this time of the year was still high (3.95 m) and can be explained by the volume of wastewater discharged into the lagoon. Although there are no data available for the studied period concerning the seasonal variation of wastewater discharges, some authors

have described similar patterns in previous years (e.g. PROCESL, 1998 in Neves, 1999; Pinto *et al.* 2001).

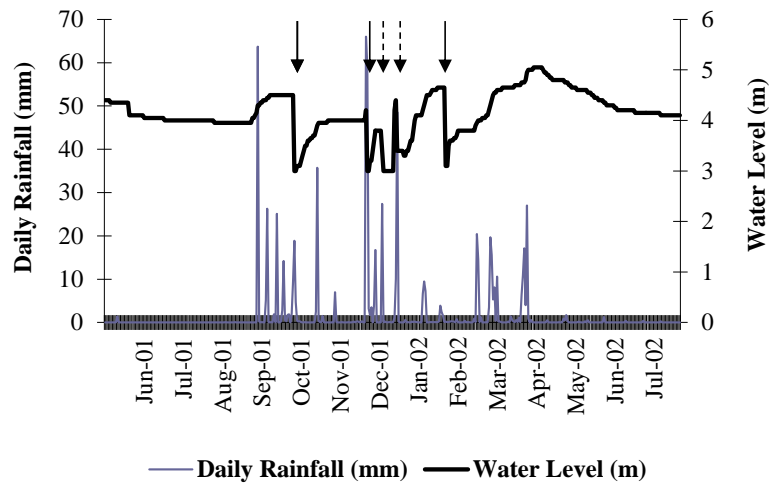


Figure 2.15 - Variation of daily rainfall registered in Algoz meteorological station and daily water level measured in the deepest part of Salgados coastal lagoon. Full arrows indicate artificial opening of the lagoon and dashed arrows represent natural opening.

While the present study was being developed, the lagoon was artificially opened to the sea in three occasions, October, December and February (Table 2.9). After the intervention made in December, two natural openings occurred (December and January). The natural destruction of the sand barrier happened with lower water levels than the artificial openings, as the sand barrier was thinner and more fragile.

Table 2.9 - Periods of connection with the sea and water level in Salgados coastal lagoon.

<i>Date</i>	<i>Opening process</i>	<i>Water level (m) before lagoon opening</i>	<i>Water level (m) when the lagoon was opened</i>	<i>Water level (m) after lagoon closing</i>
October 2001	Artificial	4.50	3.00	3.30
December 2001	Artificial	4.70	3.00	3.80
December 2001	Natural	3.80	3.00	4.20
January 2002	Natural	4.40	3.30	3.50
February 2002	Artificial	4.65	3.10	3.50

### 2.3.2.2. Water parameters

Water temperature fluctuated between 14.70 °C (December 2001) and 25.75 °C (August 2001) (Figure 2.16), presenting higher values in summer and lower values in winter.

Salinity ranged from 2.00 ‰ (October 2001) to 23.10 ‰ (January 2002). During the summer of 2001 there was no rainfall and river runoff was low, nevertheless salinity

values decreased due to the freshwater inputs from wastewater discharges, which overcame the water lost by evaporation. The starting of the raining season and the consequent raise of rivers runoff contributed to the lowering of salinity in October. In December, values were still low and somehow similar to the ones registered in October, before the artificial opening of the lagoon. January was the month with greater differences between sampling stations, revealing a gradient of salinity according to the distance from the sea. The increase of salinity and the gradient among sampling stations can be attributed to the entrance of seawater during a longer period of time in the end of December and beginning of January, when the lagoon was naturally in connection with the sea. Although the lagoon was artificially opened in February, the short period of connection and the rivers runoff associated to rainfall caused the decrease of salinity in March and May. In July, the freshwater inputs were mainly from the wastewater discharges and salinity was lower than in the summer of 2001.

The higher values of pH were observed in the upstream station except in June 2001 and July 2002. Minimum pH was 7.48 in the intermediate sampling station (June 2001) and the maximum was 9.98 in the upstream station (December 2001).

Dissolved oxygen concentration and oxygen saturation (%) presented a highly significant positive correlation ( $r = 0.997$ ;  $p < 0.001$ ). The upstream station showed higher values most of the time and the maximum ( $22.20 \text{ mg L}^{-1}$  and  $240.50 \%$ ) was registered in December 2001. Minimum values ( $4.35 \text{ mg L}^{-1}$  and  $56.00 \%$ ) were found in the intermediate station in June 2001.

Upstream and intermediate stations showed higher concentrations of total solids in suspension (TSS). The maximum values were determined in August ( $260 \text{ mg L}^{-1}$ ) and October 2001 ( $246 \text{ mg L}^{-1}$ ) and might have been caused by the increase of wastewater discharges during this period of greater tourist affluence.

During the 24 hours cycle made in May 2002, all parameters monitored except salinity, showed greater variations in the intermediate sampling station, increasing during the day and decreasing during the night (Figure 2.17). Nevertheless, apart from salinity, mean values were higher in the downstream station (Table 2.10).

Salinity was approximately the same in the two sampling stations and did not oscillate along the cycle. Water temperature varied  $8.19 \text{ }^\circ\text{C}$  in the intermediate station and  $2.48 \text{ }^\circ\text{C}$  downstream and in both stations a highly significant correlation was found between water temperature and air temperature (Intermediate:  $r = 0.855$ ,  $p < 0.001$ ;

Downstream:  $r = 0.830$ ,  $p < 0.001$ ). The greater fluctuation in the intermediate station can be explained by the lower depth (around 1 m) while the downstream station was located near the deepest part of the lagoon (> 4m).

Dissolved oxygen concentration oscillated  $9.31 \text{ mg L}^{-1}$  (113.5 % oxygen saturation) in the intermediate station and  $2.22 \text{ mg L}^{-1}$  (27.6 % oxygen saturation) downstream. The pH amplitude in the intermediate station was 0.77 and 0.12 downstream. The differences in dissolved oxygen concentration, oxygen saturation and pH observed between sampling stations, were probably related to the abundance of autotrophic organisms and bacteria in these stations. During the day, photosynthesis consumes carbon dioxide and produces oxygen and during the night, through respiration and bacteria activity, oxygen is consumed and carbon dioxide is released.

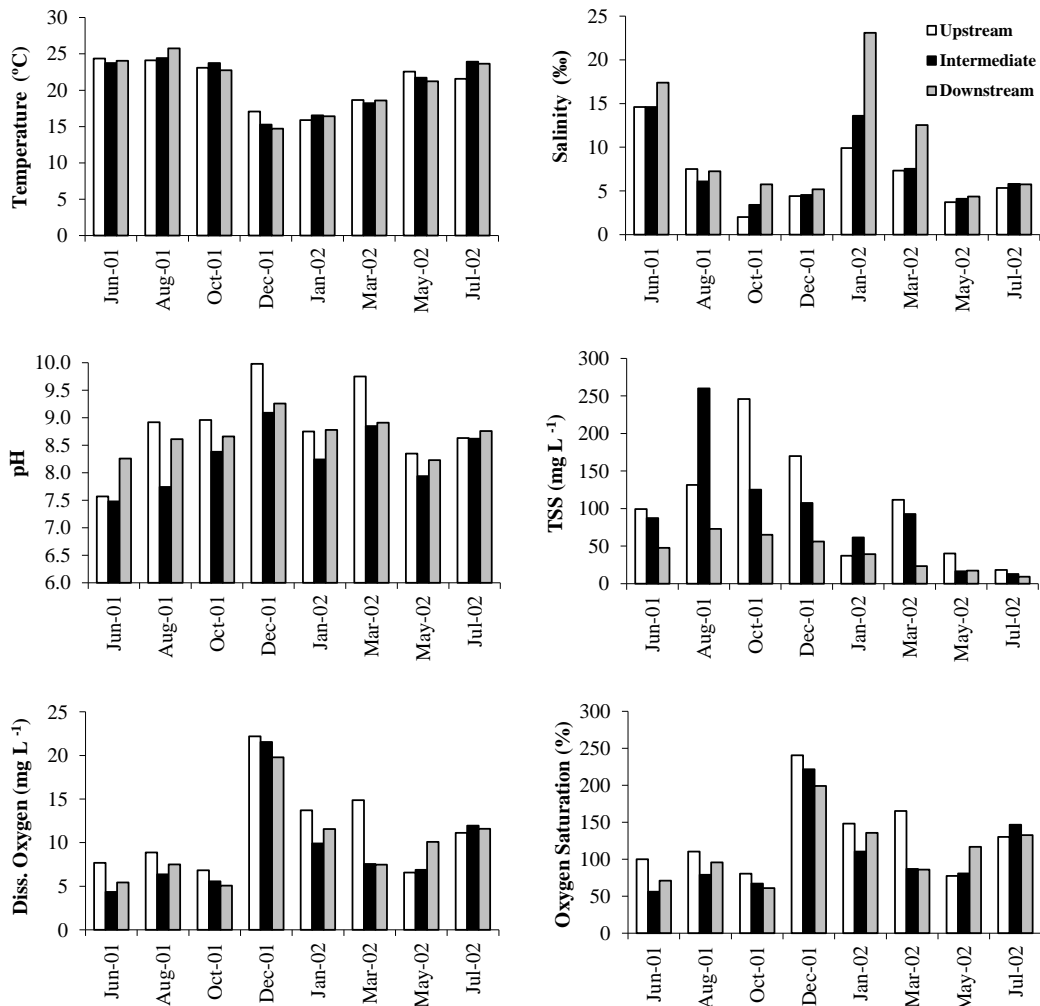


Figure 2.16 - Seasonal variation of physical and chemical water parameters (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Salgados sampling stations.

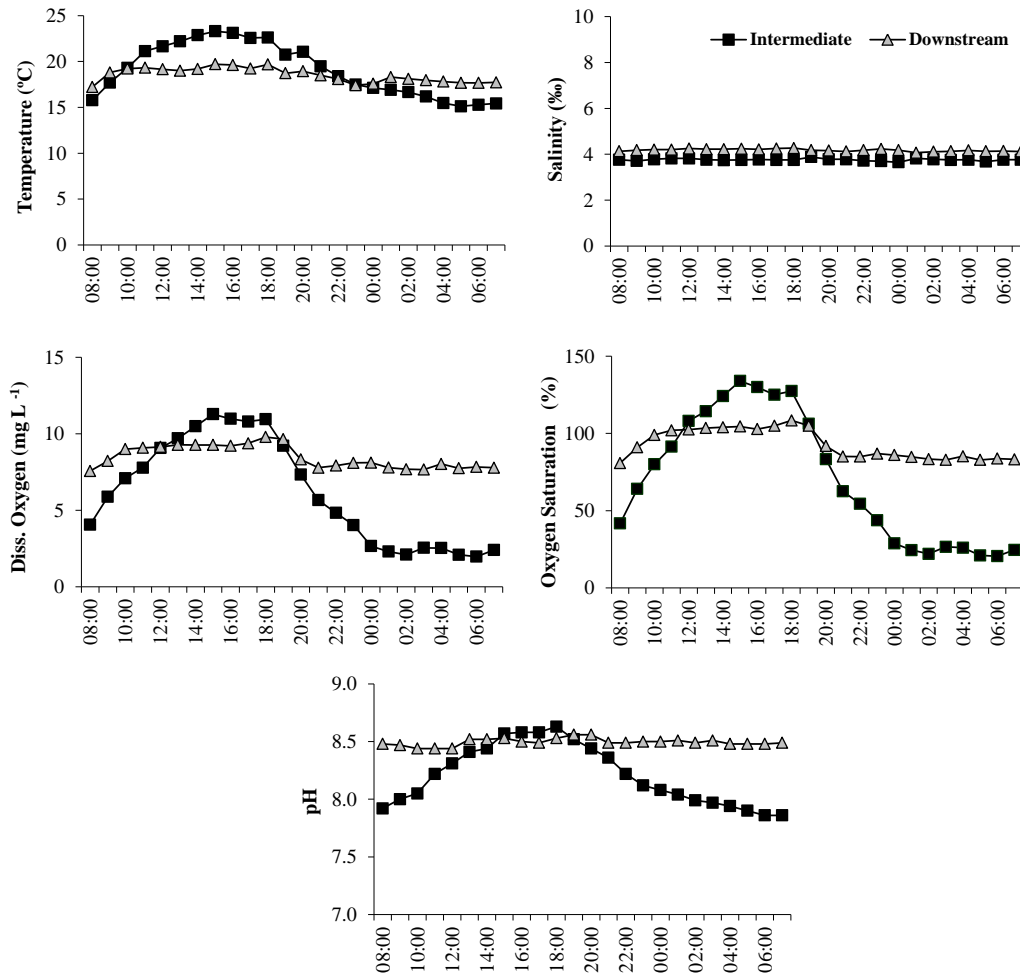


Figure 2.17 - Daily variation of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) during spring (May 2002) in the intermediate and downstream sampling stations from Salgados coastal lagoon.

Table 2.10 – Minimum, maximum and mean values of physical and chemical water parameters (temperature, salinity, pH, dissolved oxygen and oxygen saturation) monitored during a 24 hours cycle in spring (May 2002) at the intermediate and downstream sampling stations from Salgados lagoon.

	<i>Intermediate Station</i>			<i>Downstream Station</i>		
	Minimum	Maximum	Mean	Minimum	Maximum	Mean
Temperature (°C)	15.11	23.3	19.06	17.22	19.70	18.52
Salinity (%)	3.65	3.88	3.76	4.07	4.27	4.18
pH	7.86	8.63	8.21	8.44	8.56	8.50
Dissolved oxygen concentration (mg L <sup>-1</sup> )	1.97	11.28	6.16	7.57	9.97	8.48
Oxygen saturation (%)	20.50	134.00	70.20	80.80	108.40	92.89

Dissolved inorganic nitrogen concentration (DIN) reached the maximum value in August 2001 (498.52  $\mu\text{M N}$ ) because of the high concentration of ammonia (486.11  $\mu\text{M N}$ ), which represented 97.51 % of DIN (Figure 2.18). In January 2002, a smaller increase of DIN was associated to the nitrates peak (118.87  $\mu\text{M N}$ ). Nitrites maximum occurred in March 2002 (26.74  $\mu\text{M N}$ ) and August 2001 (22.83  $\mu\text{M N}$ ), but its contribution to DIN seasonal variation was less important than nitrates or ammonia.

Total phosphorus concentration (TP) followed the same seasonal fluctuation as orthophosphates, increasing during the summer (maximum TP: 161.29  $\mu\text{M P}$ ; maximum orthophosphates: 142.95  $\mu\text{M P}$ ) and decreasing in winter (minimum TP: 20.48  $\mu\text{M P}$ ; minimum orthophosphates: 14.53  $\mu\text{M P}$ ). Orthophosphates accounted 68.72 % (June 2001, upstream) to 99.11% (August 2001, downstream) of total phosphorus concentration.

The N: P ratio in all stations was lower than the Redfield ratio (16:1), ranging from 0.04 (July 2002, downstream) to 7.62 (January 2002, intermediate).

Chlorophyll *a* concentration varied from 13.23  $\mu\text{g L}^{-1}$  (March 2002, downstream) to 661.66  $\mu\text{g L}^{-1}$  (March 2002, upstream) (Figure 2.19). The upstream station was the one with higher concentrations of chlorophyll *a*. The maximum phaeo-pigments concentration (62.64  $\mu\text{g L}^{-1}$ ) was found in the downstream station, during July 2002. Margalef's pigment diversity index fluctuated between 1.75 (December 2001, intermediate) and 3.29 (May 2002, upstream).

The upstream station presented higher means of pH, total solids in suspension, dissolved oxygen concentration, chlorophyll *a* concentration, pigment diversity and lower means of salinity, nitrogen compounds (nitrates, nitrites, ammonia and total dissolved inorganic nitrogen) and N:P ratio (Table 2.11).

In the intermediate station, ammonia concentration, total dissolved inorganic nitrogen, phosphorus compounds (orthophosphates and total phosphorus), N:P ratio and phaeo-pigments concentration registered the greatest mean values, while pH and dissolved oxygen concentration showed the lowest means.

The highest means for salinity, nitrates and nitrites were determined in the downstream station. This station also had the lowest means for total solids in suspension, chlorophyll *a*, phaeo-pigments concentration and pigment diversity.

Although stations presented some differences in the mean values of water parameters, the results from one-way analysis of variance and the Kruskal-Wallis test (Appendix I.C) indicated that none of these differences among stations were statistically significant ( $p > 0.05$ ).

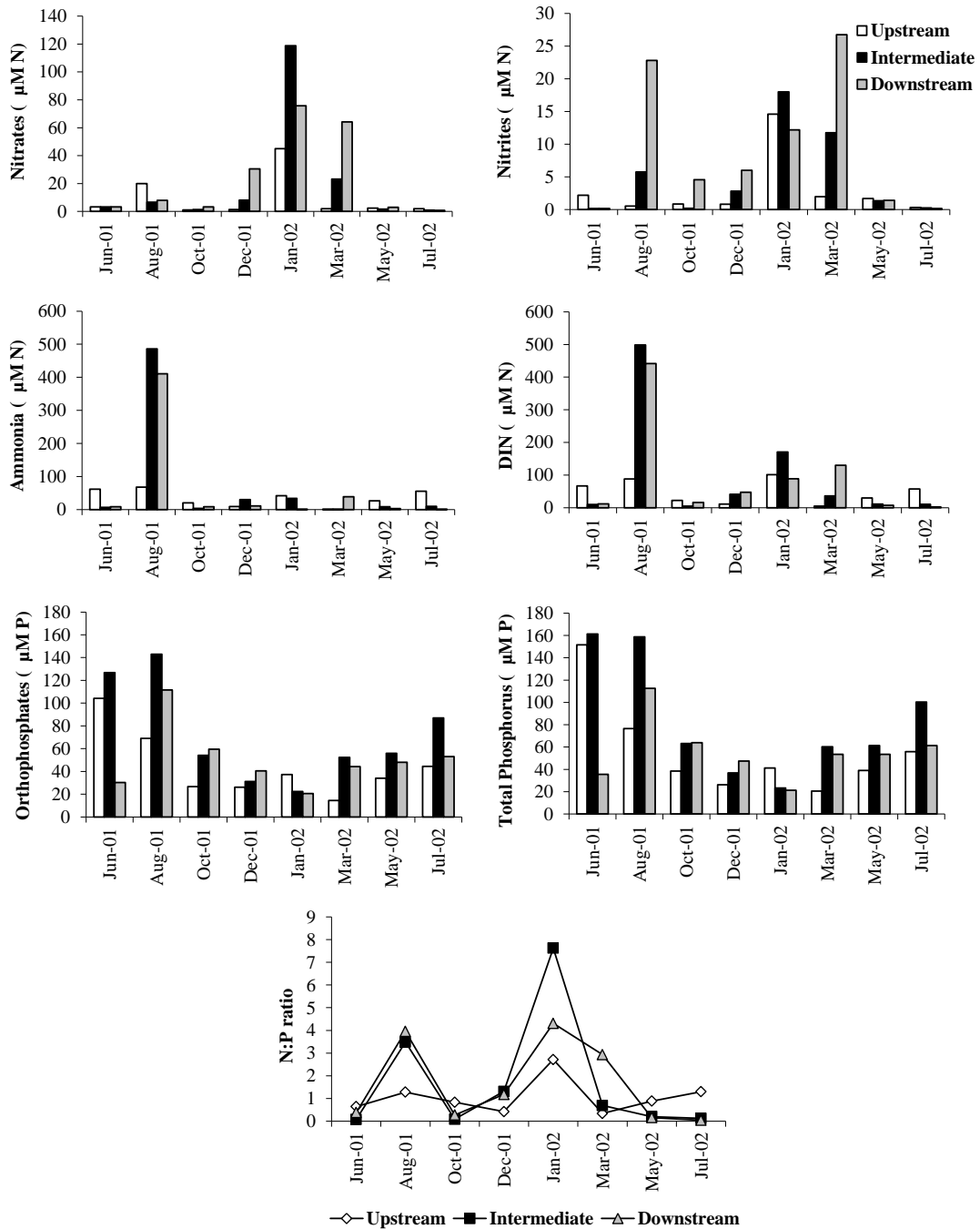


Figure 2.18 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration, N: P ratio (DIN: orthophosphates) and total solids in suspension (TSS) in Salgados sampling stations.

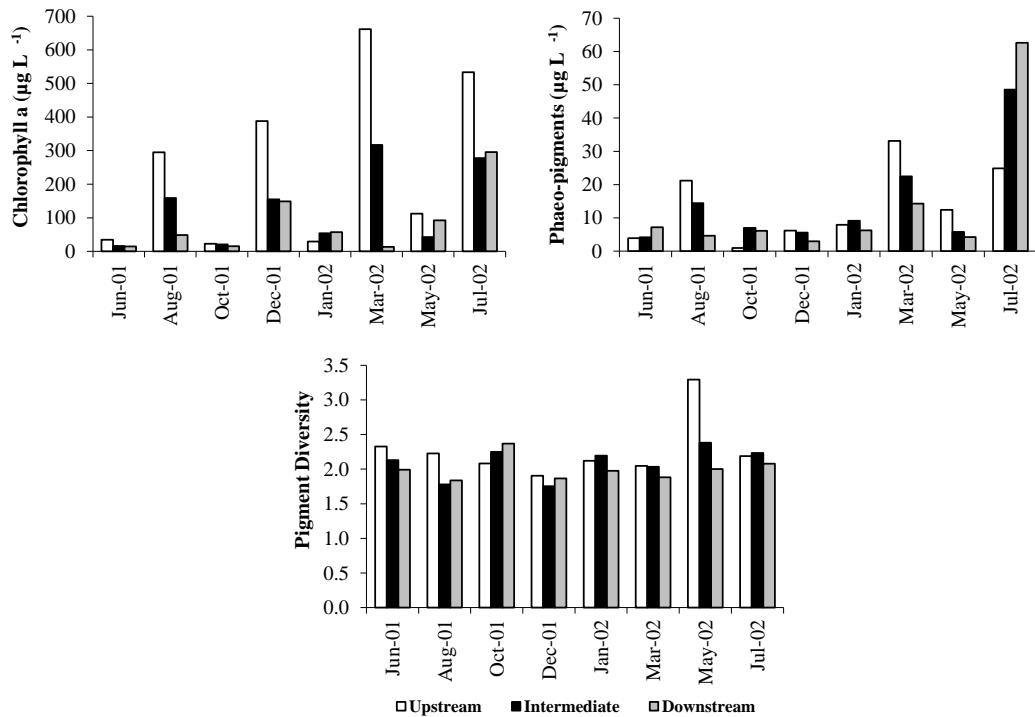


Figure 2.19 - Seasonal variation of photosynthetic pigments concentration (chlorophyll *a* and phaeo-pigments) and Margalef's pigment diversity index in Salgados sampling stations.

Table 2.11 – Annual mean values and standard deviation of water parameters in Salgados sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Temperature (°C)	20.90±3.29	20.95±3.71	20.89±3.94
Salinity (‰)	6.85±3.99	7.46±4.30	10.17±6.88
pH	8.86±0.77	8.29±0.55	8.68±0.34
Total Solids in Suspension	106.70±76.75	95.36±77.89	41.35±23.14
Dissolved oxygen concentration (mg L <sup>-1</sup> )	11.48±5.32	9.27±5.52	9.81±4.76
Nitrates concentration (µM N)	9.62±15.64	20.50±40.41	23.59±30.33
Nitrites concentration (µM N)	2.87±4.79	5.01±6.59	9.24±10.43
Ammonia concentration (µM N)	35.37±25.02	72.41±167.61	60.34±142.04
Total dissolved inorganic nitrogen concentration (µM N)	47.86±36.10	97.93±107.87	93.17±147.79
Orthophosphates concentration (µM P)	44.56±28.97	71.62±43.65	51.02±27.41
Total Phosphorus concentration (µM P)	56.16±42.30	83.16±52.42	56.13±26.73
N:P ratio	1.05±0.76	1.70±2.66	1.65±1.80
Chlorophyll a concentration (µg L <sup>-1</sup> )	259.61±249.50	130.11±117.39	85.67±96.69
Phaeo-pigments concentration (µg L <sup>-1</sup> )	13.82±11.40	14.63±14.98	13.53±20.14
Pigment diversity index (bits)	2.28±0.43	2.10±0.23	2.00±0.17

The PCA ordination biplot showed that the major environmental variables influencing sampling stations ordination along the first axis (I) were orthophosphates (component loading = 0.891), total phosphorus (component loading = 0.878), temperature (component loading = 0.853), pH (component loading = -0.712), dissolved oxygen (component loading = -0.622) and ammonia concentration (component loading = 0.605) (Figure 2.20). On the right side of the axis were plotted the stations with higher values of orthophosphates, total phosphorus, temperature, ammonia concentration and lower pH and dissolved oxygen (June and August 2001). The stations on the left side of axis I were characterized by the opposite variation of the same parameters (December 2001, January and March 2002). The first axis accounted for 27.7% of the total variance. The placement of sampling stations along the secondary axis (II) was mainly related to N: P ratio (component loading = 0.897), nitrites concentration (component loading = 0.820), nitrates concentration (component loading = 0.744) and total dissolved inorganic nitrogen (component loading = 0.735). On the upper side of the diagram were displayed samples with higher values of N: P ratio, nitrites, nitrates and total dissolved inorganic nitrogen concentrations, salinity, rainfall and lower values of phaeo-pigments concentration, chlorophyll *a* concentration, pigments diversity, water level (*e.g.* intermediate January 2002) and *vice-versa* for the bottom side of the diagram (*e.g.* downstream July 2002). The second axis explained 25.2 % of the total variance. Water level, chlorophyll *a* and phaeo-pigments concentrations, pigments diversity and TSS were less relevant for the first and second axes, presenting higher component loadings in the third and fourth axis.

The environmental variables disposed near each other on the ordination diagram presented high positive correlation coefficients: pH and dissolved oxygen; cumulative rainfall and nitrates; nitrites, N: P and salinity; DIN and ammonia; TSS, total phosphorus and orthophosphates; water level and pigment diversity; chlorophyll *a* and phaeo-pigments. Those variables pointing in opposite directions were negatively correlated, as orthophosphates concentration and pH or N:P ratio and pigments diversity index.

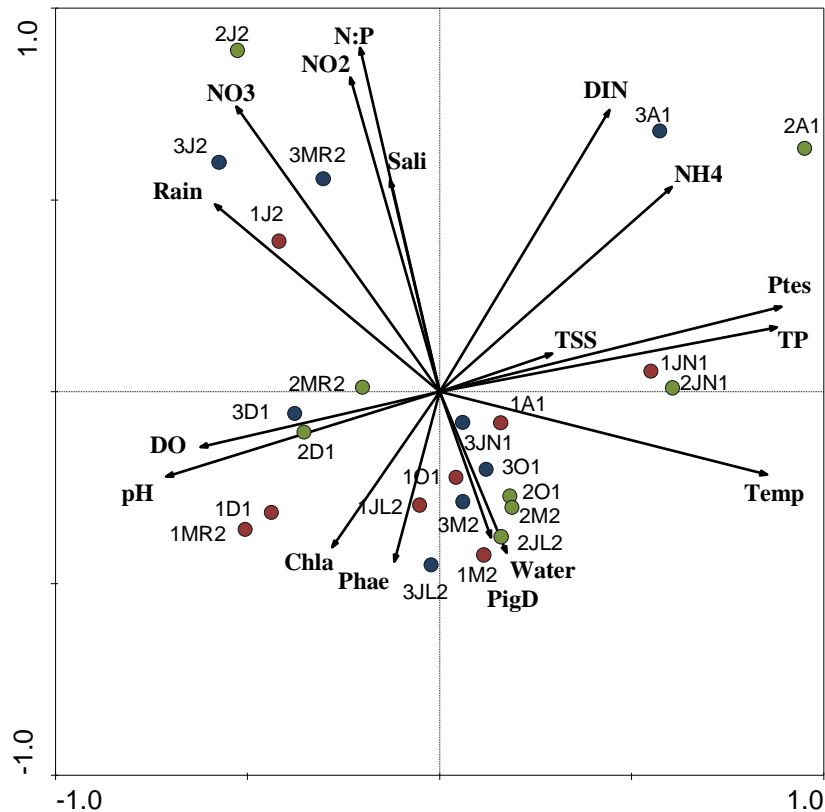


Figure 2.20 - Principal Component Analysis performed on the hydrological and water parameters from Salgados sampling stations. Cumulative percentage variance explained by axes: I – 27.7 %; I + II – 52.9 %.

*Station codes:* First character corresponds to the sampling station (1- upstream, 2- intermediate, 3- downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001, 2-2002).

*Environmental variables:* Rain- cumulative rainfall in 10 days previous to sampling; Water- water level in the lagoon; Temp- water temperature; Sali- salinity; pH; DO- dissolved oxygen concentration; TSS- total solids in suspension; NO<sub>3</sub>- nitrates concentration; NO<sub>2</sub>- nitrites concentration; NH<sub>4</sub>- ammonia concentration; DIN- total dissolved inorganic nitrogen concentration; Ptes- orthophosphates concentration; TP- total phosphorus concentration; N: P- DIN and TP ratio; Chla- Chlorophyll *a* concentration; Phae- Phaeo-pigments concentration; PigD- pigments diversity index.

### 2.3.2.3. Sediment parameters

Most upstream samples were classified as silty sandy mud (C-III). Samples collected in March and May 2002 corresponded to very silty sandy mud (C-II) and in January 2002 sediment presented a greater percentage of sand and therefore was included in the very silty sand category (B-I). All samples from the downstream station were considered as sand (Figure 2.21).

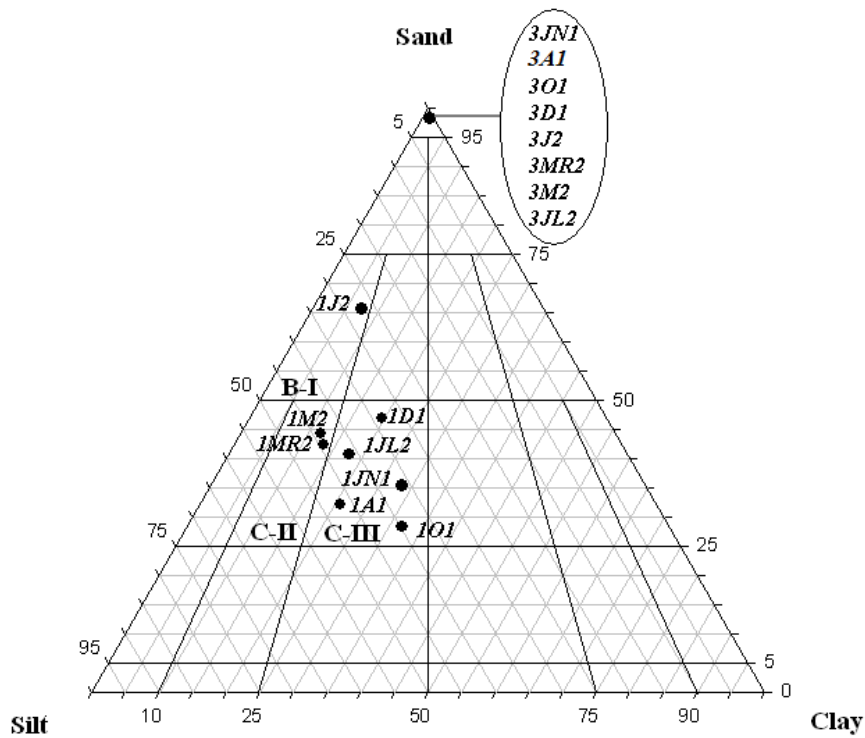


Figure 2.21 - Ternary diagram for textural classification of sediments from Salgados coastal lagoon, based on sand, silt and clay ratios (adapted from Flemming, 2000). *Station codes*: First character corresponds to the sampling station (1- upstream; 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001; 2- 2002). *Classification codes*: B-I - Very silty sand; C-II - Very silty sandy mud; C-III – Silty sandy mud.

In the upstream station, sediment presented greater diversity of grain size fractions than the sediment from the downstream station (Figure 2.22). Silt (27.1 % - 47.1 %) and clay (7.3 % - 31.9 %) were the fractions with higher percentages. Each sand fraction represented a small proportion of the sediment, nevertheless total sand ranged from 28.3 % to 65.6 %. Sediment from the downstream station was mainly composed by medium sand (40.4 % - 61.2 %) and coarse sand (25.5 % - 48.9 %).

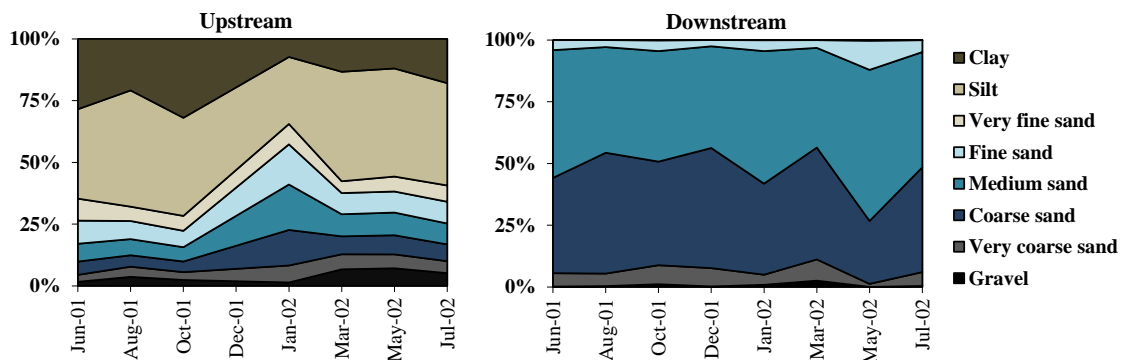


Figure 2.22 - Seasonal variation of individual grain size fractions in the two sampling stations from Salgados coastal lagoon.

Water content in the sediment ranged from 20.96 % (upstream: Jul-02) to 51.23 % (upstream: Oct-01) and till October 2001, highest values occurred upstream (Figure 2.23).

The minimum value of organic matter content (0.60 %) was registered downstream during March 2002 and the maximum value (12.95 %) was observed upstream in August 2001. The upstream station presented higher values and greater variation along time than the downstream station.

Chlorophyll *a* concentration in the sediment reached the minimum (downstream: 0.55  $\mu\text{g g}^{-1}$ ) and the maximum values (upstream: 55.75  $\mu\text{g g}^{-1}$ ) in October 2001. These concentrations were quite distinct from the ones determined during all other sampling periods. A similar situation was noticed with phaeo-pigments concentration, which also presented the minimum (downstream: 0.562  $\mu\text{g g}^{-1}$ ) and maximum (upstream: 58.50  $\mu\text{g g}^{-1}$ ) values in October 2001. Chlorophyll *a* degradation index ranged from 12.70 % in March 2002 to 55.26 % in October 2001, downstream. During the studied period, the upstream station presented higher values than the downstream station, except in October and December 2001.

Pigment diversity oscillated between 2.20 bits in June 2001 (downstream) and 3.33 bits in December 2001 (upstream), with higher values prevailing in the upstream station, except in October 2001.

The upstream station presented higher mean values for all sediment parameters, but sand content (Table 2.12).

Statistical analyses revealed that, for most parameters differences between stations were significant (Appendix I.D). Water content and chlorophyll *a* concentration were the only parameters that did not present significant differences ( $p > 0.05$ ).

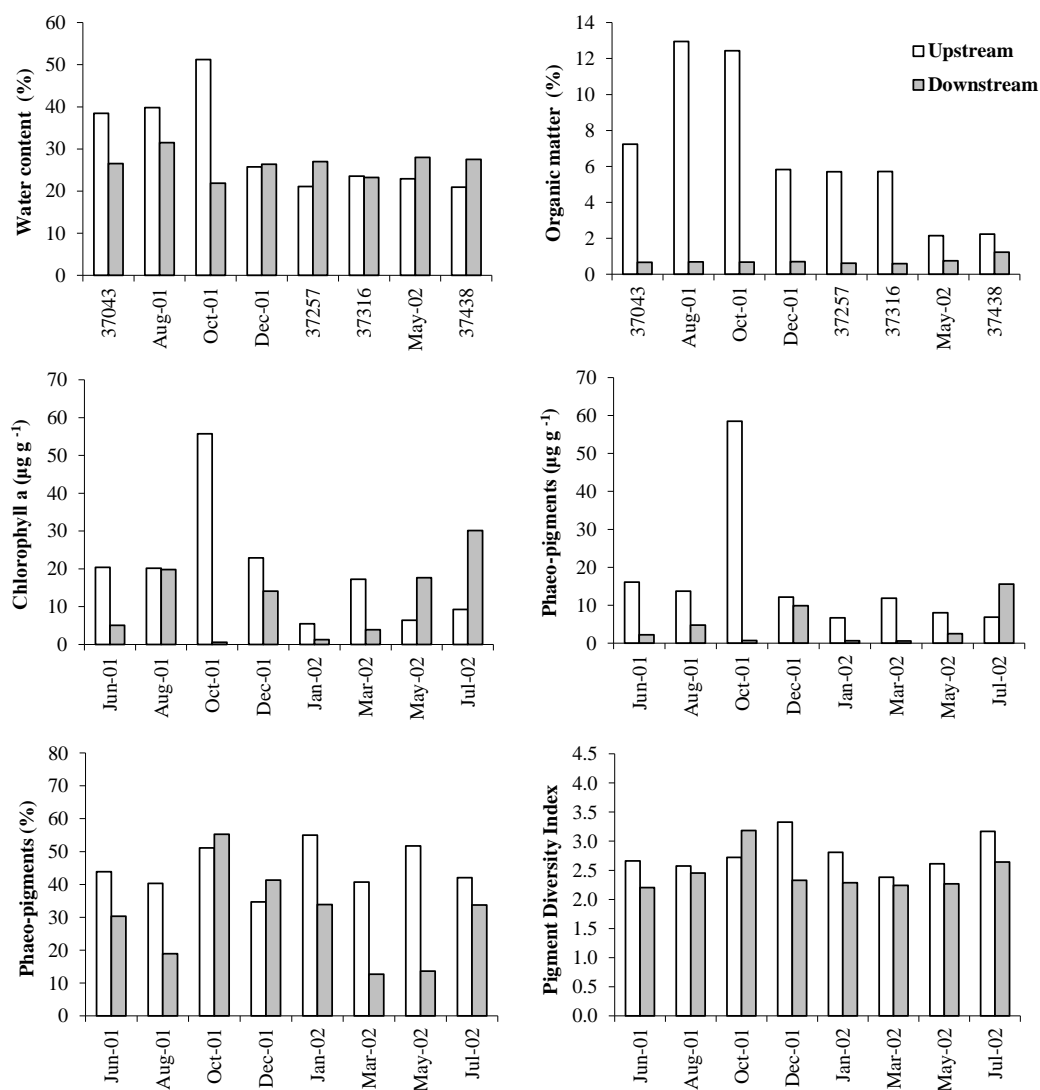


Figure 2.23 - Seasonal variation of water content, organic matter content, photosynthetic pigments concentration (chlorophyll *a* and phaeo-pigments), chlorophyll *a* degradation index (% phaeo-pigments) and Margalef's pigment diversity index in the sediment of the two sampling stations from Salgados lagoon.

Table 2.12 – Annual mean values and standard deviation of sediment parameters in Salgados sampling stations.

	<i>Upstream Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Clay content (%)	19.03±12.28	0.01±0.01
Silt content (%)	39.40±8.15	0.01±0.01
Sand content (%)	41.57±11.87	99.99±0.01
Water content (%)	30.48±11.26	26.53±2.94
Organic matter content (%)	6.78±4.06	0.74±0.20
Chlorophyll <i>a</i> concentration (µg g <sup>-1</sup> )	19.69±16.06	11.55±10.58
Phaeo-pigments concentration (µg g <sup>-1</sup> )	16.73±17.21	4.60±5.41
Chlorophyll <i>a</i> degradation index (%)	44.93±6.96	29.96±14.57
Pigment diversity index (bits)	2.78±0.32	2.45±0.33

The PCA ordination biplot (Figure 2.24) showed a separation of downstream samples (left side) from the upstream samples (right side), according to axis I. The first axis accounted for 55.1% of the total variance and the sediment parameters which contributed most for samples distribution along the axis were clay content (component loading = 0.934), organic matter content (component loading = 0.909), phaeo-pigment concentration (component loading = 0.848), silt content (component loading = 0.794), chlorophyll *a* concentration (component loading = 0.736) and water content (component loading = 0.720). The samples plotted in the right side of axis I (upstream station) had higher values of clay content, organic matter content, phaeo-pigment concentration, silt content, chlorophyll *a* concentration and water content, while the samples in the left side presented lower values for these same parameters.

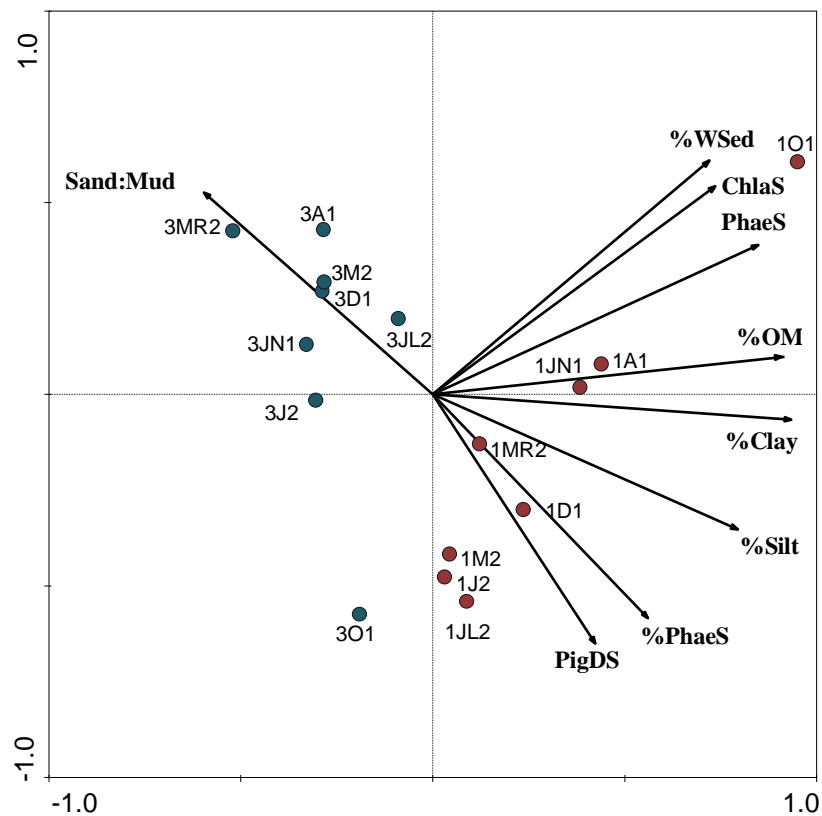


Figure 2.24 - Principal component analysis performed on sediment parameters from Salgados lagoon. Cumulative percentage variance explained by axes: I – 55.1%; I + II – 77.3%.

*Station codes:* First character corresponds to the sampling station (1 - upstream, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002). *Sediment parameters:* Sand:Mud – Sand mud ratio; %Clay – Clay content; %Silt – Silt content; %OM – Organic matter content; %WSed – Water content; ChlaS - Chlorophyll *a* concentration; PhaeS - Phaeo-pigments concentration; PigDS - Margalef's pigment diversity index; %PhaeS - Chlorophyll *a* degradation index.

The second axis explained 22.2 % of the total variance and pigments diversity index was the parameter with greater contribution to this axis (component loading = -0.650). Samples displayed in the bottom of the ordination diagram showed higher values of pigments diversity index (*e.g.* upstream July 2002 and downstream October 2001) and those samples in the upper side had lower values (*e.g.* downstream March 2002 and August 2001). Some of the parameters were highly and positively correlated being represented close to each other, namely water content and chlorophyll *a* concentration. Those parameters pointing in opposite directions were negatively correlated, as it happened with Sand: Mud ratio and chlorophyll *a* degradation index or Sand: Mud ratio and pigments diversity index.

#### **2.3.2.4. Relations between environmental parameters**

All sediment parameters were significantly correlated with at least one of the water parameters (Table 2.13). Positive correlations were found between clay content, total solids in suspension and pigments diversity in water. Silt content was also positively correlated with pigments diversity in water, while sand content presented a negative correlation. Thereby, samples with higher pigments diversity in water corresponded to the ones with greater clay content, silt content and lower sand content (upstream), and *vice-versa* (downstream). Moreover, the samples richer in clay presented higher concentrations of total solids in suspension (upstream) and *vice-versa* (downstream). Organic matter content in the sediment and total solids in suspension had a positive correlation and so, samples with greater content of organic matter matched with higher concentration of total solids in suspension (upstream) and *vice-versa* (downstream). Water content in the sediment was positively correlated to water temperature, meaning that the samples with higher water content in the sediment (upstream) tended to have higher values of temperature and *vice-versa* (downstream).

Some of these associations can be explained by the depth of sampling stations, the upstream station has a lower depth and is therefore more sensible to temperature variation and concentration of solids in suspension, while the downstream station is near the deepest part of the lagoon, where these effects were less noticed.

Chlorophyll *a* and phaeo-pigments concentrations in the sediment showed a negative linear association with salinity and nitrates concentration in water. Samples with higher

concentrations of chlorophyll *a* and phaeo-pigments in the sediment (upstream) were the ones with lower values of salinity and nitrates concentration in water, and *vice-versa* (downstream). A negative correlation was also found between chlorophyll *a* concentrations and rainfall, meaning that lower values of chlorophyll *a* in the sediment were observed in periods of greater rainfall and *vice-versa*. Chlorophyll *a* degradation index and pigments diversity index in the sediment were positively correlated with pigments diversity in water, thus sediments with higher chlorophyll *a* degradation index and pigments diversity index were associated to higher indexes of pigments diversity in water (upstream) and *vice-versa* (downstream).

Table 2.13 - Significant correlations between water and sediment parameters from Salgados lagoon. \* Correlation significant at the 0.05 level; \*\* Correlation significant at the 0.01 level.

<i>Sediment parameters</i>	<i>Water parameters</i>	<i>Results</i>
Clay content (%Clay)	Total Solids in Suspension (TSS)	Rho = 0.606; <i>p</i> = 0.013 *
	Pigment diversity index (PigDW)	Rho = 0.546; <i>p</i> = 0.029 *
Silt content (%Silt)	Pigment diversity index (PigDW)	Rho = 0.694; <i>p</i> = 0.003 **
Sand content (%Sand)	Pigment diversity index (PigDW)	Rho = -0.682; <i>p</i> = 0.004 **
Water content (%WSed)	Temperature (Temp)	Rho = 0.547; <i>p</i> = 0.028 *
Organic matter content (%OM)	Total Solids in Suspension (TSS)	Rho = 0.547; <i>p</i> = 0.028 *
Chlorophyll <i>a</i> concentration (ChlaS)	Rain	Rho = -0.527; <i>p</i> = 0.036 *
	Salinity (Sali)	R = -0.503; <i>p</i> = 0.047 *
	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.640; <i>p</i> = 0.008 **
Phaeo-pigments concentration (PhaeS)	Salinity (Sali)	R = -0.548; <i>p</i> = 0.028 *
	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.617; <i>p</i> = 0.011 *
Chlorophyll <i>a</i> degradation index (%PhaeS)	Pigment diversity index (PigDW)	Rho = 0.724; <i>p</i> = 0.002 **
Pigment diversity index (PigDS)	Pigment diversity index (PigDW)	Rho = 0.546; <i>p</i> = 0.029 *

Figure 2.25 presents the ordination of samples in a PCA biplot, according to the most relevant water and sediment parameters determined in previous PCA, performed separately for water parameters (Figure 2.20) and for sediment parameters (Figure 2.24). Only data from the upstream and downstream stations were considered, as there was no information concerning the sediment parameters in the intermediate station.

The first axis accounted for 35.9% of the total variance and mainly separated samples according to sediment parameters. Samples plotted in the left side of axis I (downstream stations) were characterized by lower values of clay content (component loading = 0.932), organic matter content (component loading = 0.851), silt content (component loading = 0.827) and phaeo-pigment concentration (component loading = 0.771), while the samples in the right side (upstream station) presented higher values for these same parameters. Nitrates concentration contributed to the first axis too

(component loading = -0.599) but it was less relevant than the other parameters, being also associated to a third axis (component loading = 0.603).

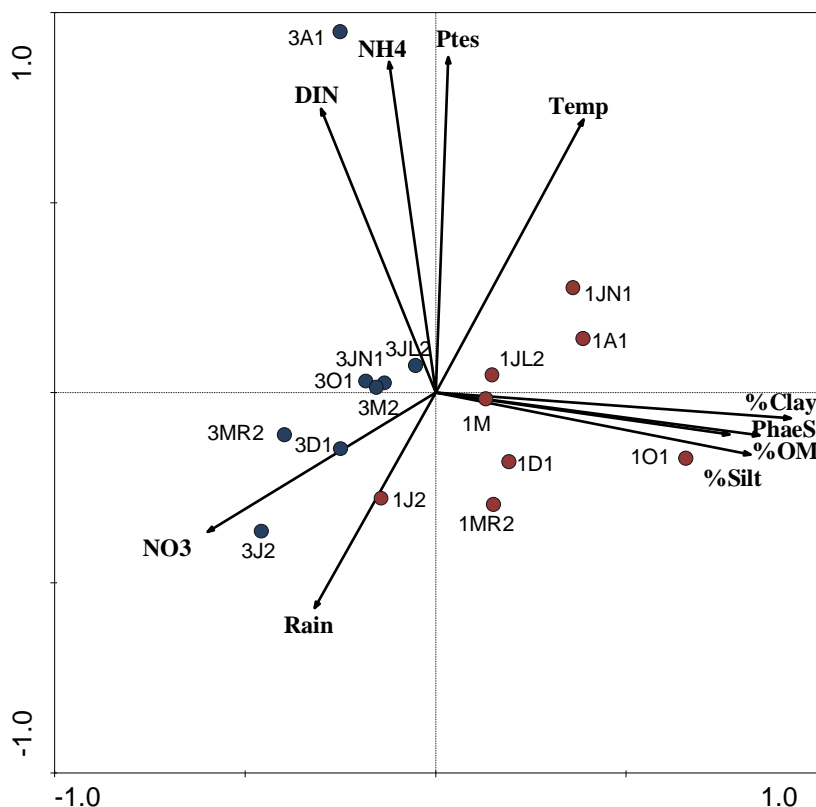


Figure 2.25 - Principal component analysis performed on environmental parameters from Salgados coastal lagoon. Cumulative percentage variance explained by axes: I – 35.9%; I + II – 67.1%.

*Station codes:* First character corresponds to the sampling station (1 - upstream, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002).

*Hydrological and water variables:* Rain- cumulative rainfall in 10 days previous to sampling; Temp – Temperature; NO<sub>3</sub>- nitrates concentration; NH<sub>4</sub>- ammonia concentration; DIN- total dissolved inorganic nitrogen concentration; Ptes- orthophosphates concentration. *Sediment variables:* %Silt – Percentage of silt; %Clay- Percentage of clay; %OM – Percentage of organic matter; PhaeS - Phaeo-pigments concentration.

The second axis explained 31.2 % of the total variance and the parameters which contributed most for samples distribution along the axis were water parameters, namely orthophosphates concentration (component loading = 0.883), ammonia concentration (component loading = 0.870), total dissolved inorganic nitrogen (component loading = 0.747) and temperature (component loading = 0.719). Samples displayed in the upper side of the ordination diagram showed higher values of these parameters (e.g. downstream August 2001) and samples in the bottom part of the diagram (e.g. downstream January 2002 and upstream March 2002) showed lower values of orthophosphates, ammonia, total dissolved inorganic nitrogen and temperature.

Cumulative rainfall had a lower contribution to samples ordination according to axis II (component loading = -0.567), as it was also associated to a third axis (component loading = 0.614).

The comparison of chlorophyll *a* in the water ( $\text{mg m}^{-2}$ ) and chlorophyll *a* in the sediment ( $\text{mg m}^{-2}$ ) upstream and downstream showed that most of the time, phytoplankton biomass in the water column was higher than microphytobenthos biomass (Figure 2.26). Just in a few samplings the opposite situation was registered, namely in June and October 2001 in the upstream station. Downstream, chlorophyll *a* in the water and chlorophyll *a* in the sediment presented closer values and similar percentages in June and August 2001, just as in March 2002.

Chlorophyll *a* in the upstream station sediment varied between  $15.43 \text{ mg m}^{-2}$  (34.3%) in January 2002 and  $157.22 \text{ mg m}^{-2}$  (87.6%) in October 2001. In July 2002, chlorophyll *a* in the sediment accounted the lowest percentage of total chlorophyll *a* (4.7%) and the maximum of 87.6% was achieved in October 2001. The annual mean value of  $55.99 \text{ mg m}^{-2}$  represented 30.3% of total chlorophyll *a* in the upstream station.

The amount of chlorophyll *a* in the water collected upstream ranged from  $22.34 \text{ mg m}^{-2}$  (12.4%) in October 2001 to  $661.66 \text{ mg m}^{-2}$  (93.2%) in March 2002 and the annual mean value was  $259.61 \text{ mg m}^{-2}$  (69.7% of total chlorophyll *a* in the upstream station).

The downstream station presented values of chlorophyll *a* in the sediment that went from  $1.57 \text{ mg m}^{-2}$  (9.3%) in October 2001 to  $86.32 \text{ mg m}^{-2}$  (22.6%) in July 2002 and the annual mean of  $32.67 \text{ mg m}^{-2}$  accounted 30.3% of total chlorophyll *a* downstream.

The lowest value of chlorophyll *a* in the water downstream was determined in March 2002 ( $13.23 \text{ mg m}^{-2}$ ; 54.3%) and the highest value was registered in July 2002 ( $295.31 \text{ mg m}^{-2}$ ; 77.4%). Throughout the studied period, chlorophyll *a* in the water accounted from 46.3% (August 2001) to 94.0% (January 2002) of total chlorophyll *a* downstream. In June 2001, August 2001 and March 2002, chlorophyll *a* in the water and chlorophyll *a* in the sediment contributed with a similar percentage to the total chlorophyll determined for each of these months.

The annual mean value of chlorophyll *a* in the water  $85.67 \text{ mg m}^{-2}$  represented 69.7% of the total chlorophyll *a* downstream.

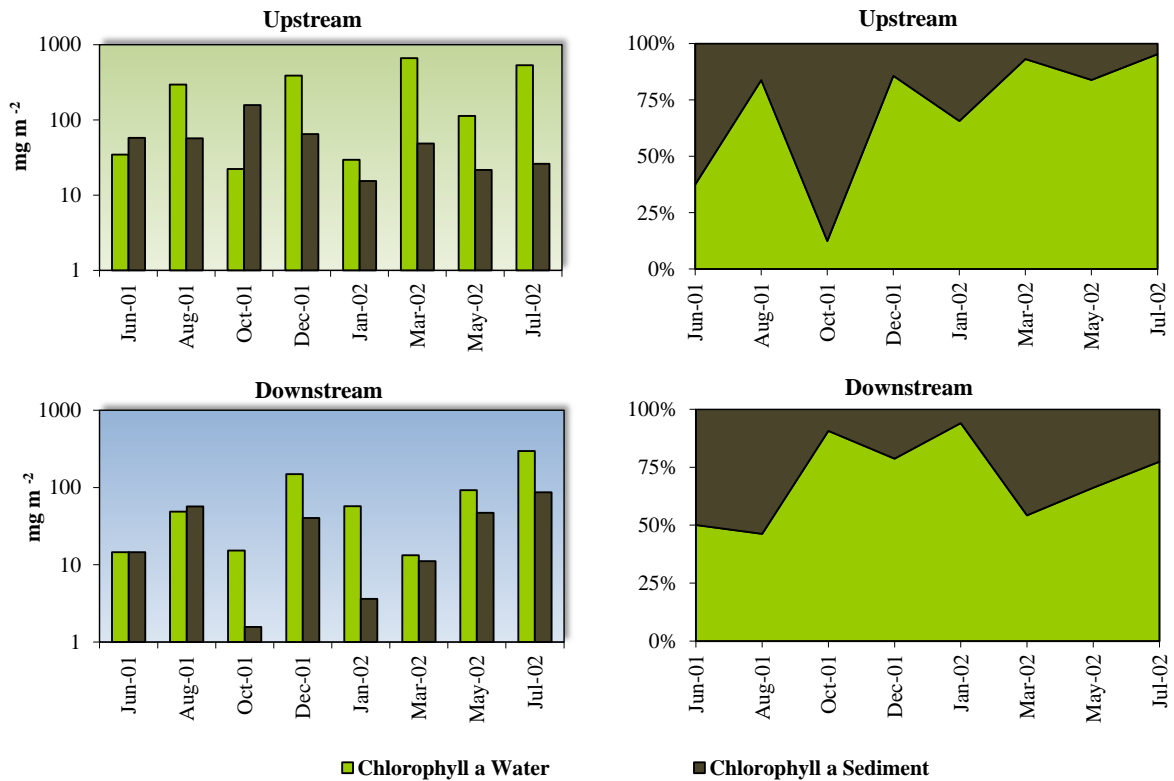


Figure 2.26 - Seasonal variation of chlorophyll *a* in the sediment, chlorophyll *a* in the water and percentage of each relative to total chlorophyll *a* in Salgados sampling stations.

### 2.3.2.5. Comparison of environmental parameters during isolation and connection of the lagoon with the sea

Just as it happened with Foz de Almagem, in January 2002 Salgados sampling coincided with a period when the lagoon was opened and with direct influence from the sea. In December 2001 the lagoon was isolated and so data these two periods were compared. Two previous openings occurred, one in the middle of October (before sampling) and another between December and January samplings.

In all stations there was a decrease in pH, total solids in suspension, dissolved oxygen and chlorophyll *a* concentrations and it was noticed an increase in salinity, nitrates, nitrites, total dissolved inorganic nitrogen, N:P ratio, phaeo-pigments concentration and pigment diversity (Table 2.14).

Temperature decreased in the upstream station and increased in the other stations, while orthophosphates and total phosphorus concentrations showed the opposite variation. In the upstream and intermediate stations was registered an increase in ammonia concentrations, whereas downstream concentrations decreased.

Table 2.14 – Water and sediment parameters in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.

	<i>Upstream station</i>		<i>Intermediate station</i>		<i>Downstream station</i>	
	<b>Jan-02</b>	<b>Variation</b>	<b>Jan-02</b>	<b>Variation</b>	<b>Jan-02</b>	<b>Variation</b>
<b>Water parameters</b>						
Temperature (°C)	15.89	<b>-1.17</b>	16.53	<b>1.27</b>	16.41	<b>1.71</b>
Salinity (‰)	9.91	<b>5.50</b>	13.59	<b>9.06</b>	23.10	<b>17.92</b>
pH	8.75	<b>-1.23</b>	8.24	<b>-0.85</b>	8.78	<b>-0.48</b>
Total Solids in Suspension (mg L <sup>-1</sup> )	37.24	<b>-132.76</b>	61.50	<b>-46.00</b>	39.33	<b>-16.67</b>
Dissolved oxygen (mg L <sup>-1</sup> )	13.71	<b>-8.49</b>	9.91	<b>-11.64</b>	11.57	<b>-8.22</b>
Nitrates concentration (µM N)	45.00	<b>43.71</b>	118.87	<b>110.79</b>	75.81	<b>45.32</b>
Nitrites concentration (µM N)	14.61	<b>13.78</b>	18.00	<b>15.20</b>	12.20	<b>6.20</b>
Ammonia concentration (µM N)	41.67	<b>32.78</b>	34.06	<b>3.94</b>	0.94	<b>-10.22</b>
Total diss. inorg. nitrogen (µM N)	101.28	<b>90.27</b>	170.93	<b>129.93</b>	88.95	<b>41.30</b>
Orthophosphates concentration (µM P)	37.26	<b>11.10</b>	22.42	<b>-8.84</b>	20.63	<b>-19.89</b>
Total Phosphorus concentration (µM P)	41.29	<b>15.08</b>	23.35	<b>-13.42</b>	21.32	<b>-26.10</b>
N:P ratio	2.72	<b>2.30</b>	7.62	<b>6.31</b>	4.31	<b>3.14</b>
Chlorophyll <i>a</i> concentration (µg L <sup>-1</sup> )	29.50	<b>-358.36</b>	53.85	<b>-101.27</b>	57.02	<b>-92.00</b>
Phaeo-pigments concentration (µg L <sup>-1</sup> )	7.94	<b>1.79</b>	9.15	<b>3.58</b>	6.25	<b>3.29</b>
Pigment diversity index	2.12	<b>0.22</b>	2.19	<b>0.44</b>	1.98	<b>0.11</b>
<b>Sediment parameters</b>						
Clay content (%)	7.31	<b>5.03</b>			0.02	<b>0.02</b>
Silt content (%)	27.13	<b>-21.32</b>			0.01	<b>0.01</b>
Sand content (%)	65.56	<b>16.28</b>			99.98	<b>-0.02</b>
Organic matter content (%)	5.70	<b>-0.12</b>			0.62	<b>-0.08</b>
Chlorophyll <i>a</i> concentration (µg g <sup>-1</sup> )	5.47	<b>-17.42</b>			1.27	<b>-12.82</b>
Phaeo-pigments concentration (µg g <sup>-1</sup> )	6.67	<b>-5.47</b>			0.65	<b>-9.23</b>
Chlorophyll <i>a</i> degradation index (%)	54.99	<b>20.33</b>			33.89	<b>-7.41</b>
Pigment diversity index	2.81	<b>-0.52</b>			2.29	<b>-0.04</b>

Major variations of water parameters were determined for chlorophyll *a*, total solids in suspension, total dissolved inorganic nitrogen, nitrates, ammonia, total phosphorus and orthophosphates concentrations.

The upstream station presented the lowest variations for temperature (-1,23°C), salinity (5.50 ‰), nitrates concentration (43.71 µM N), N:P ratio (2.30), phaeo-pigments concentration (1.79 µg L<sup>-1</sup>) and also the greatest variations of pH (-1.23), total solids in suspension (-132.76 mg L<sup>-1</sup>), ammonia (32.78 µM N) and chlorophyll *a* (-358.36 µg L<sup>-1</sup>).

The smallest variations of ammonia (3.94 µM N) and orthophosphates (-8.84 µM P) were determined in the intermediate station, just as the major variations of dissolved oxygen concentration (-11.64 mg L<sup>-1</sup>), nitrates (110.79 µM N), total dissolved inorganic nitrogen (129.93 µM N), N:P ratio (6.31), phaeo-pigments concentration (3.58 µg L<sup>-1</sup>) and pigment diversity (0.44).

Downstream was the station with the minimum variation in pH (-0.48), total solids in suspension (-16.67 mg L<sup>-1</sup>), dissolved oxygen concentration (-8.22 mg L<sup>-1</sup>), total dissolved inorganic nitrogen (41.30 µM N), chlorophyll *a* (-92.00 µg L<sup>-1</sup>) and pigment diversity (0.11). In this station it was observed the greatest variation in temperature (1.71°C), salinity (17.92 ‰) and orthophosphates concentration (-19.89 µM P).

Regarding sediment parameters, in the upstream and downstream stations there was a decrease in organic matter content, chlorophyll *a*, phaeo-pigments concentrations and pigments diversity. Chlorophyll *a* degradation index increased upstream and decreased downstream.

Silt content, chlorophyll *a* degradation index, chlorophyll *a* concentration and sand content were the parameters that presented greatest variations.

The upstream station presented the lowest variation of phaeo-pigments concentration (-5.47 µg g<sup>-1</sup>) and the greatest variations for silt content (-21.32 %), chlorophyll *a* degradation index (20.33 %), chlorophyll *a* (-17.42 µg g<sup>-1</sup>), sand content (16.28 %), clay content (5.03 %), pigment diversity (-0.52) and organic matter content (-0.12%). Downstream, the sediment parameters variation was smaller, except for phaeo-pigments concentration (-9.23 µg g<sup>-1</sup>).

#### **2.3.2.6. Trophic state and water quality**

Just as in Foz de Almargem coastal lagoon, the values of Carlsons` trophic state index determined with chlorophyll *a*, TSI (CHL), were lower than the values obtained with total phosphorus concentrations, TSI (TP) (Figure 2.27).

TSI (CHL) was minimum in March 2002 (Downstream: 55.93), June 2001 (Downstream: 56.89) and October 2001 (Downstream: 57.34), corresponding to eutrophic conditions (50 < TSI < 60). The maximum TSI (CHL) values were reached in March 2002 (Upstream: 94.31), July 2002 (Upstream: 92.20) and December 2001 (Upstream: 89.07), corresponding to hypereutrophic systems (TSI > 70). Most samples presented values that indicate hypereutrophy (54%) and eutrophy-hypereutrophy (29%). Besides chlorophyll *a* concentration, TSI (CHL) values were positively correlated with pH ( $r = 0.540$ ;  $p = 0.006$ ), dissolved oxygen ( $r = 0.511$ ;  $p = 0.011$ ) and negatively correlated to salinity ( $r = -0.438$ ;  $p = 0.032$ ). The annual mean TSI(CHL) values

determined for each station, showed a moderate decrease according to the distance from the sea (upstream:  $78.64 \pm 13.66$ ; intermediate:  $73.55 \pm 11.33$ ; downstream:  $68.78 \pm 11.38$ ), including the upstream and intermediate stations in the category of hypereutrophy, while the downstream station was classified as eutrophic-hypereutrophic.

All TSI (TP) values were near or above 100, suggesting that the lagoon presented hypereutrophic conditions (TSI > 70). The lowest values were calculated in March 2002 (Upstream: 97.21) and January 2002 (Downstream: 97.79; Intermediate: 99.10). The highest values were found in June 2001 (Intermediate: 126.97; Upstream: 126.08) and August 2001 (Intermediate: 126.74). Positive correlations were found between TSI (TP) values and orthophosphates ( $r = 0.986$ ;  $p < 0.001$ ), temperature ( $r = 0.677$ ;  $p < 0.001$ ) and ammonia ( $r = 0.443$ ;  $p = 0.030$ ), while negative correlations were determined with pH ( $r = -0.647$ ;  $p = 0.001$ ), dissolved oxygen ( $r = -0.534$ ;  $p = 0.007$ ) and cumulative rainfall ( $\rho = -0.530$ ;  $p = 0.008$ ). The intermediate station presented the greatest mean annual TSI (TP) value ( $114.73 \pm 9.70$ ), followed by the downstream ( $110.35 \pm 6.92$ ) and the upstream stations ( $109.00 \pm 9.06$ ).

The trophic state index TRIX ranged from 7.24 (Upstream, June 2001) to 10.00 (Intermediate, August 2001). According to the criteria defined by Penna *et al.* (2004) the water presented a poor quality, characteristic of a system highly productive and with the greatest trophic level (TRIX > 6). TRIX was correlated to chlorophyll *a* ( $r = 0.678$ ;  $p < 0.001$ ), dissolved oxygen ( $r = 0.444$ ;  $p = 0.030$ ), DIN ( $r = 0.409$ ;  $p = 0.047$ ) and also with the water level in the lagoon ( $\rho = -0.528$ ;  $p = 0.008$ ), pH ( $r = 0.471$ ;  $p = 0.020$ ), N: P ratio ( $\rho = 0.464$ ;  $p = 0.022$ ), cumulative rain ( $\rho = -0.454$ ;  $p = 0.026$ ) and pigment diversity ( $r = -0.444$ ;  $p = 0.033$ ).

The mean annual TRIX value in the intermediate station ( $8.96 \pm 0.74$ ) was slightly superior to the value from the upstream ( $8.89 \pm 0.86$ ) and the downstream ( $8.67 \pm 0.61$ ) stations.

When the lagoon was closed (June-December 2001; March-July 2002), the trophic state and water quality mean values of the stations were similar (TRIX) or slightly higher (TSI-CHL; TSI-TP) than the values obtained with all data from the studied period. Thereby, the trophic state and water quality classification of the stations was the same as the classification determined with all data (Table 2.15).

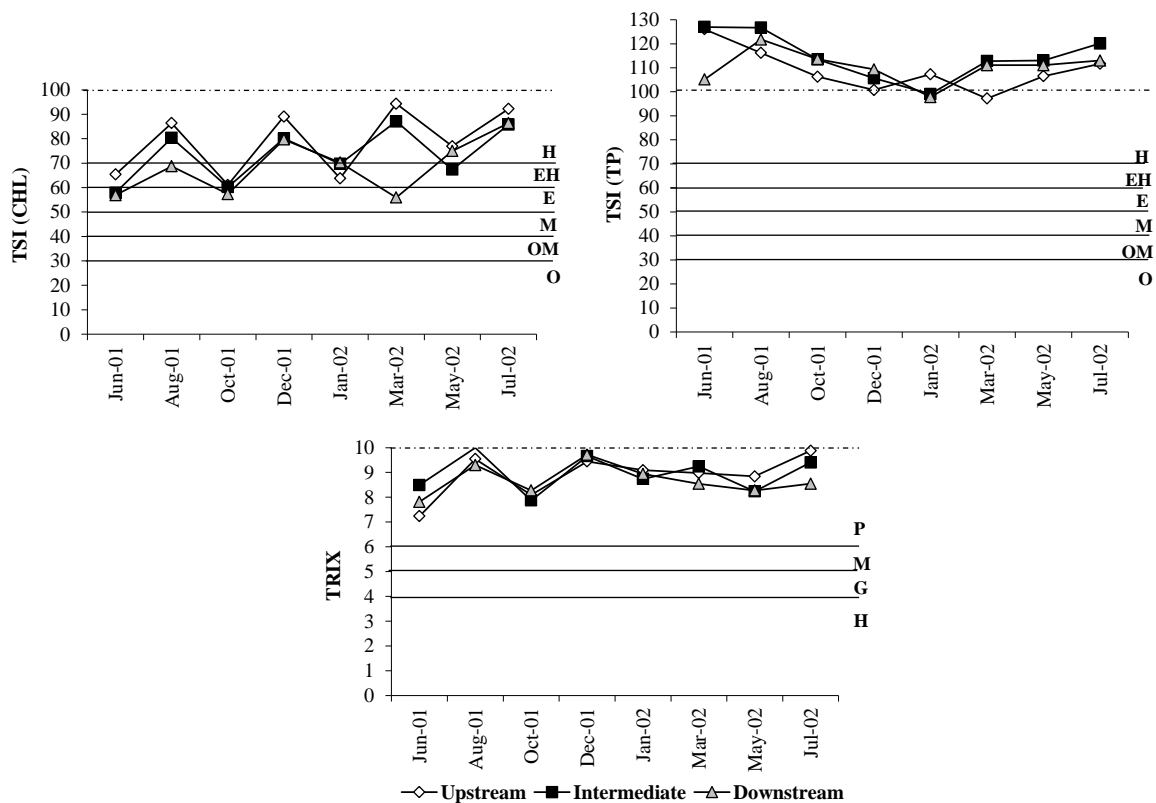


Figure 2.27 – Seasonal variation of trophic state and water quality indexes in Salgados coastal lagoon: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX. TSI: O- Oligotrophic; OM- Oligomesotrophic; M- Mesotrophic; E- Eutrophic; EH- Eutrophic to Hypereutrophic; H- Hypereutrophic. TRIX: H- High water quality; G- Good water quality; M- Mediocre water quality; P- Poor water quality. Dashed lines represent the maximum theoretical value for each index scale.

Table 2.15 – Trophic state and water quality indexes mean values, standard deviation and classification in Salgados sampling stations, determined during all studied period (June 2001- July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
TSI (CHL): All data	78.64±13.66	73.55±11.33	68.78±11.38
	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>	<b>Eutrophic-Hypereutrophic</b>
TSI (CHL): Closed period	80.76±13.26	74.10±12.12	68.57±12.28
	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>	<b>Eutrophic-Hypereutrophic</b>
TSI (TP): All data	109.00±9.06	114.73±9.70	110.35±6.92
	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>
TSI (TP): Closed period	109.24±9.76	116.96±7.95	112.14±5.09
	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>	<b>Hypereutrophic</b>
TRIX: All data	8.89±0.86	8.96±0.74	8.67±0.61
	<b>Poor water quality</b>	<b>Poor water quality</b>	<b>Poor water quality</b>
TRIX: Closed period	8.86±0.92	8.98±0.79	8.64±0.65
	<b>Poor water quality</b>	<b>Poor water quality</b>	<b>Poor water quality</b>

The comparison of data from December 2001 (lagoon closed) and January 2002 (lagoon opened) showed that these indexes had a negative variation in all stations, except TSI (TP) in the upstream station ( $\Delta = 6.58$ ). TSI (CHL) presented the lowest variation downstream ( $\Delta = -9.42$ ) and the highest variation in the upstream station ( $\Delta = -25.27$ ). According to TSI (CHL) classification, all stations decreased its trophic state from hypereutrophic (TSI CHL<sub>upstream</sub> = 89.07; TSI CHL<sub>intermediate</sub> = 80.08; TSI CHL<sub>downstream</sub> = 79.69) to eutrophic-hypereutrophic (TSI CHL<sub>upstream</sub> = 63.80; TSI CHL<sub>intermediate</sub> = 69.71; TSI CHL<sub>downstream</sub> = 70.27).

The smallest variation in TSI (TP) was determined at the upstream station ( $\Delta = 6.58$ ) and the greatest variation happened downstream ( $\Delta = -11.53$ ). Despite values variation, stations maintained the classification of hypereutrophic (TSI TP<sub>upstream</sub> = 107.32; TSI TP<sub>intermediate</sub> = 99.10; TSI TP<sub>downstream</sub> = 97.79), as when the lagoon was isolated from the sea (TSI TP<sub>upstream</sub> = 100.74; TSI TP<sub>intermediate</sub> = 105.65; TSI TP<sub>downstream</sub> = 109.32).

TRIX showed the smallest variation upstream ( $\Delta = -0.35$ ) and the highest variation in the intermediate station ( $\Delta = -0.92$ ). Although TRIX values decreased in all stations, the water quality was still classified as poor (TRIX<sub>upstream</sub> = 9.09; TRIX<sub>intermediate</sub> = 8.74; TRIX<sub>downstream</sub> = 8.94), the same way it was in December 2001 (TRIX<sub>upstream</sub> = 9.44; TRIX<sub>intermediate</sub> = 9.66; TRIX<sub>downstream</sub> = 9.72).

The 90<sup>th</sup> percentile values of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ) in Salgados stations were much higher than the reference values defined by Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012) for coastal lagoons (Table 2.16).

When the lagoon was closed, all stations were classified with a bad water quality, as the 90<sup>th</sup> percentile values of chlorophyll *a* were superior to  $101.3 \mu\text{g L}^{-1}$ , the boundary value to distinguish Bad from Poor water quality (Pereira Coutinho *et al.*, 2012). During this period, chlorophyll *a* was positively correlated with dissolved oxygen concentrations ( $r = 0.694$ ;  $p < 0.001$ ) and with pH ( $r = 0.498$ ;  $p = 0.021$ ), but no significant correlations or linear regressions were determined with nutrients concentrations. Just as it happened in Foz de Almagem, data collected in Salgados lagoon were not enough to calculate the 90<sup>th</sup> percentile values of chlorophyll *a* when the lagoon was opened (January 2002) and determine its water quality.

All the results obtained during the phytoplankton growing seasons of 2001 (June to October), 2002 (March to July) and 2001-2002 (June to October 2001; March to July 2002) were higher than  $27 \mu\text{g L}^{-1}$ , which corresponds to a bad water quality in all stations, according to Brito *et al.* (2012) classification criteria.

Table 2.16 – Water quality in Salgados sampling stations, based on the 90<sup>th</sup> percentile of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ), adapted from Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012).

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
<b>Pereira Coutinho <i>et al.</i> (2012): Semi-enclosed lagoons</b>	<i>Ref. value closed period (20 <math>\mu\text{g L}^{-1}</math>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> during closed period	584.67 $\mu\text{g L}^{-1}$	293,07 $\mu\text{g L}^{-1}$	207,54 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality during closed period</b>	<b>Bad</b> (> 101,3 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 101,3 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 101,3 $\mu\text{g L}^{-1}$ )
<b>Brito <i>et al.</i> (2012): Coastal water lagoons</b>	<i>Ref. value growing season (February-October: 5,3 <math>\mu\text{g L}^{-1}</math>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> (Jun-Oct 01)	242.88 $\mu\text{g L}^{-1}$	131.26 $\mu\text{g L}^{-1}$	42.12 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (Mar-Jul 02)	635.99 $\mu\text{g L}^{-1}$	308.75 $\mu\text{g L}^{-1}$	254.67 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (Jun-Oct 01; Mar-Jul 02)	597.51 $\mu\text{g L}^{-1}$	296.99 $\mu\text{g L}^{-1}$	193.69 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )	<b>Bad</b> (> 27 $\mu\text{g L}^{-1}$ )

### 2.3.3. Comparison of the two coastal lagoons

#### 2.3.3.1. Hydrological aspects

In both lagoons, the raining season started in September 2001 and December 2001 was the month with maximum rainfall, registering 276.7 mm in Foz de Almargem and 183.5 mm in Salgados (Figure 2.28).

In Foz de Almargem, the total rainfall determined for the studied period (June 2001-July 2002) was greater (780.3 mm) than in Salgados (688.0 mm) and also, the period of raining season lasted longer, until June 2002, while in Salgados it ended in May 2002. In Salgados lagoon, wastewater discharges also contributed to freshwater inputs, but no data were available for the studied period.

The water level in the lagoons increased during the raining season, till the sand barrier was artificially or naturally destroyed and the water from the lagoons flowed into the sea. In Foz de Almargem there were four openings of the lagoon, three of which were induced by local fishermen (October 2001, March 2002 and May 2002). During December 2001, a natural opening occurred and the lagoon stayed in connection with the sea until mid-January.

The artificial openings from October 2001 and May 2002, lasted nine and 10 days respectively. The natural opening from December 2001 (28 days: 17 in December and 11 in January) and the artificial opening from March 2002 (16 days: 6 in March and 10 in April) lasted for longer periods of time.

Salgados lagoon was artificially opened three times (October 2001, December 2001 and February 2002) by the regional environmental services (former DRAOT). In October and December 2001, the lagoon stayed opened for six days in each occasion and in February 2002, it only lasted two days. Also in December, after the artificial opening, the lagoon became naturally connected to the sea for nine days and in January 2002, the lagoon reopened naturally for more nine days, after a period of strong rainfall and sea storms.

Although Salgados lagoon was opened five times and in Foz de Almargem there were four openings, the total number of days Foz de Almargem lagoon was in connection with the sea (63 days) almost doubled the period of connection in Salgados (32 days). Both lagoons maintained the connection with the sea for longer time after natural openings, than after artificial interventions.

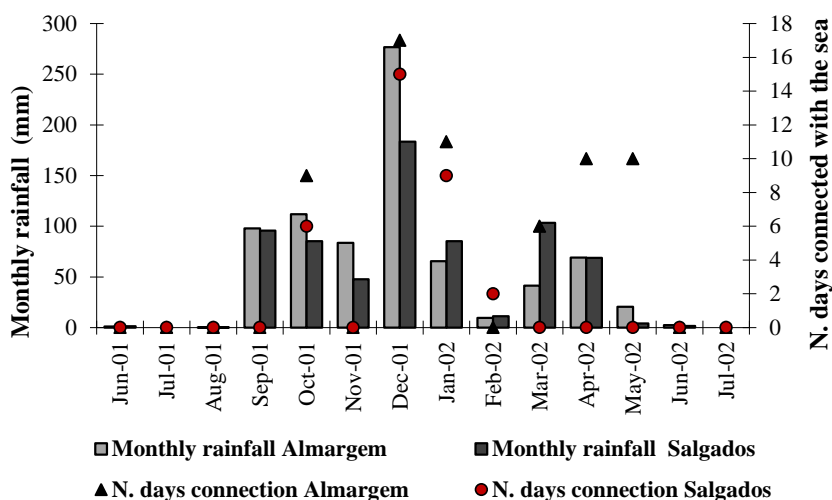


Figure 2.28 - Variation of monthly rainfall registered in Foz de Almargem (Loulé meteorological station) and Salgados (Algoz meteorological station) and duration of the connection between the lagoons and the sea (number of days *per* month).

### 2.3.3.2. Water parameters

Water temperature seasonal variation was similar in the lagoons, increasing during spring and summer and decreasing through autumn and winter, according to atmospheric temperatures (Figure 2.29).

Seasonal evolution of salinity in Foz de Almargem and Salgados presented some differences. The lowest mean values in both lagoons were registered in May 2002 (Foz de Almargem: 6.03‰; Salgados: 4.06‰) but the highest mean value in Foz de Almargem was determined in August 2001 (26.24‰), while in Salgados the highest salinities (15.53‰) occurred in June 2001 and January 2002. Based on the intervals defined by the Water Framework Directive (C.E.C., 2000), the monthly mean salinities in Foz de Almargem corresponded to a mesohaline system (5-18 ‰) most of the time, except in August and October 2001, when the values fitted in the polyhaline category (18-30 ‰). Monthly mean salinities in Salgados lagoon were also within the range of mesohaline systems during most of the studied period, but in October 2001 and May 2002 the mean values were lower corresponding to oligohaline systems (0.5-5 ‰).

In Foz de Almargem, salinity increase in the summer months was mainly due to evaporation when the water level was low. During the rest of the year, salinity variation depended on whether the lagoon was in connection to the sea or confined by the sand barrier and also on the freshwater *input* from a small river. In Salgados lagoon, besides these factors there was also the freshwater *input* from the wastewater treatment plant influencing salinity variation. This lagoon presented a lower influence from the sea, not just because of higher freshwater discharges but also due to the fact that the barrier opening frequency and the duration of connection with the sea was lower than in Foz de Almargem. Accordingly to the Water Framework Directive (C.E.C., 2000), the annual mean salinities in Foz de Almargem (13.09 ‰) and in Salgados lagoon (8.16 ‰) corresponded to mesohaline systems.

Foz de Almargem and Salgados lagoons presented different patterns of pH variation. The highest mean value in Foz de Almargem occurred in August 2001 (8.95) and in Salgados it was in December 2001 (9.44).

Total solids in suspension had greater mean values and seasonal variation in Salgados, with the maximum value occurring in August 2001 (154.76 mg L<sup>-1</sup>). This value might have been caused by higher amount of particulate organic and inorganic matter coming from wastewater discharges, as it coincided with a period of major tourist affluence in

the region. In Foz de Almagem, the highest mean value was observed in October 2001 ( $103.83 \text{ mg L}^{-1}$ ) and happened after the first autumn rains.

Dissolved oxygen concentration and oxygen saturation followed the same tendency, showing higher mean values in December 2001 (Foz de Almagem:  $11.24 \text{ mg L}^{-1}$  and  $131.83 \%$ ; Salgados:  $21.18 \text{ mg L}^{-1}$  and  $220.37 \%$ ). In Salgados, seasonal variation was greater than in Foz de Almagem.

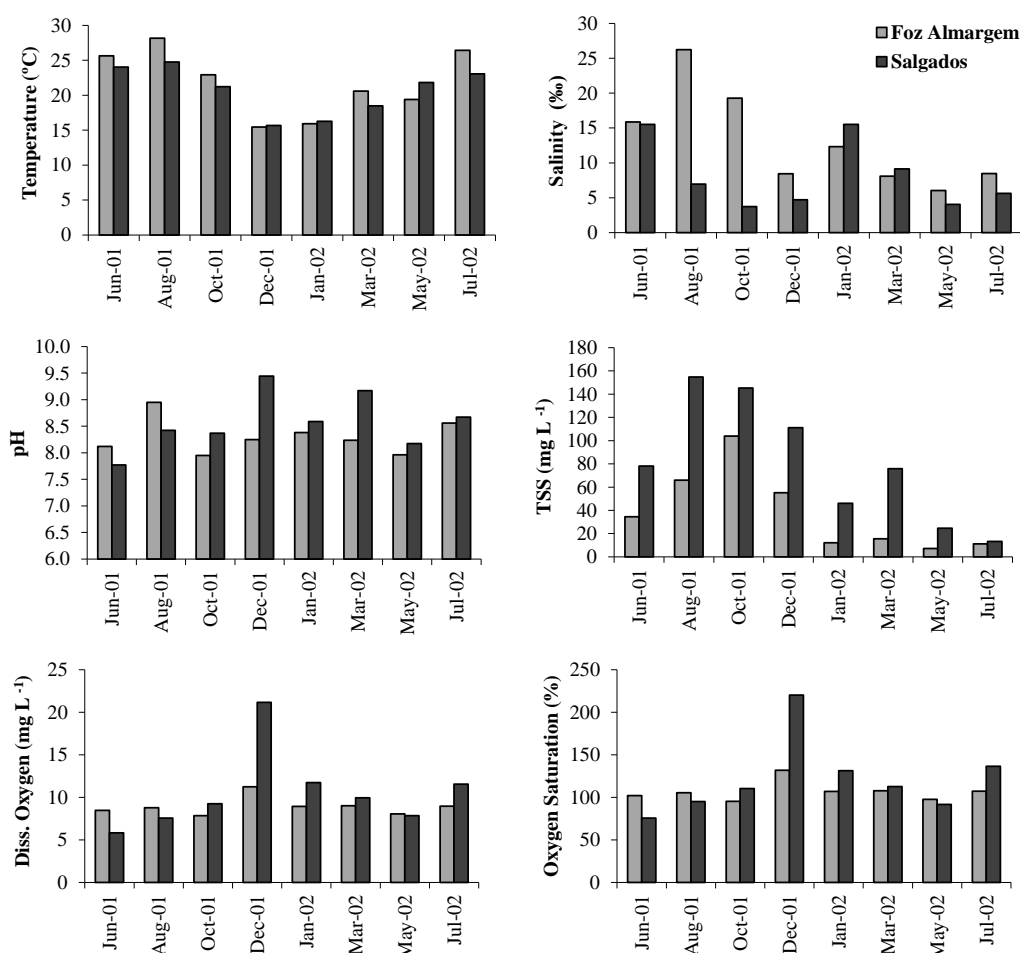


Figure 2.29 - Seasonal variation of physical and chemical water parameters mean values (salinity, temperature, pH, total solids in suspension, dissolved oxygen and oxygen saturation) in Foz de Almagem and Salgados coastal lagoons.

Total dissolved inorganic nitrogen ranged from  $4.12 \mu\text{M N}$  (August 2001) to  $96.45 \mu\text{M N}$  (January 2002) in Foz de Almagem and in Salgados mean values went from  $14.44 \mu\text{M N}$  (October 2001) to  $342.74 \mu\text{M N}$  (August 2001) (Figure 2.30). Nitrates were the most important nitrogen compounds in Foz de Almagem lagoon, representing 5.87% to 98.83% of total dissolved inorganic nitrogen determined all over the sampling period. In Salgados lagoon, nitrates mean concentrations oscillated between  $1.24 \mu\text{M N}$  (July 2002) and  $79.89 \mu\text{M N}$  (January 2002), contributing 5.24 % to 66.37 % to total

dissolved inorganic nitrogen. Ammonia was the most relevant nitrogen compound in this lagoon, representing 21.23 % to 93.80 % of total dissolved inorganic nitrogen. The lowest (10.70  $\mu\text{M N}$ ) and the highest (321.48  $\mu\text{M N}$ ) mean values of ammonia were registered in January 2002 and August 2001, respectively.

Total phosphorus and orthophosphate concentrations followed a similar tendency in both lagoons, presenting lower mean values during winter months and higher values through summer months. In Foz de Almargem, orthophosphates concentration varied between 0.20  $\mu\text{M P}$  (December 2001) and 1.87  $\mu\text{M P}$  (August 2001) and total phosphorus ranged from 0.21  $\mu\text{M P}$  (December 2001) to 2.20  $\mu\text{M P}$  (July 2002). Orthophosphates represented 40.50 % to 99.48 % of the total phosphorus determined during the sampling period. Salgados concentrations of orthophosphates and total phosphorus were considerably higher than those from Foz de Almargem. Lowest mean values (orthophosphates: 26.77  $\mu\text{M P}$ ; total phosphorus: 28.66  $\mu\text{M P}$ ) were found in January 2002 and the highest values were determined in August 2001 (orthophosphates: 107.86  $\mu\text{M P}$ ; total phosphorus: 115.91  $\mu\text{M P}$ ) and June 2001 (total phosphorus: 116.13  $\mu\text{M P}$ ). In Salgados lagoon, orthophosphates also meant a great contribution (75.00 % - 93.43 %) to total phosphorus concentrations.

In both lagoons, the lowest N: P ratio was determined in August 2001 (Foz de Almargem: 3.31; Salgados: 0.37) and the highest happened in January 2002 (Foz de Almargem: 81.72; Salgados: 4.88). In Foz de Almargem, from June till October 2001, values were under 16 (16 N: 1 P, the optimum ration for phytoplankton assimilation), indicating that nitrogen was the limiting nutrient. From December 2001 to July 2002, values were always above 16, which meant that phosphorus became the limiting nutrient, due to nitrogen increase. During the studied period, Salgados lagoon presented always values inferior to 16, which revealed that phosphorus was in excess.

Chlorophyll *a* mean concentrations in the two lagoons were quite distinct (Figure 2.31), with values oscillating between 0.38  $\mu\text{g L}^{-1}$  (June 2001) and 9.68  $\mu\text{g L}^{-1}$  (March 2002) in Foz de Almargem and varying from 19.34  $\mu\text{g L}^{-1}$  (October 2001) to 368.69  $\mu\text{g L}^{-1}$  (July 2002). Moreover, Salgados lagoon presented several episodes of high concentrations, namely in March 2002 (330.49  $\mu\text{g L}^{-1}$ ), in December 2001 (230.67  $\mu\text{g L}^{-1}$ ) and August 2001 (167.58  $\mu\text{g L}^{-1}$ ).

Phaeo-pigments mean concentrations were much lower than chlorophyll *a*, ranging from 0.21  $\mu\text{g L}^{-1}$  (January 2001) to 2.58  $\mu\text{g L}^{-1}$  (May 2002) in Foz de Almargem and from 4.69  $\mu\text{g L}^{-1}$  (October 2001) to 45.34  $\mu\text{g L}^{-1}$  (July 2002) in Salgados lagoon.

Margalef's pigment diversity in Foz de Almargem varied from 1.91 (June 2001) to 2.62 (August 2001), presenting high values also in March (2.60) and July 2002 (2.61). In Salgados lagoon, pigments diversity mean values went from 1.84 (December 2001) to 2.56 (May 2002).

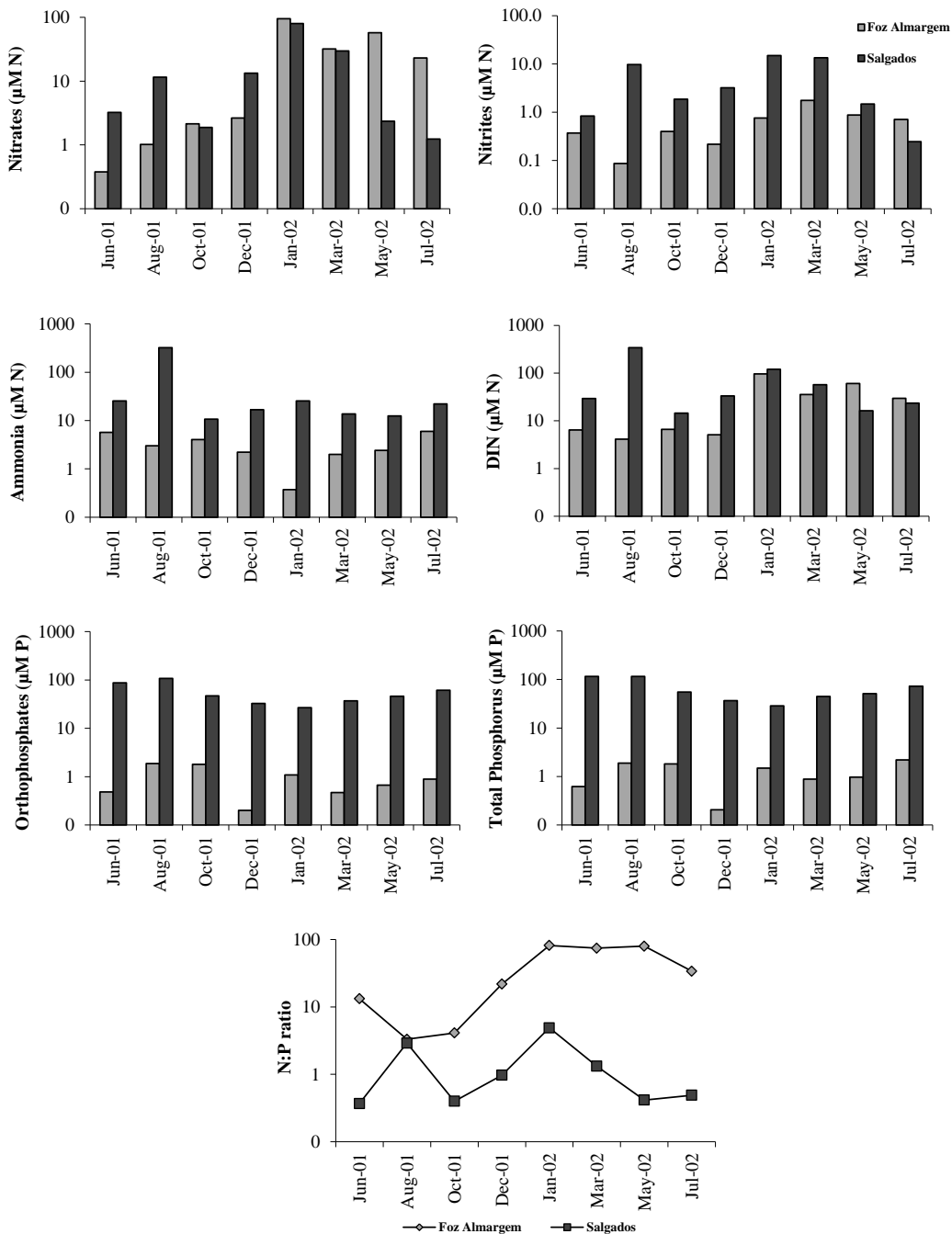


Figure 2.30 - Seasonal variation of nitrogen compounds (nitrates, nitrites and ammonia), total dissolved nitrogen (DIN), orthophosphates, total phosphorus concentration and N: P ratio (DIN: orthophosphate) mean values in Foz de Almargem and Salgados coastal lagoons.

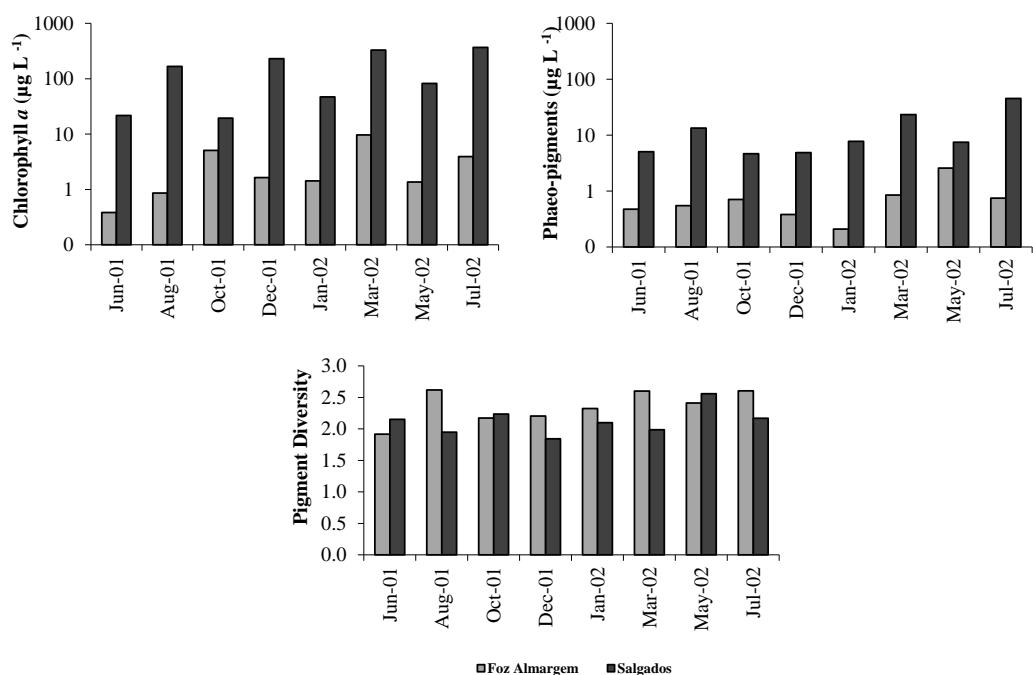


Figure 2.31 - Seasonal variation of photosynthetic pigments mean concentrations (chlorophyll *a* and phaeo-pigments) and Margalef's pigment diversity index in Foz de Almagem and Salgados coastal lagoons.

Foz de Almagem presented higher annual means of temperature, salinity, nitrates concentration, N:P ratio and pigment diversity, while in Salgados lagoon were found greater values for pH, total solids in suspension, dissolved oxygen, nitrites, ammonia, total dissolved inorganic nitrogen, orthophosphates, total phosphorus, chlorophyll *a* and phaeo-pigments concentrations (Table 2.17).

Statistical analyses found significant ( $0.01 < p \leq 0.05$ ) and highly significant ( $p \leq 0.01$ ) differences between the annual mean values of most water parameters in the two lagoons. Just for temperature, nitrates and dissolved oxygen concentrations, results indicated that differences between lagoons were not significant ( $p > 0.05$ ) (Appendix I.E).

Table 2.17 – Annual mean values and standard deviation of water parameters in Foz de Almargem and Salgados coastal lagoons.

	<i>Foz de Almargem</i>	<i>Salgados</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Temperature (°C)	21.82±4.59	20.92±3.49
Salinity (‰)	13.09±7.96	8.11±5.25
pH	8.30±0.39	8.61±0.61
Total Solids in Suspension	38.22±33.60	81.13±68.20
Dissolved oxygen concentration (mg L <sup>-1</sup> )	8.91±1.61	10.19±5.07
Nitrates concentration (µM N)	26.75±42.41	17.90±29.82
Nitrites concentration (µM N)	0.65±0.61	5.71±7.79
Ammonia concentration (µM N)	3.21±3.00	56.04±123.00
Total dissolved inorganic nitrogen concentration (µM N)	30.62±42.41	79.65±128.30
Orthophosphates concentration (µM P)	0.93±0.69	55.73±34.68
Total Phosphorus concentration (µM P)	1.26±0.76	65.15±42.04
N:P ratio	39.06±41.18	1.47±1.84
Chlorophyll a concentration (µg L <sup>-1</sup> )	3.04±3.43	158.46±177.95
Phaeo-pigments concentration (µg L <sup>-1</sup> )	0.81±0.88	13.99±15.21
Pigment diversity index (bits)	2.36±0.27	2.12±0.31

Samples distribution on the first two axes of PCA (Figure 2.32) clearly indicated the separation of Salgados and Foz de Almargem, along axis I. The first axis accounted for 29.8 % of the total variance and the parameters with greater contribution to this axis were orthophosphates (component loading = 0.855), total phosphorus (component loading = 0.820), chlorophyll *a* (component loading = 0.716), ammonia (component loading = 0.699), pigments diversity (component loading = -0.683), N: P ratio (component loading = -0.678) and total solids in suspension (component loading = 0.676). Salgados samples were plotted in the right side of axis I, as they showed higher concentrations of orthophosphates, total phosphorus, chlorophyll *a*, ammonia, total solids in suspension and lower values of pigments diversity and N:P ratio. In the left side of axis I, Foz de Almargem samples presented the opposite variation for these parameters.

The second axis explained 17.3 % of the total variance and disposed samples according to a seasonal gradient, as it was mainly defined by temperature (component loading = -0.804), dissolved oxygen concentration (component loading = 0.727) and pH (component loading = 0.579). On the upper side of the diagram were displayed samples with lower values of temperature and higher values of dissolved oxygen and pH (*e.g.* Salgados January 2002), while on the bottom side were represented samples with higher temperature and lower dissolved oxygen concentration and pH (*e.g.* Salgados June

2001). Nitrates, total dissolved inorganic nitrogen, salinity and water level were less important for the first and second axes ordination, presenting major component loadings in the third and fourth axes.

Parameters disposed near each other on the ordination diagram presented high positive correlation coefficients: total solids in suspension, orthophosphates, total phosphorus and ammonia; chlorophyll *a* and phaeo-pigments; cumulative rainfall and dissolved oxygen concentration. Those parameters pointing in opposite directions were negatively correlated, as orthophosphates concentration and N:P ratio or chlorophyll *a* and pigments diversity.

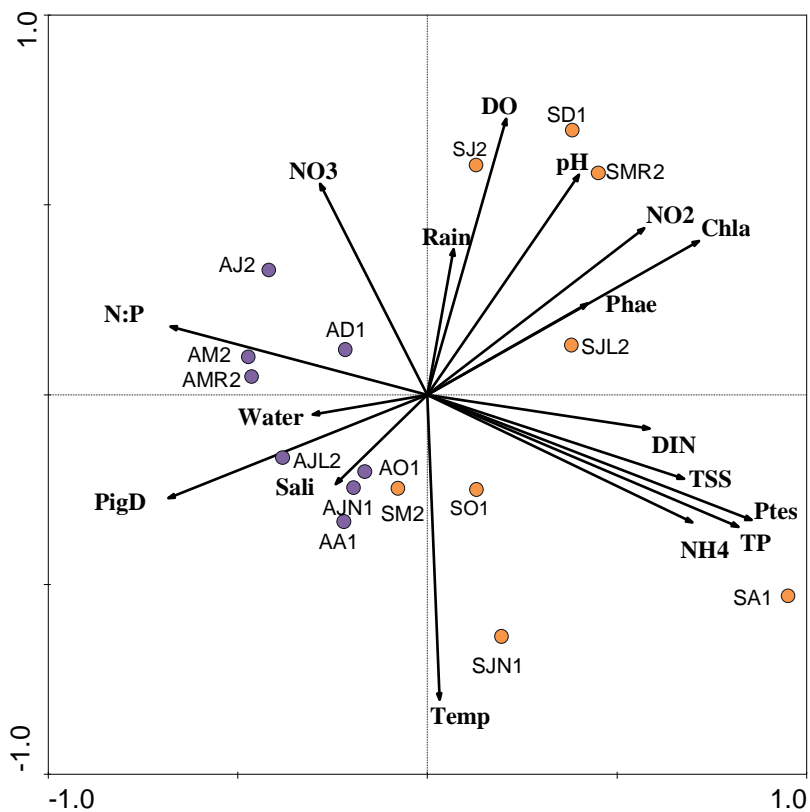


Figure 2.32 - Principal Component Analysis performed on the hydrological and water parameters mean values from Foz de Almargem and Salgados coastal lagoons. Cumulative percentage variance explained by axes: I – 29.8 %; I + II – 47.2 %.

*Station codes:* First character corresponds to the coastal lagoon (A – Foz de Almargem; S – Salgados) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001, 2-2002).

*Environmental variables:* Rain- cumulative rainfall in 10 days previous to sampling; Water- water level in the lagoon; Temp- water temperature; Sali- salinity; pH; DO- dissolved oxygen concentration; TSS- total solids in suspension; NO3- nitrates concentration; NO2- nitrites concentration; NH4- ammonia concentration; DIN- total dissolved inorganic nitrogen concentration; Ptes- orthophosphates concentration; TP- total phosphorus concentration; N: P- DIN and TP ratio; Chla- Chlorophyll *a* concentration; Phae- Phaeo-pigments concentration; PigD- pigments diversity index.

### 2.3.3.3. Sediment parameters

During the studied period, Foz de Almargem presented a variation in the mean values of sand content (13.42%) and clay content (3.44 %) smaller than the variation in Salgados (sand content: 18.60%; clay content: 16.97%) (Figure 2.33). In Foz de Almargem, sand content oscillated between 71.72% (May 2002) and 85.14% (July 2002), while in Salgados values went from 64.17% (August 2001) to 82.77% (January 2002). Minimum and maximum clay content in Foz de Almargem was 1.80 % (July 2002) and 5.24 % (August 2001). Mean values in Salgados ranged from 1.14 % (December 2001) to 18.11 % (July 2002).

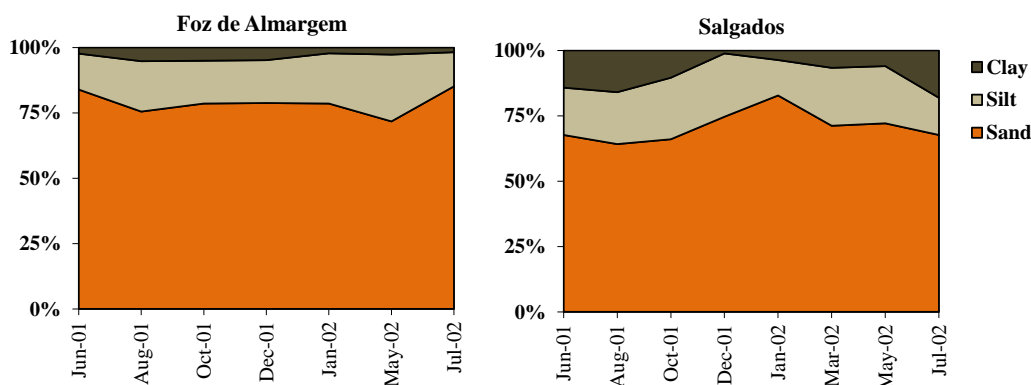


Figure 2.33 - Seasonal variation of mean sand, silt and clay contents in Foz de Almargem and Salgados coastal lagoons.

Mean water content in the sediment of the two lagoons followed a similar seasonal trend (Figure 2.34), but in May 2002, a sudden increase was registered in Foz de Almargem (41.33%). In Salgados, water content did not go above 36.57% (October 2001).

Organic matter content was higher in Salgados lagoon, till December 2001. From January to July 2002, greater values occurred in Foz de Almargem. Maximum values in both lagoons were determined in August 2001 (Foz de Almargem: 4.56 %; Salgados: 6.82 %).

Most of the time, higher

chlorophyll *a* and phaeo-pigments concentrations were observed in Salgados lagoon, except in January and May 2002. In Foz de Almargem, May 2002 was the month when the highest mean values were determined (chlorophyll *a* = 16.80  $\mu\text{g g}^{-1}$ ; phaeo-pigments = 18.46  $\mu\text{g g}^{-1}$ ) and the lowest values occurred in June 2001 (chlorophyll *a* = 2.16  $\mu\text{g g}^{-1}$

<sup>1</sup>) and December 2001 (phaeo-pigments = 2.07  $\mu\text{g g}^{-1}$ ). In Salgados, the highest mean values were registered in October 2001 (chlorophyll *a* = 28.15  $\mu\text{g g}^{-1}$ ; phaeo-pigments = 29.59  $\mu\text{g g}^{-1}$ ) and in January 2002 were observed the lowest values (chlorophyll *a* = 3.37  $\mu\text{g g}^{-1}$ ; phaeo-pigments = 3.66  $\mu\text{g g}^{-1}$ ).

Concerning chlorophyll *a* degradation index, Foz de Almargem presented higher values than Salgados, except in October 2001 (when the highest value in Salgados was achieved – 53.20%) and July 2002. The highest value in Foz de Almargem (57.91%) occurred in January 2002 and the lowest value (31.37%) happened in July 2002. In Salgados, the lowest value (26.72%) was registered in March 2002.

In terms of pigment diversity, Foz de Almargem showed mean values greater than Salgados during most of the sampling periods, apart from December 2001 and July 2002. Highest values in Foz de Almargem and Salgados were determined in October 2001 (Salgados: 2.95) and January 2002 (Foz de Almargem: 3.31).

Foz de Almargem presented annual mean values of clay content, silt content, chlorophyll *a* and phaeo-pigments concentrations lower than Salgados and higher means of sand content, water content, chlorophyll *a* degradation index and pigments diversity (Table 2.18).

Statistical analyses performed with the mean annual values revealed that, for most sediment parameters differences between lagoons were not significant (Appendix I.F). Only chlorophyll *a* concentration and pigments diversity showed evidences of significant differences in the two lagoons ( $0.01 < p \leq 0.05$ ).

Table 2.18 – Annual mean values and standard deviation of sediment parameters in Foz de Almargem and Salgados coastal lagoons.

	<i>Foz de Almargem</i>	<i>Salgados</i>
	Mean±Std.dev.	Mean±Std.dev.
Clay content (%)	2.99±3.38	9.46±11.29
Silt content (%)	15.59±21.51	19.58±20.70
Sand content (%)	81.42±24.06	70.96±30.98
Water content (%)	30.33±8.22	28.50±8.21
Organic matter content (%)	2.96±2.87	3.76±4.18
Chlorophyll <i>a</i> concentration ( $\mu\text{g g}^{-1}$ )	7.05±6.66	15.62±13.79
Phaeo-pigments concentration ( $\mu\text{g g}^{-1}$ )	6.89±7.84	10.66±13.83
Chlorophyll <i>a</i> degradation index (%)	44.44±15.31	37.45±13.47
Pigment diversity index (bits)	2.96±0.54	2.62±0.36

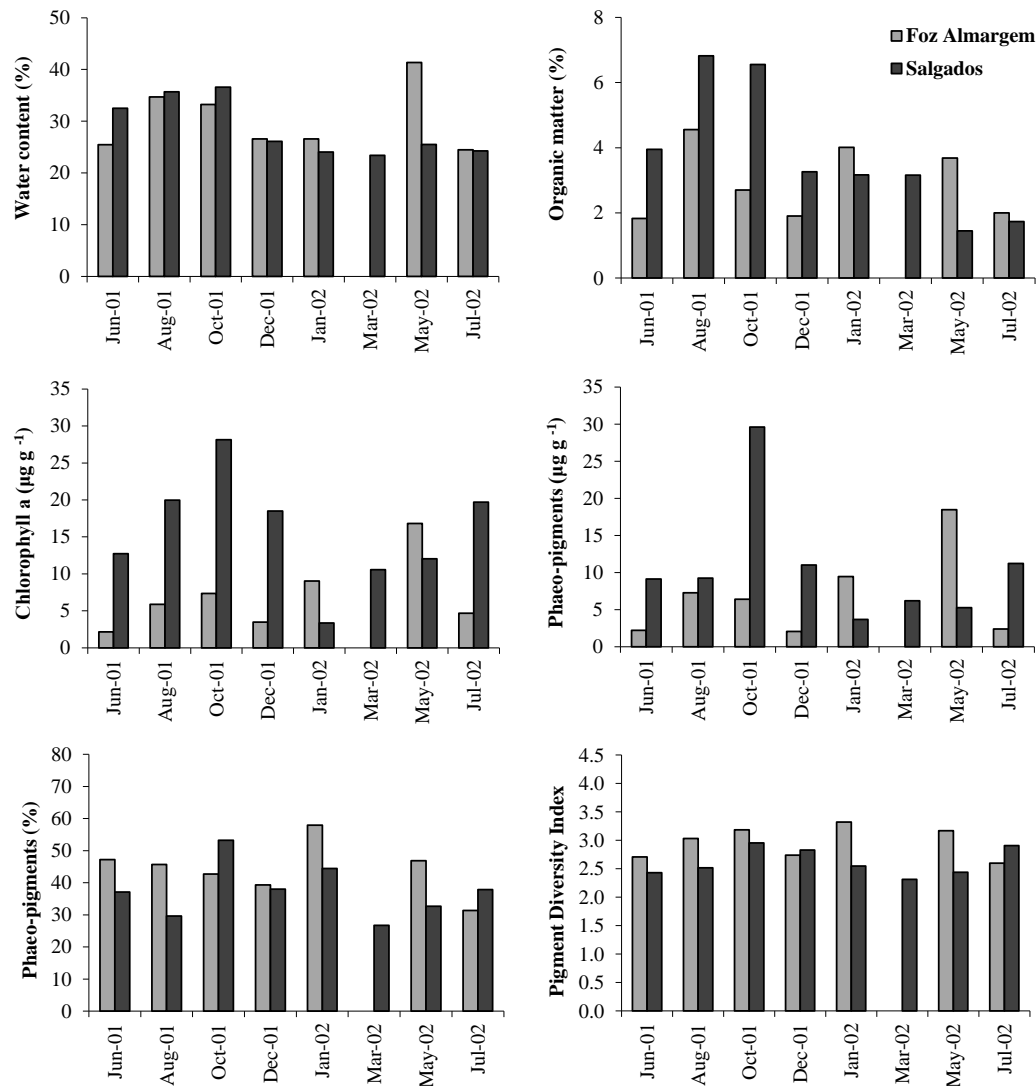


Figure 2.34 - Seasonal variation of mean values for water content, organic matter content, photosynthetic pigments concentration (chlorophyll *a* and phaeo-pigments), chlorophyll *a* degradation index (% phaeo-pigments) and Margalef's pigment diversity index in Foz de Almagrem and Salgados coastal lagoons.

The PCA ordination biplot showed that phaeo-pigments concentration (component loading = 0.935), chlorophyll *a* concentration (component loading = 0.876), organic matter content (component loading = 0.811), water content (component loading = 0.733) and clay content (component loading = 0.702) were the major sediment parameters influencing lagoon samples ordination along the first axis (Figure 2.35).

On the right side of the axis were plotted the samples with higher values of these parameters (*e.g.* Salgados October 2001) and on the left side were the samples characterized by lower values (*e.g.* Foz de Almagrem July 2002). The first axis accounted for 42.0% of the total variance. The secondary axis (II) separated Foz de Almagrem samples from Salgados samples. The parameters with greater contribution to

this axis were pigment diversity (component loading = 0.893) and chlorophyll *a* degradation index (component loading = 0.779).

Foz de Almagem samples were displayed on the upper part of the diagram, as they presented higher values of pigment diversity and chlorophyll *a* degradation index. On the bottom part of the diagram were plotted Salgados samples, due to lower values of these parameters. The second axis explained 25.3 % of the total variance. Sand: Mud ratio and silt content were less relevant for the first and second axis, presenting higher component loadings in the third and fourth axes.

Parameters as phaeo-pigments concentration and organic matter content, silt and clay content, pigment diversity and chlorophyll *a* degradation index, had high positive correlation coefficients, as they were represented close to each other.

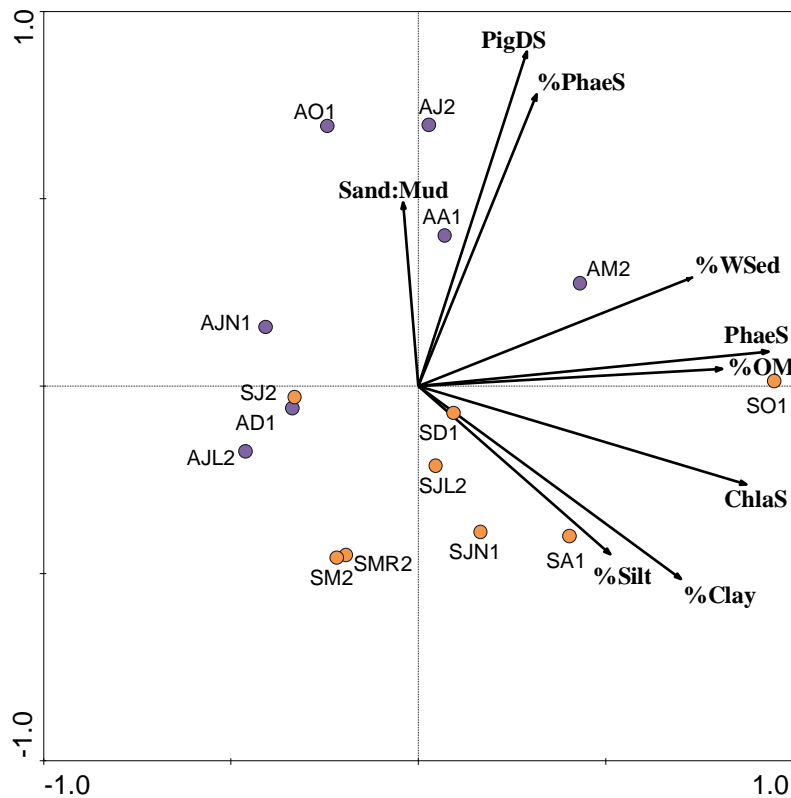


Figure 2.35 - Principal Component Analysis performed on the sediment parameters mean values from Foz de Almagem and Salgados coastal lagoons. Cumulative percentage variance explained by axes: I – 42.0 %; I + II – 67.3 %.

*Station codes:* First character corresponds to the coastal lagoon (A – Foz de Almagem; S – Salgados) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001, 2-2002).

*Sediment parameters:* Sand:Mud – Sand mud ratio; %Clay – Percentage of clay; %Silt – Percentage of silt; %OM – Percentage of organic matter; %WSed – Water content; ChlaS - Chlorophyll *a* concentration; PhaeS - Phaeo-pigments concentration; PigDS - Margalef's pigment diversity index; %PhaeS - Chlorophyll *a* degradation index.

#### 2.3.3.4. Relations between environmental parameters

The analysis of environmental parameters relations and its relevance in the characterization of Foz de Almargem and Salgados samples was performed through a PCA, considering the most important water and sediment parameters determined in previous analyses (Figure 2.32 and 2.35) performed separately for each type of parameter.

PCA ordination biplot disposed all samples from Foz de Almargem in the left side of axis I and most samples of Salgados lagoon in the right side of this axis (Figure 2.36). The first axis accounted for 37.6 % of the total variance and the parameters with greater contribution to this axis were clay content (component loading = 0.934), chlorophyll *a* in the sediment (component loading = 0.871), orthophosphates (component loading = 0.858), organic matter content (component loading = 0.658) and phaeo-pigments concentration in the sediment (component loading = 0.655). Phaeo-pigments concentration in the sediment also had a relevant contribution to axis II (component loading = 0.637) and jointly with pigments diversity in water (component loading = 0.744), N:P ratio (component loading = 0.624), chlorophyll *a* in water (component loading = -0.591) and dissolved oxygen (component loading = -0.551), determined ordination along axis II. The second axis explained 24.3 % of the total variance. Although dissolved oxygen could be associated to axis II, its major contribution was to axis III (component loading = 0.711), together with temperature (component loading = -0.815). Temperature was highly and positively correlated to organic matter content. Thereby, the majority of Salgados samples were characterized by higher mean values of clay content, chlorophyll *a* in the sediment, orthophosphates concentration, organic matter content, phaeo-pigments concentration in the sediment, chlorophyll *a* in the water, dissolved oxygen concentration and lower mean values of pigments diversity in water and N:P ratio.

Foz de Almargem samples had lower mean values of clay content, chlorophyll *a* in the sediment, orthophosphates concentration. Some samples showed higher mean values of pigments diversity in water, N:P ratio, phaeo-pigments concentration in the sediment and lower values of chlorophyll *a* in the water and dissolved oxygen concentration (*e.g.* May and January 2002). Some other samples presented the opposite variation of these parameters (*e.g.* June and December 2001).

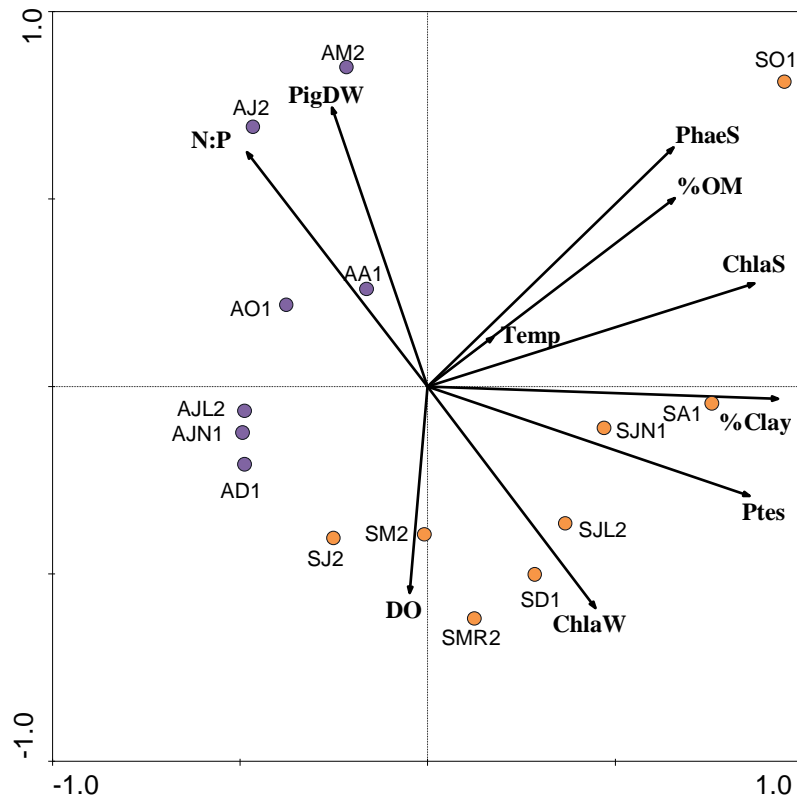


Figure 2.36 - Principal Component Analysis performed on the environmental parameters mean values from Foz de Almagem and Salgados coastal lagoons. Cumulative percentage variance explained by axes: I – 37.6 %; I + II – 61.9 %.

*Station codes:* First character corresponds to the coastal lagoon (A – Foz de Almagem; S – Salgados) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; MR- March; M- May; JL- July) and year of survey (1- 2001, 2-2002).

*Water variables:* Temp – Temperature; DO- dissolved oxygen concentration; Ptes- orthophosphates concentration; N: P- DIN and TP ratio; ChlaW- Chlorophyll *a* concentration; PigDW- pigments diversity index.

*Sediment variables:* %Clay- Percentage of clay; %OM – Percentage of organic matter; ChlaS - Chlorophyll *a* concentration; PhaeS- Phaeo-pigments concentration.

The assessment of chlorophyll *a* in the water ( $\text{mg m}^{-2}$ ) and chlorophyll *a* in the sediment ( $\text{mg m}^{-2}$ ) in the two lagoons revealed that in Foz de Almagem the amount of benthic chlorophyll was greater than pelagic chlorophyll, while in Salgados lagoon it was observed the inverse situation. Just in June and October 2001, chlorophyll *a* in Salgados sediment presented higher values than chlorophyll *a* in the water (Figure 2.37).

During the studied period, monthly mean values of chlorophyll *a* in Foz de Almagem sediment went from  $5.45 \text{ mg m}^{-2}$  (93.5%) in June 2001 to  $49.22 \text{ mg m}^{-2}$  (97.3%) in May 2002 and the annual mean was  $20.65 \text{ mg m}^{-2}$ , accounting 89.3 % of total chlorophyll *a* determined in the lagoon.

Mean values of chlorophyll *a* in the water were lower than in the sediment and varied between 0.38 mg m<sup>-2</sup> (6.5%) in June 2001 and 5.06 mg m<sup>-2</sup> (18.3%) in October 2001. The annual mean value of 2.10 mg m<sup>-2</sup> (without data from March 2002) represented 10.7% of total chlorophyll *a* in Foz de Almargem.

In Salgados lagoon, chlorophyll *a* in the sediment presented the minimum value in January 2002 (9.53 mg m<sup>-2</sup>; 18.1%), the maximum value in October 2001 (79.40 mg m<sup>-2</sup>; 80.9%) and an annual mean value of 44.33 mg m<sup>-2</sup> corresponding to 30.6% of total chlorophyll *a*. June and October 2001 were the only months with greater amount of chlorophyll *a* in the sediment than in the water column, accounting 59.5% (36.03 mg m<sup>-2</sup>) and 80.9 % (79.40 mg m<sup>-2</sup>) of total chlorophyll determined in these months.

Mean monthly values of chlorophyll *a* in the water (without the intermediate station data) ranged from 18.80 mg m<sup>-2</sup> (19.1%) in October 2001 to 414,33 mg m<sup>-2</sup> (88.1%) in July 2002 and the annual mean value determined was 172.6 mg m<sup>-2</sup> representing 69.4 % of total chlorophyll *a*.in Salgados lagoon.

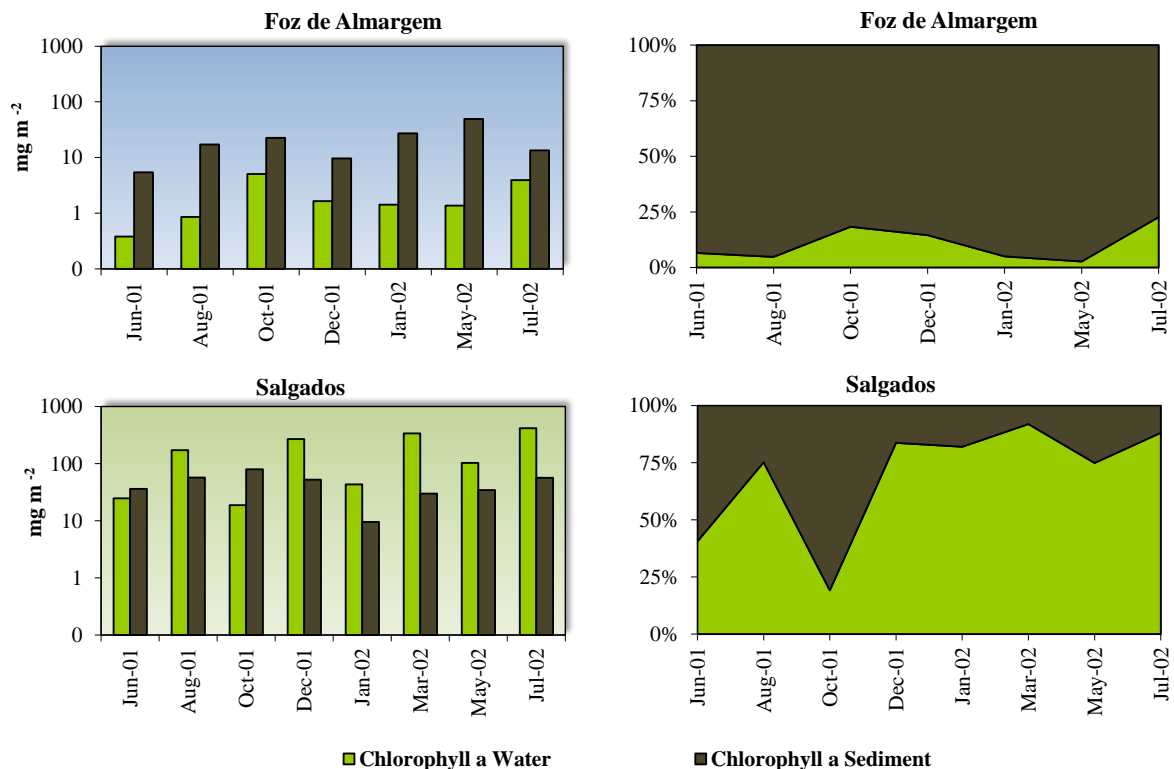


Figure 2.37 - Seasonal variation of chlorophyll *a* in the sediment, chlorophyll *a* in the water and percentage of each relative to total chlorophyll *a* in Foz de Almargem and Salgados lagoons.

### 2.3.3.5. Comparison of environmental parameters during isolation and connection of the lagoons with the sea

During December 2001 samplings, Foz de Almargem and Salgados lagoons were isolated from the sea and in January 2002, the two lagoons were in connection with the sea and samplings took place during the water outflow from the lagoons. Thus, mean data from these two periods were compared.

Both lagoons presented a decrease in total solids in suspension, dissolved oxygen, chlorophyll *a* concentrations and showed an increase in temperature, salinity, nitrates, nitrites, total dissolved inorganic nitrogen, N:P ratio and pigments diversity (Table 2.19). Orthophosphates, total phosphorus and pH increased in Foz de Almargem and decreased in Salgados lagoon, while ammonia and phaeo-pigments concentrations followed the opposite tendency.

Table 2.19 – Water and sediment parameters in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).

	<i>Foz de Almargem lagoon</i>		<i>Salgados lagoon</i>	
	Jan-02	Variation	Jan-02	Variation
<b>Water parameters</b>				
Temperature (°C)	15.92	<b>0.45</b>	16.28	<b>0.60</b>
Salinity (‰)	12.34	<b>3.91</b>	15.53	<b>11.20</b>
pH	8.38	<b>0.13</b>	8.59	<b>-0.85</b>
Total Solids in Suspension (mg L <sup>-1</sup> )	12.21	<b>-42.99</b>	46.02	<b>-65.14</b>
Dissolved oxygen (mg L <sup>-1</sup> )	8.94	<b>-2.30</b>	11.73	<b>-9.45</b>
Nitrates concentration (µM N)	95.32	<b>92.68</b>	79.89	<b>66.61</b>
Nitrites concentration (µM N)	0.76	<b>0.54</b>	14.93	<b>11.72</b>
Ammonia concentration (µM N)	0.37	<b>-1.85</b>	25.56	<b>8.83</b>
Total diss. inorg. nitrogen (µM N)	96.45	<b>91.37</b>	120.38	<b>87.17</b>
Orthophosphates concentration (µM P)	1.09	<b>0.89</b>	26.77	<b>-5.88</b>
Total Phosphorus concentration (µM P)	1.48	<b>1.28</b>	28.66	<b>-8.15</b>
N:P ratio	81.72	<b>59.88</b>	4.88	<b>3.91</b>
Chlorophyll <i>a</i> concentration (µg L <sup>-1</sup> )	1.43	<b>-0.21</b>	46.79	<b>-183.88</b>
Phaeo-pigments concentration (µg L <sup>-1</sup> )	0.21	<b>-0.17</b>	7.78	<b>2.89</b>
Pigment diversity index	2.32	<b>0.12</b>	2.10	<b>0.26</b>
<b>Sediment parameters</b>				
Clay content (%)	2.23	<b>-2.61</b>	3.66	<b>2.52</b>
Silt content (%)	19.22	<b>2.82</b>	13.57	<b>-10.65</b>
Sand content (%)	78.55	<b>-0.20</b>	82.77	<b>8.13</b>
Organic matter content (%)	4.01	<b>2.11</b>	3.16	<b>-0.10</b>
Chlorophyll <i>a</i> concentration (µg g <sup>-1</sup> )	9.03	<b>5.54</b>	3.37	<b>-7.35</b>
Phaeo-pigments concentration (µg g <sup>-1</sup> )	9.47	<b>7.40</b>	3.66	<b>-7.35</b>
Chlorophyll <i>a</i> degradation index (%)	57.91	<b>18.60</b>	44.44	<b>6.46</b>
Pigment diversity index	3.32	<b>0.58</b>	2.55	<b>-0.28</b>

The water parameters with major variations were chlorophyll *a*, nitrates, total dissolved inorganic nitrogen, total solids in suspension and N:P ratio.

Foz de Almargem lagoon presented the greatest variations for nitrates concentration (92.68  $\mu\text{M N}$ ), total dissolved inorganic nitrogen (91.37  $\mu\text{M N}$ ) and N:P ratio (59.88). January mean values of nitrates (95.32  $\mu\text{M N}$ ), N:P ratio (81.72) and pigments diversity (2.32) were also higher than in Salgados lagoon.

The remaining water parameters had greater variations in Salgados lagoon: temperature (0.60°C), salinity (11.20 ‰), pH (-0.85), total solids in suspension (-65.14  $\text{mg L}^{-1}$ ), dissolved oxygen concentration (-9.45  $\text{mg L}^{-1}$ ), nitrites (11.72  $\mu\text{M N}$ ), ammonia (8.83  $\mu\text{M N}$ ), orthophosphates (-5.88  $\mu\text{M P}$ ), total phosphorus (-8.15  $\mu\text{M P}$ ), chlorophyll *a* (-183.88  $\mu\text{g L}^{-1}$ ), phaeo-pigments concentration (2.89  $\mu\text{g L}^{-1}$ ) and pigments diversity (0.26).

Concerning sediment parameters, both lagoons had an increase in chlorophyll *a* degradation index. This parameter, just as silt content, sand content, chlorophyll *a* and phaeo-pigments concentrations were the ones with major variations.

In Foz de Almargem was determined a greater variation of chlorophyll *a* degradation index (18.60), phaeo-pigments concentration (7.50  $\mu\text{g g}^{-1}$ ), clay content (-2.61 ‰), organic matter content (2.11 ‰) and pigments diversity (0.58).

The variation of other sediment parameters was higher in Salgados lagoon: silt content (-10.65%), sand content (8.13) and chlorophyll *a* concentration (-7.35  $\mu\text{g g}^{-1}$ ).

#### **2.3.3.6. Trophic state and Water quality**

The evaluation of the trophic state and water quality, through TSI (CHL), TSI (TP) and TRIX, showed that Foz de Almargem presented a lower trophic state and better water quality than Salgados lagoon (Figure 2.38).

In Foz de Almargem, TSI (CHL) mean monthly values ranged from 21.02 in June 2001 (oligotrophic) to 52.47 in March 2002 (eutrophic), but during most of the months the lagoon presented a trophic state of oligotrophy (25%), oligomesotrophy (37.5%) or mesotrophy (25%). The TSI (CHL) annual mean of 35.48 suggested a system with oligomesotrophic conditions.

TSI (TP) mean monthly values were higher, varying from 29.62 in December 2001 (oligotrophic) to 62.01 in October 2001 (eutrophic-hypereutrophic) and this index

classified a greater number of months as eutrophic (50%) and eutrophic-hypereutrophic (25%). The TSI (TP) annual mean of 51.78 indicated an eutrophic system.

The lowest and highest TRIX mean monthly values were determined in December 2001 (4.24) and October 2001 (5.98), to which corresponded a good water quality (system moderately productive with a mean trophic level) and a mediocre water quality (system moderate to highly productive and with a high trophic level), respectively. Most of the months were classified with a mediocre water quality (75%), which was the same category determined by the TRIX annual mean (5.38).

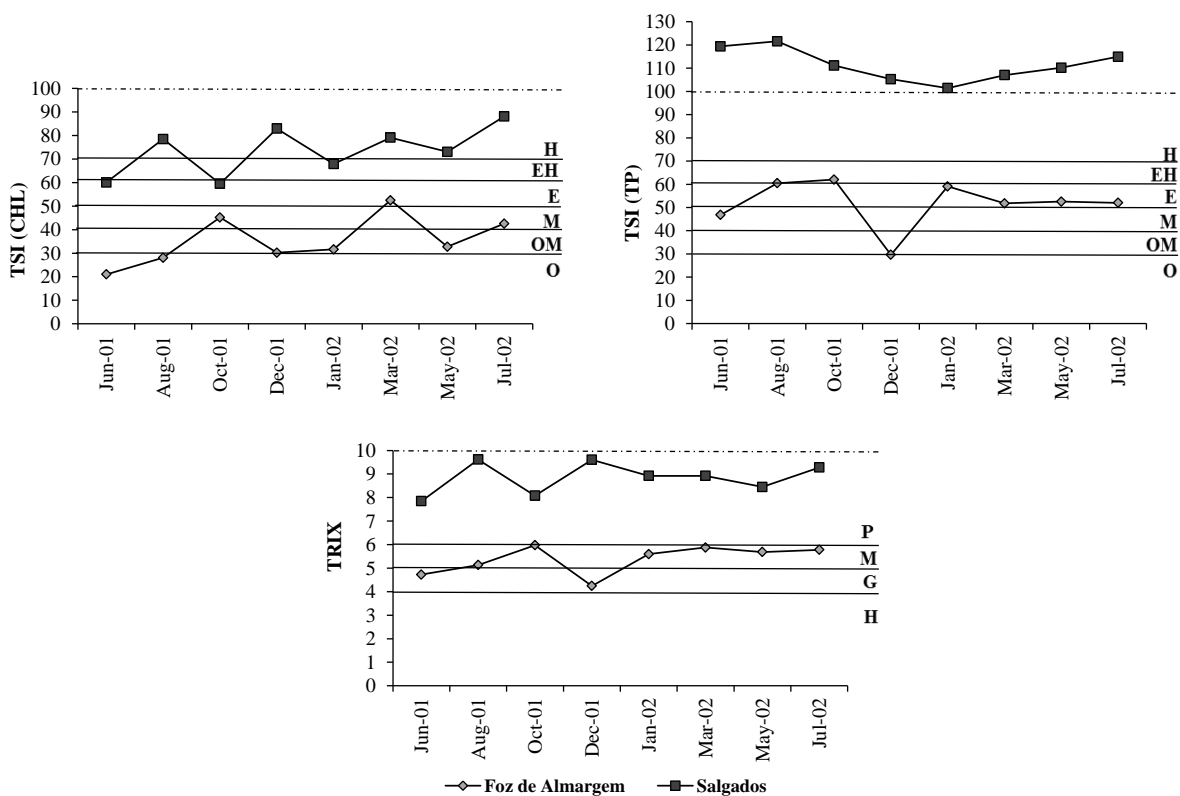


Figure 2.38 – Seasonal variation of trophic state and water quality indexes (means) in Foz de Almargem and Salgados coastal lagoons: TSI with chlorophyll (CHL), TSI with total phosphorus concentrations (TP) and TRIX.

TSI: O- Oligotrophic; OM- Oligomesotrophic; M- Mesotrophic; E- Eutrophic; EH- Eutrophic to Hypereutrophic; H- Hypereutrophic. TRIX: H- High water quality; G- Good water quality; M- Mediocre water quality; P- Poor water quality. Dashed lines represent the maximum theoretical value for each index scale.

In Salgados lagoon, all three indexes presented higher values than in Foz de Almargem. TSI (CHL) monthly mean values varied from 59.53 in October 2001 (eutrophic) to 88.13 in July 2002 (hypereutrophic), with most samples being classified as hypereutrophic (62.5%) or eutrophic-hypereutrophic (25%). The TSI (CHL) annual mean of 73.66 indicated a system with hypereutrophic conditions.

All the monthly mean values determined with TSI (TP) were above 100 (hypereutrophic), with a minimum of 101.40 in January 2002 and a maximum value occurring in August 2001 (121.57). The TSI (TP) annual mean was 111.36.

According to TRIX monthly mean values, all samples had a poor water quality (TRIX > 6) indicating a highly productive system. The lowest mean value (7.85) was determined in June 2001 and the highest value (9.61) was registered in December 2001. TRIX annual mean value in Salgados lagoon was 8.84.

The mean values of trophic state and water quality indexes determined for each lagoon with all data and with the data from sampling periods when the lagoons were closed (June-December 2001; March-July 2002) were similar using TRIX and slightly higher if TSI (CHL) was applied. TSI (TP) values with data from the closed period were lower in Foz de Almargem and higher in Salgados lagoon, compared to the TSI (TP) values obtained with all data. Despite these small variations in the trophic state and water quality values, the classification categories of each lagoon were the same using all data or the data when the lagoons were closed (Table 2.20).

Table 2.20 – Trophic state and water quality indexes mean values, standard deviation and classification in Foz de Almargem and Salgados lagoons, determined during all studied period (June 2001-July 2002) and when the lagoon was closed (June-December 2001; March-July 2002).

	<i>Foz de Almargem lagoon</i>	<i>Salgados lagoon</i>
	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
TSI (CHL): All data	35.48±11.45	73.66±12.34
	<b>Oligomesotrophic</b>	<b>Hypereutrophic</b>
TSI (CHL): Closed period	36.04±11.89	74.48±12.97
	<b>Oligomesotrophic</b>	<b>Hypereutrophic</b>
TSI (TP): All data	51.78±10.81	111.36±8.63
	<b>Eutrophic</b>	<b>Hypereutrophic</b>
TSI (TP): Closed period	50.74±11.14	112.78±8.12
	<b>Eutrophic</b>	<b>Hypereutrophic</b>
TRIX: All data	5.38±0.77	8.84±0.72
	<b>Mediocre water quality</b>	<b>Poor water quality</b>
TRIX: Closed period	5.35±0.81	8.83±0.77
	<b>Mediocre water quality</b>	<b>Poor water quality</b>

When data from January 2002 (lagoons opened) were compared with data from December 2001 (closed lagoons), all trophic state and water quality indexes in Foz de Almargem presented a positive variation, while in Salgados lagoon the variation was negative.

TSI (CHL) in Foz de Almargem showed a smaller variation ( $\Delta = 1.40$ ) than the one recorded in Salgados ( $\Delta = -15.02$ ). Foz de Almargem value in January 2002 (TSI CHL = 31.62), still corresponded to an oligomesotrophic system, as in December 2001 (TSI CHL = 30.23), but in Salgados lagoon, the trophic state decreased from hypereutrophic (TSI CHL = 82.95) to eutrophic-hypereutrophic (TSI CHL = 67.92).

The smallest variation in TSI (TP) was determined in Salgados ( $\Delta = -3.83$ ) and the trophic state classification in January 2002 (TSI TP = 101.40) was the same as in December 2001 (TSI TP = 105.23), hypereutrophic. The positive variation registered in Foz de Almargem ( $\Delta = 29.46$ ) raised the classification from oligotrophic (TSI TP = 29.62) to eutrophic (TSI TP = 59.08).

The increase of TRIx in Foz de Almargem ( $\Delta = 1.35$ ) meant a deterioration in water quality from good (TRIX = 4.24) to mediocre (TRIX = 5.59). Although TRIx decreased in Salgados lagoon ( $\Delta = -0.68$ ), water quality classification in January 2002 (TRIX = 8.92) was still poor, as in December 2001 (TRIX = 9.61).

The 90<sup>th</sup> percentile values of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ) in Foz de Almargem lagoon were considerably lower than the values determined in Salgados lagoon (Table 2.21).

According to the criteria defined by Pereira Coutinho *et al.* (2012) for semi-enclosed lagoons, during the sampling periods lagoons were closed (June-December 2001; March-July 2002), water quality was high in Foz de Almargem (Chlorophyll *a* 90<sup>th</sup> percentile  $< 30 \mu\text{g L}^{-1}$ ) and bad in Salgados lagoon (Chlorophyll *a* 90<sup>th</sup> percentile  $> 101.3 \mu\text{g L}^{-1}$ ). Data from both lagoons were insufficient for the determination of water quality when the lagoons were opened (January 2002).

The adaptation of Brito *et al.* (2012) classification system to the data available in the studied lagoons, suggested a high water quality (Chlorophyll *a* 90<sup>th</sup> percentile  $< 8 \mu\text{g L}^{-1}$ ) in Foz de Almargem lagoon during the growing seasons of 2001 (June to October) and 2001-2002 (June to October 2001; March to July 2002), while in 2002 (March to July) water quality would be good ( $8 \mu\text{g L}^{-1} < \text{Chlorophyll } a \text{ 90}^{\text{th}} \text{ percentile} < 12 \mu\text{g L}^{-1}$ ). In Salgados lagoon, water quality was considered bad (Chlorophyll *a* 90<sup>th</sup> percentile  $> 27 \mu\text{g L}^{-1}$ ) during the growing seasons of 2001, 2002 and 2001-2002.

Table 2.21 – Water quality in Foz de Almagem and Salgados lagoons, based on the 90<sup>th</sup> percentile of chlorophyll *a* ( $\mu\text{g L}^{-1}$ ), adapted from Brito *et al.* (2012) and Pereira Coutinho *et al.* (2012).

	<i>Foz de Almagem lagoon</i>	<i>Salgados lagoon</i>
<b>Pereira Coutinho <i>et al.</i> (2012): Semi-enclosed lagoons</b>		
<i>Ref. value during closed period (20 <math>\mu\text{g L}^{-1}</math>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> during closed period	7.56 $\mu\text{g L}^{-1}$	387.85 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality during closed period</b>	<b>High</b> ( $< 30 \mu\text{g L}^{-1}$ )	<b>Bad</b> ( $> 101,3 \mu\text{g L}^{-1}$ )
<b>Brito <i>et al.</i> (2012): Coastal water lagoons</b>		
<i>Ref. value during growing season (February-October: 5,3 <math>\mu\text{g L}^{-1}</math>)</i>		
90 <sup>th</sup> percentile chlorophyll <i>a</i> (June-October 2001)	5.98 $\mu\text{g L}^{-1}$	186.17 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>High</b> ( $< 8 \mu\text{g L}^{-1}$ )	<b>Bad</b> ( $> 27 \mu\text{g L}^{-1}$ )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (March-July 2002)	9.70 $\mu\text{g L}^{-1}$	559.01 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>Good</b> ( $8-12 \mu\text{g L}^{-1}$ )	<b>Bad</b> ( $> 27 \mu\text{g L}^{-1}$ )
90 <sup>th</sup> percentile chlorophyll <i>a</i> (June-October 2001; March-July 2002)	7.92 $\mu\text{g L}^{-1}$	381.61 $\mu\text{g L}^{-1}$
<b>Classification of Water Quality</b>	<b>High</b> ( $< 8 \mu\text{g L}^{-1}$ )	<b>Bad</b> ( $> 27 \mu\text{g L}^{-1}$ )

### **3. PHYTOPLANKTON COMMUNITIES**

#### **3.1. Specific Aims**

Concerning phytoplankton communities, three specific aims were established:

1. Characterize seasonal variation in phytoplankton communities; compare spatial patterns along the gradient of distance to the sea and between lagoons.
2. Study the relations between phytoplankton communities and environmental parameters in the two lagoons, namely hydrological and water parameters.
3. Evaluate the effect of lagoons opening in phytoplankton communities.
4. Determine the occurrence of potentially harmful phytoplankton and its relation with environmental parameters.
5. Evaluate the relation between phytoplankton communities, water quality and trophic state in the lagoons.

#### **3.2. Material and methods**

##### **3.2.1. Field procedures and laboratory analyses**

Field work was done from June 2001 to July 2002, with an interval of approximately 45 days and at the same time as water monitoring. In each lagoon, sampling took place in three stations along a gradient of distance from the sea (E1- Upstream; E2- Intermediate; E3- Downstream).

Phytoplankton was analysed in terms of biomass and abundance. The most common method of measuring phytoplankton biomass is to quantify chlorophyll *a* (Hartnett and Nash, 2004).

Water samples were collected in the superficial water layer (50 cm depth) and preserved in cold, dark conditions for laboratory quantification of chlorophyll *a* concentration (1 L volume) and phytoplankton count and identification (0.3 L volume).

Chlorophyll *a*, phaeo-pigments and pigments diversity determinations were described before in Chapter 2.

Counting and identification of phytoplankton cells was made at 400 x magnification with an inverted microscope, following Utermöhl's technique (Utermöhl, 1958), using the deposited algae contained in 300 ml of samples fixed with lugol. At least 50 fields or 100 individuals of the most abundant species were counted in each sample (Venrick, 1978). Identification was undertaken with reference to various authors, namely Newell and Newell (1977), Sykes (1981), Dodge (1982), Sournia (1984), Sournia (1986), Chrétiennot-Dinet (1990), Cox (1996), Tomas and Hasle (1997), Trigueros *et al.* (2000), John *et al.* (2002).

### 3.2.2. Data analysis

The phytoplankton community was studied in terms of taxonomic composition, species richness and abundance, diversity - Shannon-Wiener diversity index – (Shannon and Weaver, 1963) and evenness (Pielou, 1966).

In both lagoons, phytoplankton parameters were compared before and during the lagoons connection with the sea, based on data from December 2001 and January 2002, respectively.

A first approach considered data from each lagoon separately in order to analyse sampling stations variation and only then, mean values from the lagoons were compared.

Differences in phytoplankton parameters among stations and between the lagoons were tested through One-Way ANOVA and Student T test, respectively. Previously, a logarithmic transformation of data was done ( $\ln x + 1$ ) and the normality of data distribution just as the homogeneity of variances was investigated. Whenever data distribution did not present normality or variances were not homogeneous, the non-parametric test of Kruskal-Wallis or the Mann-Whitney U test, were applied. The LSD Fisher multiple comparison test was used to determine which of the three stations differed significantly (Maroco, 2010). The confidence level used in all statistical tests was 95 % ( $\alpha = 0.05$ ).

In order to see how the phytoplankton was associated with the environmental variables studied, canonical correspondence analyses (CCA) were applied to data of each sampling station and to the mean values of each lagoon. Phytoplankton data was presented as the abundance of the main taxonomic groups, after root transformation. Just the environmental variables that the PCA previously performed for each lagoon and

for the mean values of the lagoons showed not to be highly correlated were included in CCA (see Chapter 2).

Pearson and Spearman correlation coefficients were used to evaluate the bivariate linear associations between phytoplankton parameters and also, between environmental parameters, phytoplankton abundances and indexes, depending on data normality after transformation ( $\ln x+1$ ).

The softwares used for data analysis were CANOCO (Ter Braak, version 4.54, 1988-2005 Biometris) and SPSS (version 19, IBM SPSS Statistics).

### **3.3. Results and discussion**

#### **3.3.1. Foz de Almargem coastal lagoon**

##### **3.3.1.1. Phytoplankton communities**

During the studied period 29 phytoplankton *taxa* were identified in the lagoon (Appendix II.A), most of them (17) belonging to Bacillariophyceae. The remaining *taxa* were Chlorophyceae (2), Cryptophyceae (2), Dinophyceae (5), Euglenophyceae (1), Cyanophyceae (1) and pico-nano flagellate algae (< 20  $\mu\text{m}$ ). Three *taxa* (*Prorocentrum minimum* accounting 37%, *Gymnodinium* sp. and *Protoperidinium* sp.) constituted 70% of total phytoplankton cells sampled. Five *taxa* (*Cocconeis* sp., *Cyclotella* spp., *Navicula* spp., *Gymnodinium* sp., *Prorocentrum minimum*,) were represented in more than 50% of the samples and seven *taxa* occurred in more than 25% of the samples (*Diploneis* sp., *Grammatophora* sp., *Nitzschia* sp., *Rhodomonas* sp., *Protoperidinium* sp., *Scrippsiella trochoidea* and *Eutreptiella* sp.). Pico-nano flagellate algae accounted 6% of total abundance and were represented in 54% of the samples.

Taxonomic richness in the stations ranged from four (May 2002, intermediate; March 2002, downstream) to 12 (March 2002, intermediate) and till December, the three stations showed a similar pattern of variation. From January to July, the downstream station presented a different tendency of evolution compared to the other stations. During the studied period, the intermediate station was the one with greater variation in the number of *taxa*, followed by the downstream and upstream stations (Figure 3.1).

Taxonomic richness was positively correlated with Shannon-Wiener diversity ( $\rho = 0.609$ ;  $p = 0.002$ ) and Bacillariophyceae abundance ( $r = 0.728$ ;  $p < 0.001$ ).

Total phytoplankton abundance in the three stations showed different patterns of evolution during the studied period, except in March and May 2002. From June to October 2001, the upstream and intermediate stations followed the same tendency, decreasing in August and increasing in October. In December 2001 and January 2002 was observed an increment of total phytoplankton in the intermediate station, while upstream and downstream abundances decreased. During March and May 2002, all stations presented an augment and then a reduction, respectively. In July 2002, total phytoplankton downstream was higher than the abundances registered during the summer months of 2001, but in the upstream and intermediate stations, the abundances were lower and closer to the values observed in August 2001.

The intermediate station presented the lowest ( $32.77 \times 10^3 \text{ cell L}^{-1}$ ) and the highest ( $966.19 \times 10^3 \text{ cell L}^{-1}$ ) phytoplankton abundance from the three sampling stations. The greatest variation in time was registered in the intermediate station ( $933.42 \times 10^3 \text{ cell L}^{-1}$ ), while downstream ( $684.65 \times 10^3 \text{ cell L}^{-1}$ ) and upstream ( $531.17 \times 10^3 \text{ cell L}^{-1}$ ) the variation was smaller. Total phytoplankton abundance was negatively correlated with Shannon-Wiener diversity ( $\rho = -0.593$ ;  $p = 0.002$ ) and evenness ( $\rho = -0.764$ ;  $p < 0.001$ ) and positively correlated with Dinophyceae abundance ( $r = 0.812$ ;  $p < 0.001$ ).

The Shannon-Wiener diversity index and the evenness were lowest in March 2002 (downstream:  $H' = 0.09$ ;  $E = 0.05$ ), which coincided with the highest phytoplankton density that was caused by an increase of *Prorocentrum minimum* (Dinophyceae) representing 90 – 99 % of the total phytoplankton cells. Also in October 2001 the diversity and evenness were lower than the other sampling periods (upstream:  $H' = 0.30$ ;  $E = 0.13$ ), due to another Dinophyceae species growth, *Protoperdinium* sp. (77 – 96 % of total phytoplankton cells). In July 2002, maximum diversity and evenness were observed (upstream:  $H' = 2.77$ ;  $E = 0.82$ ). From June to October 2001, diversity and evenness increased with the proximity to the sea (upstream – downstream gradient), but after December no regular pattern occurred. The lowest variation in diversity and evenness during the studied period was observed in the intermediate station ( $H' = 2.13$ ;

$E = 0.64$ ) and greater variations were determined in the downstream ( $H' = 2.32$ ;  $E = 0.72$ ) and upstream stations ( $H' = 2.47$ ;  $E = 0.71$ ).

Shannon-Wiener diversity was positively correlated with evenness ( $\rho = 0.878$ ;  $p < 0.001$ ), Bacillariophyceae abundance ( $\rho = 0.607$ ;  $p = 0.002$ ), Cryptophyceae abundance ( $\rho = 0.407$ ;  $p = 0.021$ ) and it was negatively correlated with Dinophyceae abundance ( $\rho = -0.778$ ;  $p < 0.001$ ). Evenness also showed a negative correlation with Dinophyceae abundance ( $\rho = -0.769$ ;  $p < 0.001$ ).

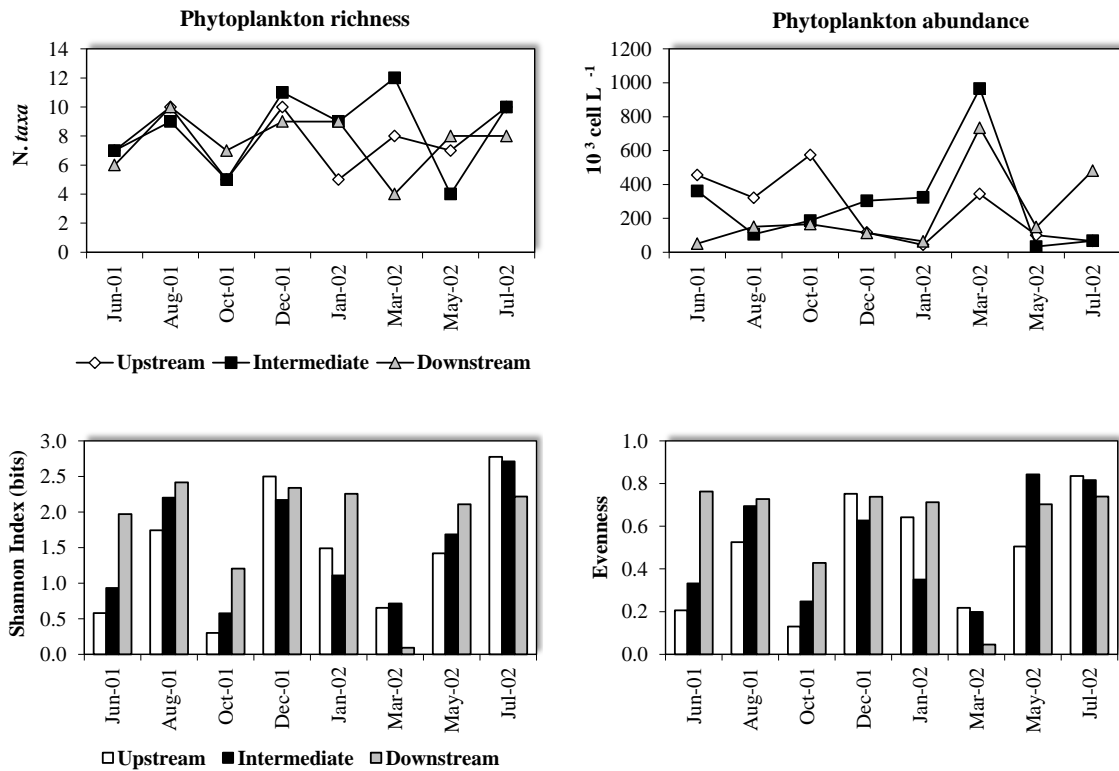


Figure 3.1 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Foz de Almargem sampling stations.

Through time, the upstream and intermediate stations were dominated by the same phytoplankton taxonomic classes, except in January 2002 when the lagoon was in connection with the sea (Jun-01: Dinophyceae; Aug-01: Pico-nano flagellate algae; Oct-01: Dinophyceae; Dec-01: Bacillariophyceae; Jan-02: Pico-nano flagellate algae / Dinophyceae; Mar-02: Dinophyceae; May-02: Dinophyceae; Jul-02: Bacillariophyceae). At the downstream station, Dinophyceae was the most relevant class, being replaced by Bacillariophyceae in August 2001, January and July 2002 (Figure 3.2).

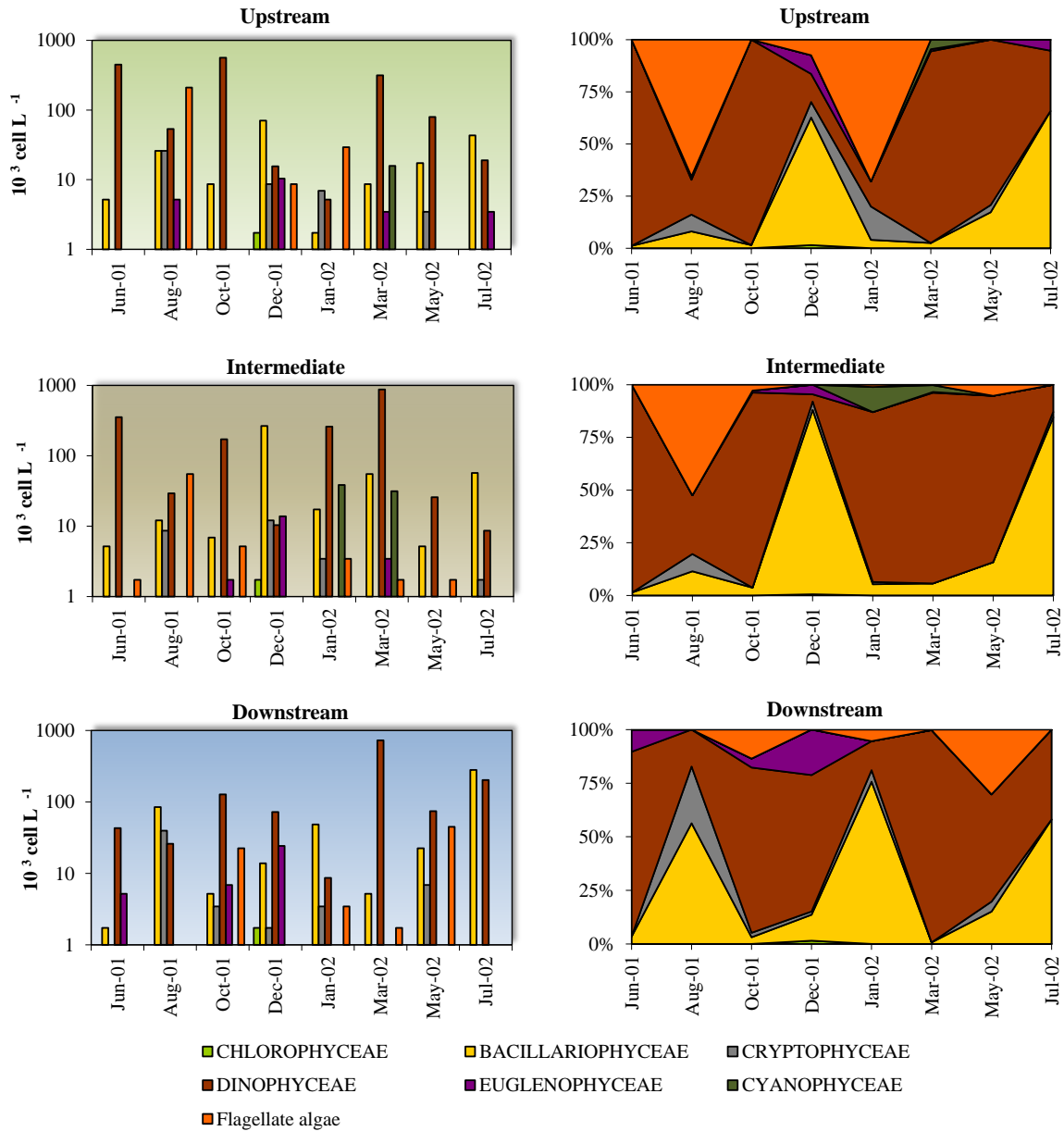


Figure 3.2 - Evolution of phytoplankton abundance *per class* and relative frequency of each class in Foz de Almargem sampling stations.

Pico-nano flagellate algae occurred in the upstream station just in three months (August 2001, December 2001, January 2002), but in two of them dominated the phytoplankton community, August 2001 ( $210 \times 10^3 \text{ cell L}^{-1}$ ; 66%) and January 2002 ( $29 \times 10^3 \text{ cell L}^{-1}$ ; 68%).

In December 2001 and July 2002 pico-nano flagellate algae were absent from the intermediate station, presenting the maximum abundance in August 2001 ( $55 \times 10^3 \text{ cell L}^{-1}$ ), which corresponded to 52% of total phytoplankton in the station.

The occurrence of pico-nano flagellate algae downstream was more irregular than in the intermediate station; the maximum abundance was lower than the highest values found in the other stations and was observed in May 2002 ( $45 \times 10^3 \text{ cell L}^{-1}$ ), accounting 30% of total phytoplankton in the station.

In the upstream station, Dinophyceae abundances went from  $5 \times 10^3 \text{ cell L}^{-1}$  in January 2002 (12% of total phytoplankton) to  $566 \times 10^3 \text{ cell L}^{-1}$  in October 2001 (98%), with high values also registered in June 2001 ( $450 \times 10^3 \text{ cell L}^{-1}$ ; 99%) and March 2002 ( $316 \times 10^3 \text{ cell L}^{-1}$ ; 92%). In May 2002, Dinophyceae abundance was lower ( $79 \times 10^3 \text{ cell L}^{-1}$ ; 79%), although it dominated the phytoplankton community (Figure 3.2). The most important *taxa* were *Gymnodinium* sp. ( $416 \times 10^3 \text{ cell L}^{-1}$ ; 91%) in June 2001, *Protoperidinium* sp. ( $552 \times 10^3 \text{ cell L}^{-1}$ ; 96%) in October 2001 and *Prorocentrum minimum* in March ( $312 \times 10^3 \text{ cell L}^{-1}$ ; 91%) and May 2002 ( $74 \times 10^3 \text{ cell L}^{-1}$ ; 75%) (Figure 3.3).

Dinophyceae abundances in the intermediate station oscillated between  $9 \times 10^3 \text{ cell L}^{-1}$  in July 2002 (13%) and  $874 \times 10^3 \text{ cell L}^{-1}$  in March 2002 (90%). Dinophyceae was the major phytoplankton class in June 2001 ( $353 \times 10^3 \text{ cell L}^{-1}$ ; 98%), October 2001 ( $172 \times 10^3 \text{ cell L}^{-1}$ ; 93%), January 2002 ( $260 \times 10^3 \text{ cell L}^{-1}$ ; 81%), March 2002 ( $874 \times 10^3 \text{ cell L}^{-1}$ ; 90%) and May 2002 ( $26 \times 10^3 \text{ cell L}^{-1}$ ; 79%). The most relevant *taxa* in June 2001 (*Gymnodinium* sp.:  $305 \times 10^3 \text{ cell L}^{-1}$ ; 85%), October 2001 (*Protoperidinium* sp.:  $171 \times 10^3 \text{ cell L}^{-1}$ ; 91%), March 2002 (*Prorocentrum minimum*:  $871 \times 10^3 \text{ cell L}^{-1}$ ; 90%) and May 2002 (*Prorocentrum minimum*:  $16 \times 10^3 \text{ cell L}^{-1}$ ; 48%) were the same as in the upstream station. In January 2002, *Gymnodinium* sp. ( $259 \times 10^3 \text{ cell L}^{-1}$ ) accounted 80% of the total phytoplankton abundance.

Downstream, Dinophyceae varied between  $9 \times 10^3 \text{ cell L}^{-1}$  in January 2002 (14%) and  $728 \times 10^3 \text{ cell L}^{-1}$  in March 2002 (99%), being the most represented class in June 2001 ( $43 \times 10^3 \text{ cell L}^{-1}$ ; 86%), October 2001 ( $128 \times 10^3 \text{ cell L}^{-1}$ ; 77%), December 2001 ( $72 \times 10^3 \text{ cell L}^{-1}$ ; 64%), March 2002 ( $728 \times 10^3 \text{ cell L}^{-1}$ ; 99%) and May 2002 ( $74 \times 10^3 \text{ cell L}^{-1}$ ; 50%). The *taxon* with greater relevance in June 2001 (*Prorocentrum minimum*:  $26 \times 10^3 \text{ cell L}^{-1}$ ; 53%) was different from the one found in the other stations (*Gymnodinium* sp.), but in October 2001 (*Protoperidinium* sp.:  $128 \times 10^3 \text{ cell L}^{-1}$ ; 77%), March 2002 (*Prorocentrum minimum*:  $728 \times 10^3 \text{ cell L}^{-1}$ ; 99%) and May 2002 (*Prorocentrum minimum*:  $67 \times 10^3 \text{ cell L}^{-1}$ ; 45%), the most abundant *taxa* downstream were the same as in the upstream and intermediate stations. In December 2001, the

dominance of Dinophyceae was due to *Gymnodinium* sp. ( $36 \times 10^3 \text{ cell L}^{-1}$ ) and *Scrippsiella trochoidea* ( $34 \times 10^3 \text{ cell L}^{-1}$ ), which accounted 32% and 30% of the total phytoplankton abundance.

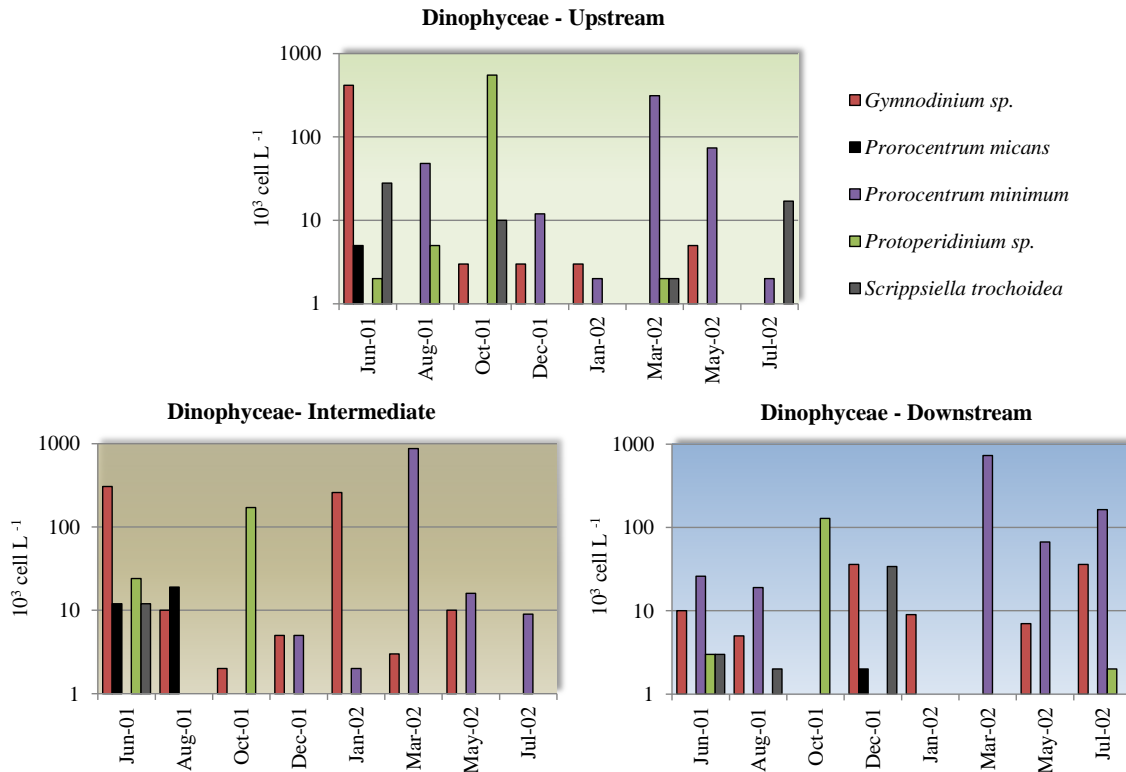


Figure 3.3 - Seasonal variation of Dinophyceae *taxa* abundance in Foz de Almagem sampling stations.

Bacillariophyceae abundances in the upstream station ranged from  $2 \times 10^3 \text{ cell L}^{-1}$  in January 2002 (4%) to  $71 \times 10^3 \text{ cell L}^{-1}$  in December 2001 (61%) and in July 2002 abundance was  $43 \times 10^3 \text{ cell L}^{-1}$ , accounting 66% of total phytoplankton abundance (Figure 3.2). *Fragilaria* spp. ( $59 \times 10^3 \text{ cell L}^{-1}$ ; 51%) were the most abundant diatoms in December 2001, while in July 2002 *Navicula* spp. ( $17 \times 10^3 \text{ cell L}^{-1}$ ; 26%) and *Cocconeis* sp. ( $12 \times 10^3 \text{ cell L}^{-1}$ ; 18%) represented the major *taxa* (Figure 3.4).

In the intermediate station, Bacillariophyceae minimum abundance was determined in June 2001 ( $5 \times 10^3 \text{ cell L}^{-1}$ ; 1%) and May 2002 ( $5 \times 10^3 \text{ cell L}^{-1}$ ; 16%), while the maximum abundance was registered in December 2001 ( $266 \times 10^3 \text{ cell L}^{-1}$ ; 88%). The most abundant diatoms in December 2001 were *Leptocylindrus danicus* ( $160 \times 10^3 \text{ cell L}^{-1}$ ; 53%) and *Cyclotella* spp. ( $69 \times 10^3 \text{ cell L}^{-1}$ ; 23%). In July 2002, diatoms abundance was much lower ( $57 \times 10^3 \text{ cell L}^{-1}$ ) than in December 2001 ( $266 \times 10^3 \text{ cell L}^{-1}$ ), nevertheless Bacillariophyceae dominated the phytoplankton community (85%), being

mostly represented by *Cocconeis* sp. ( $26 \times 10^3$  cell L<sup>-1</sup>; 38%) and *Cyclotella* spp. ( $12 \times 10^3$  cell L<sup>-1</sup>; 17%).

Downstream, Bacillariophyceae abundances went from  $2 \times 10^3$  cell L<sup>-1</sup> in June 2001 (3%) to  $279 \times 10^3$  cell L<sup>-1</sup> in July 2002 (58%). In August 2001 ( $85 \times 10^3$  cell L<sup>-1</sup>; 56%), January 2002 ( $48 \times 10^3$  cell L<sup>-1</sup>; 76%) and July 2002 ( $279 \times 10^3$  cell L<sup>-1</sup>; 58%), Bacillariophyceae was the most important phytoplankton class. *Cocconeis* sp. ( $60 \times 10^3$  cell L<sup>-1</sup>; 40%), *Diploneis* sp. ( $34 \times 10^3$  cell L<sup>-1</sup>; 55%) and *Fragilaria* spp. ( $200 \times 10^3$  cell L<sup>-1</sup>; 34%) were the most abundant diatoms in August 2001, January and July 2002, respectively.

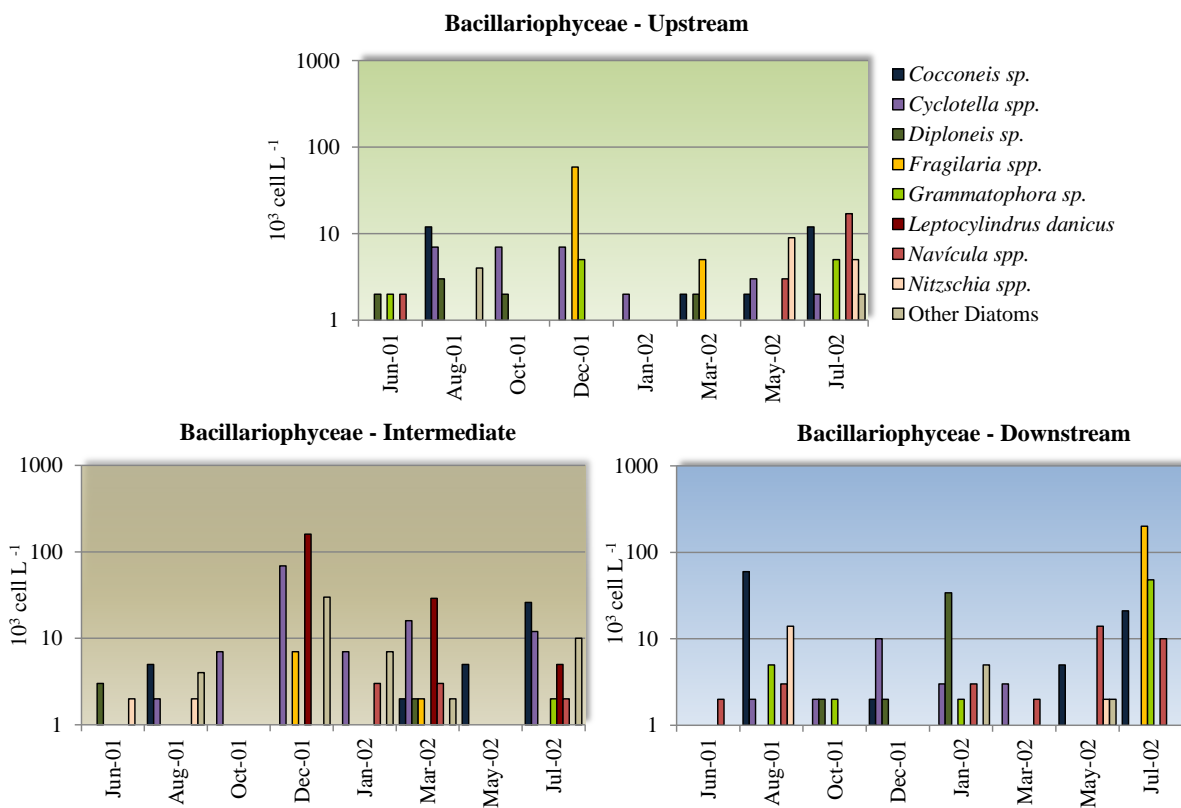


Figure 3.4 - Evolution of Bacillariophyceae *taxa* abundance in Foz de Almagem sampling stations.

The presence of Chlorophyceae, Cryptophyceae, Euglenophyceae and Cyanophyceae *taxa* was irregular and most of the time abundances were low in all stations (Figure 3.5). Chlorophyceae only occurred during December 2001 (upstream: *Cosmarinum* sp.; intermediate and downstream: *Staurastrum* sp.), with abundances under  $2 \times 10^3$  cell L<sup>-1</sup>. Cryptophyceae *taxa* were identified in all stations, generally presenting abundances lower than  $10 \times 10^3$  cell L<sup>-1</sup>, except in August and December 2001. *Rhodomonas* sp.

reached the maximum abundance upstream in August 2001 ( $26 \times 10^3 \text{ cell L}^{-1}$ ; 8%), while in the intermediate and downstream stations the highest Cryptophyceae abundances were determined for *Cryptomonas* sp. in December (12  $\times 10^3 \text{ cell L}^{-1}$ ; 4%) and August 2001 ( $38 \times 10^3 \text{ cell L}^{-1}$ ), respectively. *Cryptomonas* sp. contributed 25% to the total phytoplankton abundance downstream, in August 2001.

Euglenophyceae (*Eutreptiella* sp.) was observed in the three stations, but it was mostly represented downstream in December 2001, with an abundance of  $24 \times 10^3 \text{ cell L}^{-1}$ , which corresponded to 21% of the total phytoplankton downstream.

Cyanophyceae (*Anabaena flos-aqua*) was absent from the downstream station during the studied period, being found in the upstream station in March 2002 ( $16 \times 10^3 \text{ cell L}^{-1}$ ; 5%) and in the intermediate station in January ( $38 \times 10^3 \text{ cell L}^{-1}$ ; 12%) and March 2002 ( $32 \times 10^3 \text{ cell L}^{-1}$ ; 3%).

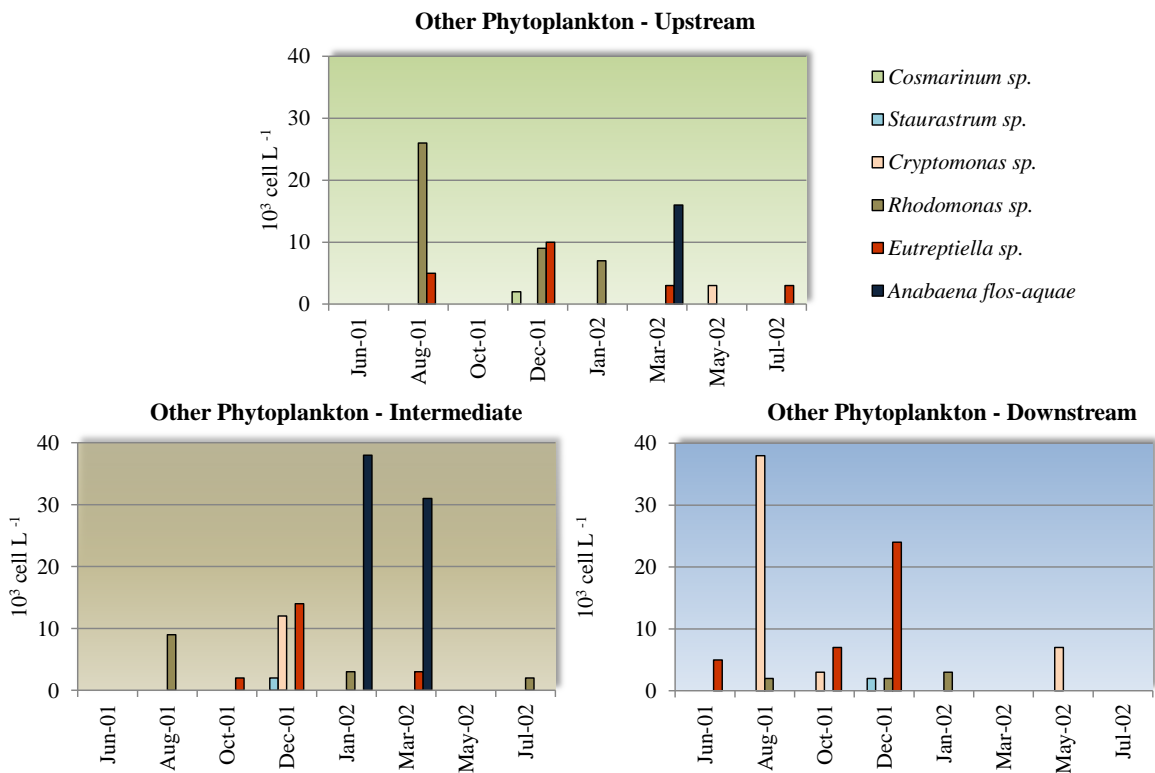


Figure 3.5 - Evolution of Chlorophyceae (*Cosmarinum* sp.; *Staurastrum* sp.), Cryptophyceae (*Cryptomonas* sp.; *Rhodomonas* sp.), Euglenophyceae (*Eutreptiella* sp.) and Cyanophyceae (*Anabaena flos-aqua*) taxa abundances in Foz de Almagem sampling stations.

The majority of phytoplankton classes did not present significant correlations between their abundances, just Cryptophyceae was positively correlated with pico-nano flagellate algae ( $\rho = 0.432$ ;  $p = 0.035$ ) and negatively correlated to Dinophyceae ( $\rho = -0.570$ ;  $p = 0.004$ ).

The upstream station presented lower annual means of Shannon-Wiener diversity, evenness, Bacillariophyceae abundance and higher mean abundance of pico-nano flagellate algae (Table 3.1). In the intermediate station, Cryptophyceae, Euglenophyceae and pico-nano flagellate algae showed the lowest mean abundances, while total phytoplankton abundance, taxonomic richness, Dinophyceae and Cyanophyceae abundances registered the greatest mean values.

The lowest means for total phytoplankton abundance, taxonomic richness and Dinophyceae abundance were determined in the downstream station. This station also presented the highest means for Shannon-Wiener diversity, evenness, Bacillariophyceae, Cryptophyceae, Euglenophyceae abundances.

Table 3.1 – Annual mean annual and standard deviation of phytoplankton parameters in Foz de Almagem sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
<b>Phytoplankton classes</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total phytoplankton abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	252.25 ± 199.43	293.08 ± 298.59	238.00 ± 241.40
Taxonomic richness	7.75 ± 2.12	8.38 ± 2.83	7.63 ± 1.92
Shannon-Wiener diversity (bits)	1.43 ± 0.90	1.51 ± 0.79	1.82 ± 0.80
Evenness	0.48 ± 0.27	0.51 ± 0.26	0.61 ± 0.25
Chlorophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	0.22 ± 0.61	0.22 ± 0.61	0.22 ± 0.61
Bacillariophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	22.63 ± 23.62	53.03 ± 88.53	57.56 ± 93.94
Cryptophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	5.60 ± 8.88	3.23 ± 4.65	6.90 ± 13.45
Dinophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	187.98 ± 223.37	216.86 ± 295.81	160.17 ± 237.47
Euglenophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	2.80 ± 3.68	2.37 ± 4.78	4.53 ± 8.40
Cyanophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	1.97 ± 5.56	8.74 ± 16.30	0.00 ± 0.00
Pico-nano flagellate algae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	31.04 ± 73.18	8.62 ± 18.89	9.05 ± 16.35

Although stations had different annual means for the studied phytoplankton parameters, the comparison performed with parametric and non-parametric tests did not reveal statistical significant differences among the mean values of the stations (Appendix I.G). These results might be explained by the high standard deviation values, which indicate a great oscillation through time and therefore, the variation within each station was greater than the variation among stations. Statistical tests were not applied to Chlorophyceae or Cyanophyceae because the number of occurrences was too low.

As described before in Chapter 2 (2.3.1.2. Water quality) chlorophyll *a* concentration in the lagoon was under 10 µg.L<sup>-1</sup> most of the year, except in March 2002 (Figure 2.5). The intermediate station presented the greatest seasonal variation (13.05 µg L<sup>-1</sup>) and

maximum chlorophyll *a* concentration (13.48  $\mu\text{g L}^{-1}$ ). Phaeo-pigments concentration was lower than chlorophyll *a*, except in May 2002. Pigment diversity index was similar in the three stations, presenting higher values in August 2001 (2.71) and March 2002 (2.74). Despite the variation of chlorophyll *a*, phaeo-pigments and pigments diversity in the three sampling stations, statistical analyses showed that there were no significant differences in the mean annual values of these parameters among stations (Appendix I.A).

No significant correlations were found between chlorophyll *a* concentration, phaeo-pigments concentration, pigment diversity and the total phytoplankton abundance, taxonomic richness, phytoplankton diversity and evenness. Only Bacillariophyceae abundance showed a positive correlation with pigment diversity ( $r = 0.480$ ;  $p = 0.018$ ).

### **3.3.1.2. Environmental parameters and phytoplankton communities**

No linear associations were determined between total phytoplankton abundance and environmental parameters ( $p > 0.05$ ), but Shannon-Wiener diversity, taxonomic richness and evenness were negatively correlated to cumulative rainfall. For Shannon-Wiener diversity and evenness, positive correlations were also found with pH (Table 3.2).

Bacillariophyceae and Cryptophyceae abundances were negatively correlated to cumulative rainfall. Cryptophyceae abundance also presented negative correlations with water level in the lagoon and nitrites concentration. Dinophyceae abundance showed a positive correlation with cumulative rainfall and a negative correlation with pH. A positive correlation was determined between Euglenophyceae abundance and dissolved oxygen concentration, while negative correlations were found with orthophosphates and total phosphorus concentrations.

The strongest and most significant correlations were determined between: 1) Cumulative rainfall vs. Shannon-Wiener diversity, evenness, taxonomic richness, Dinophyceae abundance and Bacillariophyceae abundance; 2) pH vs. Shannon-Wiener diversity.

Table 3.2 - Significant correlations between phytoplankton and environmental parameters in Foz de Almagem lagoon. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Phytoplankton parameters</i>	<i>Environmental parameters</i>	<i>Results</i>
Taxonomic richness	Cumulative rainfall	Rho = -0.619; $p = 0.001$ **
Shannon-Wiener diversity	Cumulative rainfall	Rho = -0.811; $p < 0.001$ **
	pH	Rho = 0.581; $p = 0.003$ **
Evenness	Cumulative rainfall	Rho = -0.634; $p = 0.001$ **
	pH	Rho = 0.440; $p = 0.031$ *
Bacillariophyceae abundance	Cumulative rainfall	Rho = -0.563; $p = 0.004$ **
Cryptophyceae abundance	Water level	Rho = -0.546; $p = 0.006$ **
	Cumulative rainfall	Rho = -0.482; $p = 0.017$ *
	Nitrites concentration	Rho = -0.500; $p = 0.013$ *
Dinophyceae abundance	Cumulative rainfall	Rho = 0.599; $p = 0.002$ **
	pH	Rho = -0.427; $p = 0.038$ *
Euglenophyceae abundance	Dissolved oxygen concentration	Rho = 0.415; $p = 0.044$ *
	Orthophosphates concentration	Rho = -0.433; $p = 0.035$ *
	Total phosphorus concentration	Rho = -0.457; $p = 0.025$ *

The multivariate approach given by the Canonical Correspondence Analysis (CCA) allowed the simultaneous incorporation of several environmental parameters and phytoplankton groups' abundances, all in a triplot diagram which simplified the interpretation of the relations between environmental parameters and phytoplankton abundances in the different sampling stations and months.

For the CCA (Figure 3.6) were included just the environmental variables that in the PCA (Figure 2.6) presented longest arrows and so, the most relevant for differentiating sampling stations, namely water level, total dissolved inorganic nitrogen concentration (DIN), salinity, total phosphorus and total solids in suspension concentration (TSS). The projections of the environmental vectors in the CCA were not very different from those derived from the PCA; however salinity showed greater correlation with axis II than in the PCA.

The Bacillariophyceae and Dinophyceae were placed near the origin of the ordination diagram, meaning that these phytoplankton groups were present in all samples and were not associated to any of these environmental variables. Euglenophyceae and Chlorophyceae seemed to be related to stations and months with lower DIN, salinity, total phosphorus and higher TSS. The increase of salinity, total phosphorus, TSS and the decrease of water level caused the rise of Cryptophyceae density. Cyanophyceae growth was stimulated by higher DIN, salinity, total phosphorus and lower TSS.

Pico-nano flagellate algae were positively associated with salinity and total phosphorus and negatively with water level. The relation between phytoplankton classes and environmental variables was highly significant (Monte Carlo test:  $p = 0.005$ ).

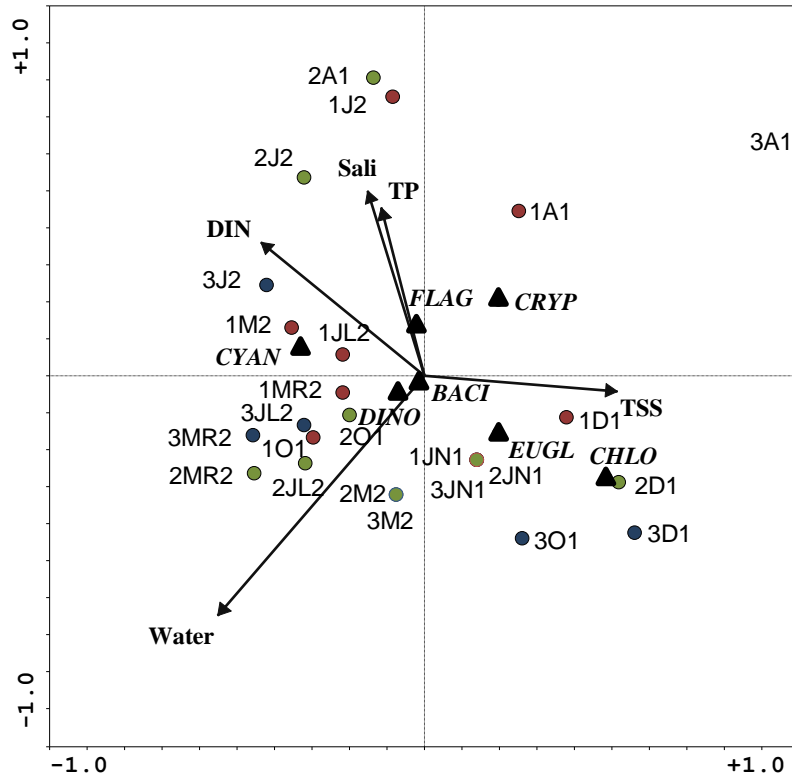


Figure 3.6 - Canonical correspondence analysis performed with the phytoplankton groups (total density *per* station) from Foz de Almargem sampling stations. Cumulative percentage variance explained by axes: Species - I = 16.9 % and I + II = 28.2 %; and Species-environment relation - I = 42.5 % and I + II = 70.8 %. Monte Carlo test of all canonical axes  $p = 0.005$

*Station codes:* First character corresponds to the sampling station (1- upstream, 2- intermediate, 3- downstream) and subsequent ones to month and year of survey (1- 2001, 2-2002).

*Environmental variables:* Water- water level in the lagoon; Sali- salinity; TSS- total solids in suspension; DIN- total dissolved inorganic nitrogen concentration; TP- total phosphorus concentration.

*Phytoplankton:* CRYP – Cryptophyceae; CHLO – Chlorophyceae; EUGL – Euglenophyceae; DINO – Dinophyceae; BACI – Bacillariophyceae; CYAN – Cyanophyceae; FLAG – Pico-nano flagellate algae (< 20  $\mu\text{m}$ ).

### 3.3.1.3. Phytoplankton communities during isolation and connection of the lagoon with the sea

The comparison of phytoplankton communities before and during the lagoon connection with the sea was done based on data from December 2001 and January 2002.

Before December, there was a first opening of the lagoon in the end of October and between December and January samplings, the lagoon opened for a second time.

All stations presented a decrease in Shannon-Wiener diversity, evenness, Chlorophyceae abundance and Euglenophyceae abundance and registered an increase in pico-nano flagellate algae abundance (Table 3.3). Chlorophyceae and Euglenophyceae were absent from all stations. Cyanophyceae were found only in the intermediate station, showing an augment in abundance. Total phytoplankton abundance and Dinophyceae abundance increased in the intermediate station and decreased in the other stations. There was a reduction in taxonomic richness in the upstream and intermediate stations, but the number of *taxa* in the intermediate and downstream stations was the same, with six *taxa* in common. Bacillariophyceae and Cryptophyceae abundances diminished in the upstream and intermediate stations, increasing downstream. The phytoplankton classes that presented greatest variations in abundance were the Bacillariophyceae ( $-248 \times 10^3 \text{ cell L}^{-1}$ ) and Dinophyceae ( $250 \times 10^3 \text{ cell L}^{-1}$ ) in the intermediate station, which meant a shift in the dominant taxonomic group.

Table 3.3 – Phytoplankton communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Foz de Almagem sampling stations.

	<i>Upstream station</i>		<i>Intermediate station</i>		<i>Downstream station</i>	
	Jan-02	Variation	Jan-02	Variation	Jan-02	Variation
<b>Phytoplankton communities</b>						
Total phytoplankton abundance ( $10^3 \text{ cell L}^{-1}$ )	43	<b>-72</b>	323	<b>19</b>	64	<b>-50</b>
Taxonomic richness	5	<b>-5</b>	9	<b>-2</b>	9	<b>0</b>
Shannon-Wiener diversity (bits)	1.49	<b>-1.01</b>	1.11	<b>-1.06</b>	2.26	<b>-0.08</b>
Evenness	0.64	<b>-0.11</b>	0.35	<b>-0.28</b>	0.71	<b>-0.03</b>
Chlorophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	0	<b>-2</b>	0	<b>-2</b>	0	<b>-2</b>
Bacillariophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	2	<b>-69</b>	17	<b>-248</b>	48	<b>34</b>
Cryptophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	7	<b>-2</b>	3	<b>-9</b>	3	<b>2</b>
Dinophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	5	<b>-10</b>	260	<b>250</b>	9	<b>-64</b>
Euglenophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	0	<b>-10</b>	0	<b>-14</b>	0	<b>-24</b>
Cyanophyceae abundance ( $10^3 \text{ cell L}^{-1}$ )	0	<b>0</b>	38	<b>38</b>	0	<b>0</b>
Pico-nano flagellate abundance ( $10^3 \text{ cell L}^{-1}$ )	29	<b>21</b>	3	<b>3</b>	3	<b>3</b>

Despite the variation between the two periods, when the lagoon was in connection with the sea, the upstream station had the greatest abundances of Cryptophyceae, pico-nano flagellate algae and the smallest abundances of Bacillariophyceae, Dinophyceae and values of taxonomic richness. In the intermediate station were found the highest values of total phytoplankton abundance, Dinophyceae abundances, Cyanophyceae abundances

and the lowest values of Shannon-Wiener diversity and evenness. Downstream, Bacillariophyceae abundances were higher, just as Shannon-Wiener diversity and evenness. Both intermediate and downstream stations presented the greater values for taxonomic richness and the smallest abundances for Cryptophyceae and pico-nano flagellate algae.

### **3.3.2. Salgados coastal lagoon**

#### **3.3.2.1. Phytoplankton communities**

In Salgados lagoon, 40 phytoplankton *taxa* were identified (Appendix II.B), 14 Chlorophyceae, 10 Cyanophyceae, 7 Bacillariophyceae, 5 Euglenophyceae, 2 Cryptophyceae, 1 Dinophyceae and pico-nano flagellate algae (< 20 µm). Only a *taxon* was present in more than 50% of the samples (*Cyclotella* spp. accounting 75% of the samples). Other *taxa* were identified in more than 25% of the samples, namely *Planktothrix* sp., *Navicula* spp., *Rhodomonas* sp., *Gymnodinium* sp., *Anabaena spiroides*, *Microcystis aeruginosa* and *Scenedesmus opoliensis*. The cyanobacteria *Microcystis aeruginosa* was the most abundant species, accomplishing 75 % of the total phytoplankton cells sampled.

Taxonomic richness in the stations varied from three (upstream: March 2002; downstream: June 2001, December 2001, May 2002) to 19 *taxa* (upstream: July 2002) and most of the time stations presented a similar tendency of evolution, except the upstream station in August 2001, March and May 2002 (Figure 3.7). From June to October 2001, the number of *taxa* increased in the intermediate and downstream stations, but in December values went down to the same numbers as in June 2001 and till May 2002 there was some irregularity in stations tendencies. In July 2002, all stations showed a considerable increase. Along the year, the upstream station had a greater variation in the number of *taxa* (16) than the intermediate (12) and the downstream (9) stations. Taxonomic richness was positively and highly correlated with total phytoplankton abundance ( $r = 0.732$ ;  $p < 0.001$ ), Chlorophyceae abundance ( $\rho = 0.759$ ;  $p < 0.001$ ), Bacillariophyceae abundance ( $\rho = 0.675$ ;  $p < 0.001$ ),

Dinophyceae abundance ( $\rho = 0.714$ ;  $p < 0.001$ ), Euglenophyceae abundance ( $\rho = 0.568$ ;  $p = 0.004$ ) and Cyanophyceae abundance ( $r = 0.581$ ;  $p = 0.003$ ).

Total phytoplankton lowest and highest abundances were found in the upstream station, during December 2001 ( $169 \times 10^3 \text{ cell L}^{-1}$ ) and July 2002 ( $621066 \times 10^3 \text{ cell L}^{-1}$ ), respectively. All stations followed a similar seasonal trend, increasing in August 2001, January and July 2002. June and December 2001 were the months with lower values, while in August 2001 and July 2002 were observed the higher values of total phytoplankton abundance in the three stations.

The upstream station showed the greatest variation in time ( $620897 \times 10^3 \text{ cell L}^{-1}$ ), followed by the intermediate ( $157863 \times 10^3 \text{ cell L}^{-1}$ ) and the downstream ( $126974 \times 10^3 \text{ cell L}^{-1}$ ) stations.

Total phytoplankton abundance was negatively correlated to evenness ( $r = -0.629$ ;  $p = 0.001$ ) and positively correlated to Cyanophyceae abundance ( $r = 0.919$ ;  $p < 0.001$ ), taxonomic richness ( $r = 0.732$ ;  $p < 0.001$ ), Bacillariophyceae abundance ( $\rho = 0.733$ ;  $p < 0.001$ ), Chlorophyceae abundance ( $\rho = 0.679$ ;  $p < 0.001$ ), Dinophyceae abundance ( $\rho = 0.526$ ;  $p = 0.008$ ) and Euglenophyceae abundance ( $\rho = 0.452$ ;  $p = 0.026$ ).

In January 2002 were determined the lowest values of Shannon-Wiener diversity index (upstream:  $H' = 0.13$ ; intermediate:  $H' = 0.12$ ) and evenness (upstream:  $E = 0.05$ ; intermediate:  $E = 0.06$ ), due to the dominance of *Microcystis aeruginosa*, which represented 99% of the total phytoplankton cells. The maximum values of Shannon-Wiener diversity ( $H' = 2.82$ ) and evenness ( $E = 0.95$ ) were found in October 2001 (intermediate) and March 2002 (upstream), respectively. During the studied period, the lowest variation in diversity and evenness was observed downstream ( $H' = 1.59$ ;  $E = 0.61$ ) and greater variations were determined in the upstream ( $H' = 2.53$ ;  $E = 0.90$ ) and intermediate stations ( $H' = 2.70$ ;  $E = 0.67$ ). No regular patterns were found in the seasonal variation of diversity or evenness from the stations.

Shannon-Wiener diversity was positively correlated to evenness ( $r = 0.849$ ;  $p < 0.001$ ), while evenness had a negative correlation with total phytoplankton abundance ( $r = -0.629$ ;  $p = 0.001$ ) and with Cyanophyceae abundance ( $r = -0.601$ ;  $p = 0.002$ ).

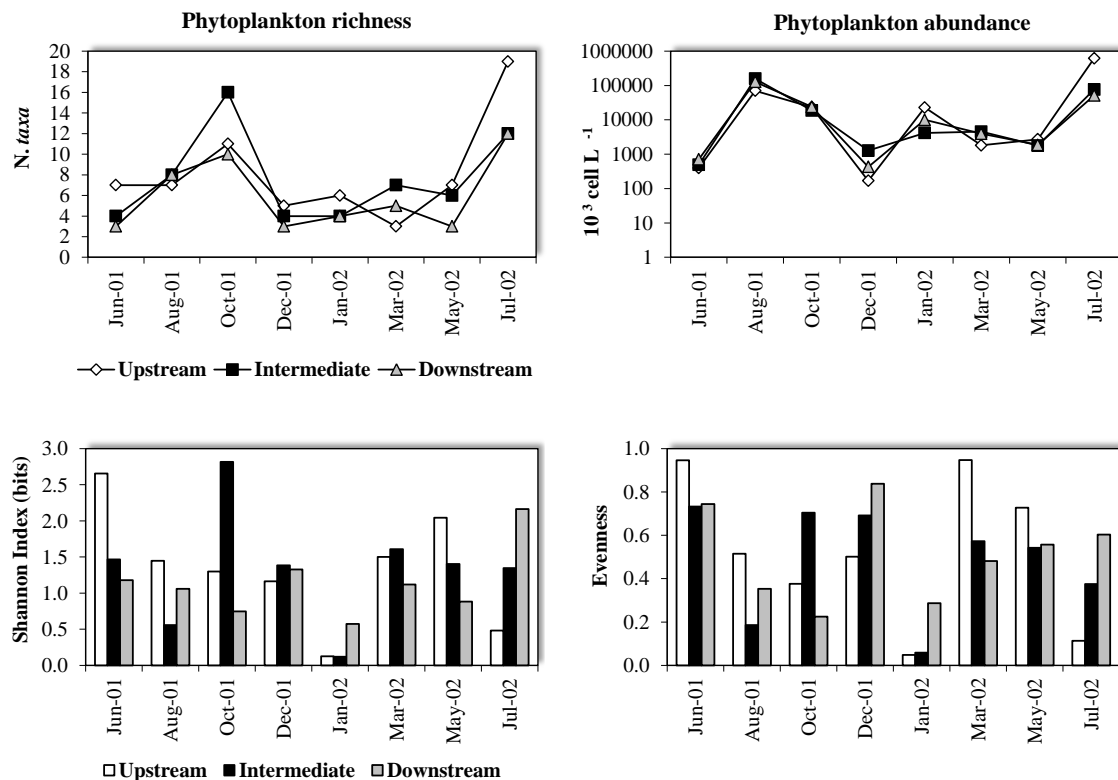


Figure 3.7 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Salgados sampling stations.

Most of the time, Cyanophyceae dominated the phytoplankton communities of the stations, with a few exceptions. Bacillariophyceae became the main taxonomic group in December 2001; Chlorophyceae was the major class upstream in June 2001 and downstream in July 2002; pico-nano flagellate algae accounted a high percentage of the intermediate and downstream phytoplankton in March and May 2002 (Figure 3.8).

Pico-nano flagellate algae were observed in the upstream station just in August 2001 ( $13797 \times 10^3 \text{ cell L}^{-1}$ ) and May 2002 ( $655 \times 10^3 \text{ cell L}^{-1}$ ), being the second major group (after Cyanophyceae) and accounting 20% and 24% of total phytoplankton determined in each of the months.

In the intermediate station, pico-nano flagellate algae occurred in March ( $2426 \times 10^3 \text{ cell L}^{-1}$ ) and May 2002 ( $1130 \times 10^3 \text{ cell L}^{-1}$ ), dominating the communities with 53% and 63% of total phytoplankton, respectively.

Downstream, just as in the intermediate station, pico-nano flagellate algae were identified only in March 2002 ( $1661 \times 10^3 \text{ cell L}^{-1}$ ) and May 2002 ( $1513 \times 10^3 \text{ cell L}^{-1}$ ), being the most abundant group in March (42%) jointly with Cyanophyceae and dominating the phytoplankton community in May 2002 (81%).

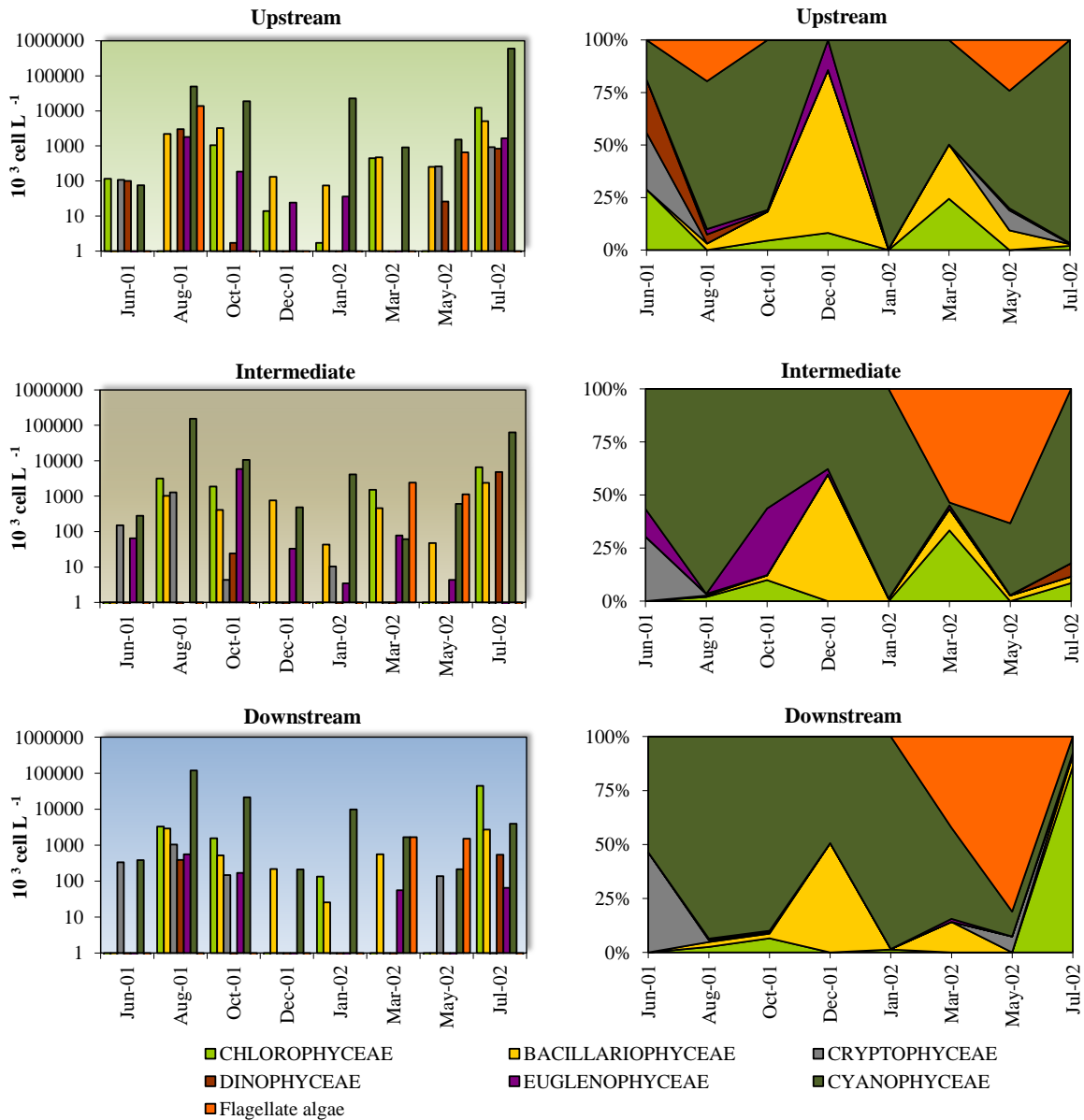


Figure 3.8 - Evolution of phytoplankton abundance *per class* and relative frequency of each class in Salgados sampling stations.

In the upstream station, Cyanophyceae was present in all samplings except December 2001. During the remaining period, abundances went from  $75 \times 10^3 \text{ cell L}^{-1}$  in June 2001 (19% of total phytoplankton abundance) to  $600263 \times 10^3 \text{ cell L}^{-1}$  in July 2002 (97%), showing high values also in August 2001 ( $49581 \times 10^3 \text{ cell L}^{-1}$ ; 70%), October 2001 ( $18970 \times 10^3 \text{ cell L}^{-1}$ ; 81%) and January 2002 ( $22676 \times 10^3 \text{ cell L}^{-1}$ ; 99%). Cyanophyceae abundance was lower in March 2002 ( $905 \times 10^3 \text{ cell L}^{-1}$ ) and May 2002 ( $1522 \times 10^3 \text{ cell L}^{-1}$ ), nevertheless it was the major phytoplankton class in these months (March: 50%; May: 56%). The most important *taxa* were *Microcystis aeruginosa*, in August 2001 ( $48288 \times 10^3 \text{ cell L}^{-1}$ ; 69%), January 2002 ( $22470 \times 10^3 \text{ cell L}^{-1}$ ; 99%) and

July 2002 ( $589619 \times 10^3 \text{ cell L}^{-1}$ ; 95%) and *Anabaena spiroides* ( $17461 \times 10^3 \text{ cell L}^{-1}$ ) in October 2001, accounting 74% of total phytoplankton (Figure 3.9).

In the intermediate station, Cyanophyceae abundances varied between  $61 \times 10^3 \text{ cell L}^{-1}$  in March 2002 (1% of total phytoplankton abundance) and  $152912 \times 10^3 \text{ cell L}^{-1}$  in August 2001 (97%). High abundances were also observed in October 2001 ( $10554 \times 10^3 \text{ cell L}^{-1}$ ; 57%), January 2002 ( $4108 \times 10^3 \text{ cell L}^{-1}$ ; 99%), July 2002 ( $62885 \times 10^3 \text{ cell L}^{-1}$ ; 82%) and during these months, Cyanophyceae dominated the phytoplankton communities. In June 2001, although abundance was lower ( $280 \times 10^3 \text{ cell L}^{-1}$ ), it was also the predominant phytoplankton class (57%), due to the occurrence of *Planktothrix* sp. During August and October 2001, *Planktothrix* sp. had higher abundances than in June 2001 ( $7761 \times 10^3 \text{ cell L}^{-1}$  and  $1716 \times 10^3 \text{ cell L}^{-1}$ ), but other *taxa* were more relevant, namely *Microcystis aeruginosa* in August ( $145151 \times 10^3 \text{ cell L}^{-1}$ ; 92%) and *Anabaena spiroides* ( $4109 \times 10^3 \text{ cell L}^{-1}$ ; 22%), *Chroococcus limneticus* ( $2897 \times 10^3 \text{ cell L}^{-1}$ ; 16%) and *Lyngbya* sp. ( $1832 \times 10^3 \text{ cell L}^{-1}$ ; 10%) in October. *Microcystis aeruginosa* was the only Cyanophyceae *taxon* present in January 2002 ( $4108 \times 10^3 \text{ cell L}^{-1}$ ; 99%), and in July 2002 besides *Microcystis aeruginosa* ( $60503 \times 10^3 \text{ cell L}^{-1}$ ; 79%) two other *taxa* occurred, *Merismopedia punctate* ( $1836 \times 10^3 \text{ cell L}^{-1}$ ; 2%) and *Anabaena spiroides* ( $546 \times 10^3 \text{ cell L}^{-1}$ ;  $\approx 1\%$ ). The most important *taxa* in August 2001 (*Microcystis aeruginosa*), October 2001 (*Anabaena spiroides*), January 2002 (*Microcystis aeruginosa*) and July 2002 (*Microcystis aeruginosa*) were the same as in the upstream station.

At the downstream station, Cyanophyceae were more abundant during August 2001 ( $119254 \times 10^3 \text{ cell L}^{-1}$ ), October 2001 ( $21471 \times 10^3 \text{ cell L}^{-1}$ ) and January 2002 ( $9854 \times 10^3 \text{ cell L}^{-1}$ ), representing 94%, 90% and 98% of the total phytoplankton determined in these months. Just as in the intermediate station during June 2001, Cyanophyceae abundance was lower ( $384 \times 10^3 \text{ cell L}^{-1}$ ) but still accounted 54% of the phytoplankton. In December 2001 ( $212 \times 10^3 \text{ cell L}^{-1}$ ) and March 2002 ( $1661 \times 10^3 \text{ cell L}^{-1}$ ), Cyanophyceae represented 49 % and 42% of the phytoplankton communities. *Anabaena flos-aqua* was the only species identified in June 2001 ( $384 \times 10^3 \text{ cell L}^{-1}$ ; 54%). Three species occurred in August 2001, *Microcystis aeruginosa* ( $102914 \times 10^3 \text{ cell L}^{-1}$ ; 81%), *Planktothrix* sp. ( $14874 \times 10^3 \text{ cell L}^{-1}$ ; 12%) and *Anabaena flos-aqua* ( $1466 \times 10^3 \text{ cell L}^{-1}$ ;  $\approx 1\%$ ). In October 2001 there was only *Anabaena spiroides* ( $21471 \times 10^3 \text{ cell L}^{-1}$ ; 90%), while in January 2002 two *taxa* were observed, *Microcystis aeruginosa* ( $8922 \times 10^3 \text{ cell L}^{-1}$ ; 89%) and *Chroococcus limneticus* ( $932 \times 10^3 \text{ cell L}^{-1}$ ; 9%). During

March 2002, unidentified coccoid Cyanophyceae accounted 42% ( $1661 \times 10^3 \text{ cell L}^{-1}$ ) of the total phytoplankton abundance. Although Cyanophyceae accounted a small percentage of total phytoplankton in July 2001 ( $3913 \times 10^3 \text{ cell L}^{-1}$ ; 7%), *Microcystis aeruginosa* ( $2230 \times 10^3 \text{ cell L}^{-1}$ ; 4%) was the most abundant Cyanophyceae species. *Lyngbya* sp. ( $1683 \times 10^3 \text{ cell L}^{-1}$ ; 3%) was the second major *taxa* observed in this month.

The most relevant species in August 2001, January 2002, July 2002 (*Microcystis aeruginosa*) and in October 2001 (*Anabaena spiroides*) were the same in the three sampling stations.

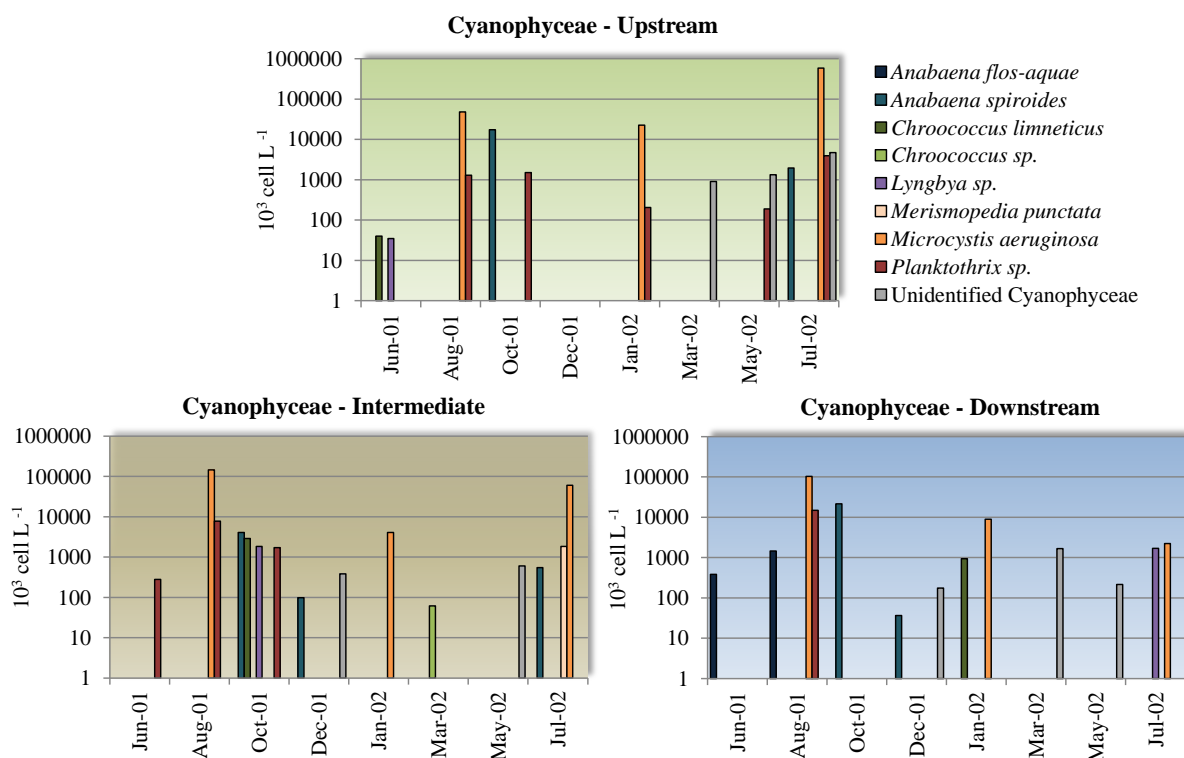


Figure 3.9 - Evolution of Cyanophyceae *taxa* abundance in Salgados sampling stations.

Bacillariophyceae were absent from the upstream station in June 2001 and from August 2001 till July 2002 abundances ranged from  $74 \times 10^3 \text{ cell L}^{-1}$  in January 2002 (< 1%) to  $5066 \times 10^3 \text{ cell L}^{-1}$  in July 2002 ( $\approx$  1%). Although Bacillariophyceae abundance was lower in December 2001 ( $131 \times 10^3 \text{ cell L}^{-1}$ ), it dominated the phytoplankton community (78%). In March 2002, Bacillariophyceae abundance ( $470 \times 10^3 \text{ cell L}^{-1}$ ) represented 26% of the total phytoplankton. *Cyclotella* spp. were the only Bacillariophyceae *taxa* present in the station during December 2001 ( $131 \times 10^3 \text{ cell L}^{-1}$ ; 78%) and March 2002 ( $470 \times 10^3 \text{ cell L}^{-1}$ ; 26%). In July 2002, three *taxa* were

observed: *Cyclotella* spp. ( $3471 \times 10^3 \text{ cell L}^{-1}$ ; < 1%), *Navicula* spp. ( $1574 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) and *Nitzschia* sp. ( $22 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) (Figure 3.10).

In the intermediate station, Bacillariophyceae occurred from August 2001 till July 2002, being absent in June 2001, just as it happened in the upstream station. In January and May 2002 were determined the lower abundances,  $43 \times 10^3 \text{ cell L}^{-1}$  (1%) and  $47 \times 10^3 \text{ cell L}^{-1}$  (3%) respectively, while the highest abundance was observed in July 2002 ( $2372 \times 10^3 \text{ cell L}^{-1}$ ; 3%). Bacillariophyceae dominated the phytoplankton community in December 2001 (60%), with the *taxa Cyclotella* spp. ( $764 \times 10^3 \text{ cell L}^{-1}$ ). In July 2002, besides *Cyclotella* spp. ( $2113 \times 10^3 \text{ cell L}^{-1}$ ; 3%), just *Navicula* spp. ( $259 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) occurred.

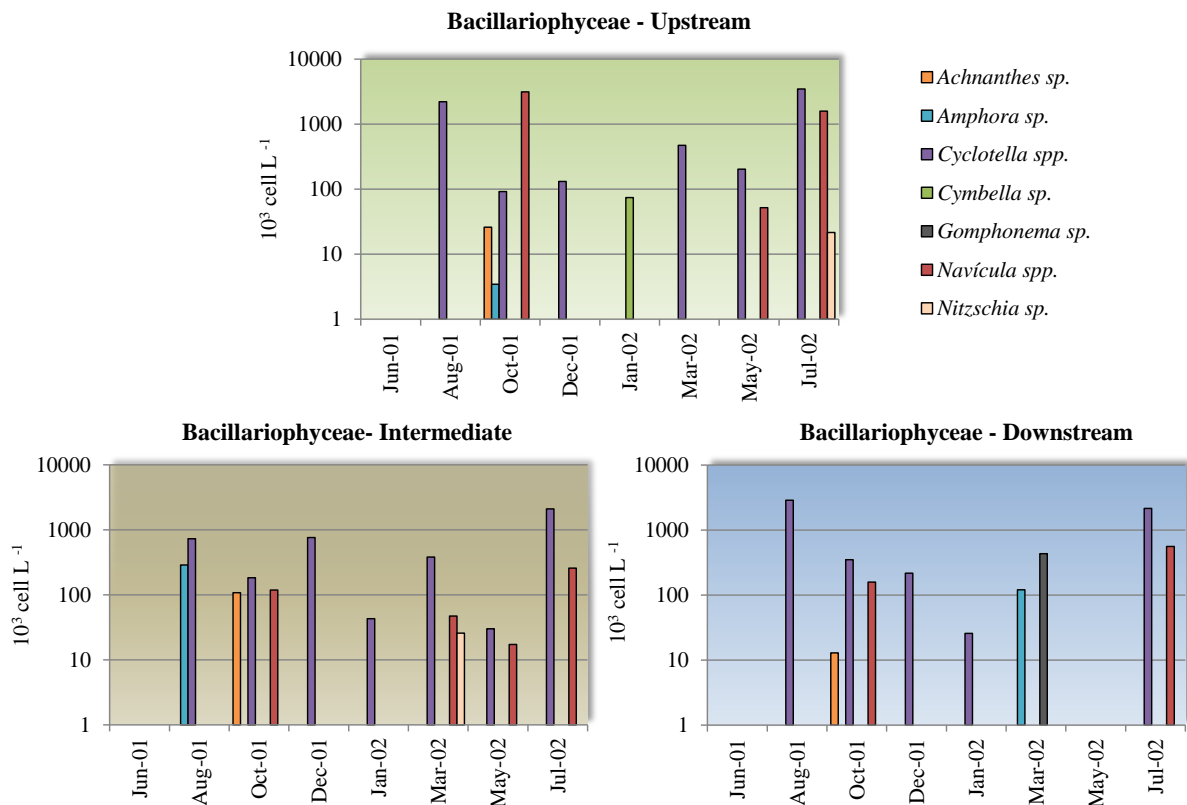


Figure 3.10 - Evolution of Bacillariophyceae *taxa* abundance in Salgados sampling stations.

Bacillariophyceae were observed downstream from August 2001 to March 2002 and in July 2002, presenting the higher abundances in August 2001 ( $2889 \times 10^3 \text{ cell L}^{-1}$ ; 2%) and July 2002 ( $2716 \times 10^3 \text{ cell L}^{-1}$ ; 5%). Abundances decreased from August till January 2002 ( $26 \times 10^3 \text{ cell L}^{-1}$ ) and increased in March and July 2002. In December 2001, Bacillariophyceae represented 51% of the phytoplankton community ( $217 \times 10^3 \text{ cell L}^{-1}$ ) and in March 2002 accounted 14% ( $556 \times 10^3 \text{ cell L}^{-1}$ ). *Cyclotella* spp. were the

only diatoms identified in August ( $2889 \times 10^3 \text{ cell L}^{-1}$ ; 2%) and December 2001 ( $217 \times 10^3 \text{ cell L}^{-1}$ ; 51%), while in July 2002, besides *Cyclotella* spp. ( $2156 \times 10^3 \text{ cell L}^{-1}$ ; 4%), *Navicula* spp. ( $560 \times 10^3 \text{ cell L}^{-1}$ ; 1%) were also found.

No Chlorophyceae *taxa* were observed upstream in August 2001, January and May 2002. In December 2001, the abundance was  $14 \times 10^3 \text{ cell L}^{-1}$  (8 %) and the maximum value was obtained in July 2002 ( $12309 \times 10^3 \text{ cell L}^{-1}$ ; 2%). Despite abundances in June 2001 ( $115 \times 10^3 \text{ cell L}^{-1}$ ) and March 2002 ( $444 \times 10^3 \text{ cell L}^{-1}$ ) were not as high as in July 2002, Chlorophyceae accounted 29% and 24% respectively, of the total phytoplankton abundance. In June 2001, only two species were identified *Crucigenia quadrata* ( $101 \times 10^3 \text{ cell L}^{-1}$ ; 25%) and *Ankistrodesmus acicularis* ( $14 \times 10^3 \text{ cell L}^{-1}$ ; 4%), while in March 2002 there was only one Chlorophyceae species, *Geminella interrupta* ( $444 \times 10^3 \text{ cell L}^{-1}$ ; 24%) (Figure 3.11). During the peak abundance of Chlorophyceae in July 2002, eight *taxa* were registered (*Ankistrodesmus falcatus*, *Ankistrodesmus acicularis*, *Kirchneriella lunaris*, *Scenedesmus acutus*, *Scenedesmus acuminatus*, *Scenedesmus opoliensis*, *Kirchneriella obesa*, *Coelastrum microporum*), with abundances varying from  $561 \times 10^3 \text{ cell L}^{-1}$  (*Ankistrodesmus falcatus*) to  $3104 \times 10^3 \text{ cell L}^{-1}$  (*Coelastrum microporum*).

In the intermediate station, Chlorophyceae were present in August and October 2001, March and July 2002, with abundances oscillating from  $1513 \times 10^3 \text{ cell L}^{-1}$  in March 2002 (33%) to  $6510 \times 10^3 \text{ cell L}^{-1}$  in July 2002 (9%). In March 2002, Chlorophyceae was the second major group (after Pico-nano flagellate algae), due to the abundance of *Geminella interrupta* ( $1513 \times 10^3 \text{ cell L}^{-1}$ ), which accounted 33% of total phytoplankton. In the intermediate station, during July 2002 were observed six of the species identified in this month upstream (*Ankistrodesmus falcatus*, *Scenedesmus acutus*, *Coelastrum microporum*, *Scenedesmus opoliensis*, *Scenedesmus acuminatus*, *Kirchneriella obesa*) and species abundances went from  $86 \times 10^3 \text{ cell L}^{-1}$  (*Ankistrodesmus falcatus*) to  $2113 \times 10^3 \text{ cell L}^{-1}$  (*Kirchneriella obesa*).

Chlorophyceae occurred downstream during August and October 2001, January and July 2002, dominating the phytoplankton community in July 2002 (86%), when the maximum abundance was determined ( $44300 \times 10^3 \text{ cell L}^{-1}$ ). In August and October 2001, there was only one *taxon* *Oocystis lacustris*, which presented abundances of  $3277 \times 10^3 \text{ cell L}^{-1}$  (3%) and  $798 \times 10^3 \text{ cell L}^{-1}$  (3%), respectively. *Geminella interrupta* occurred in January 2002 ( $135 \times 10^3 \text{ cell L}^{-1}$ ; 1%), and in July 2002 were identified six

species (*Selenastrum gracile*, *Oocystis lacustris*, *Coelastrum microporum*, *Kirchneriella obesa*, *Scenedesmus acutus*, *Scenedesmus opoliensis*), four of which were common with the other stations (*Coelastrum microporum*, *Kirchneriella obesa*, *Scenedesmus acutus*, *Scenedesmus opoliensis*). *Selenastrum gracile* was the species with lower abundance ( $237 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) and *Scenedesmus opoliensis* had the higher abundance ( $29921 \times 10^3 \text{ cell L}^{-1}$ ; 58%).

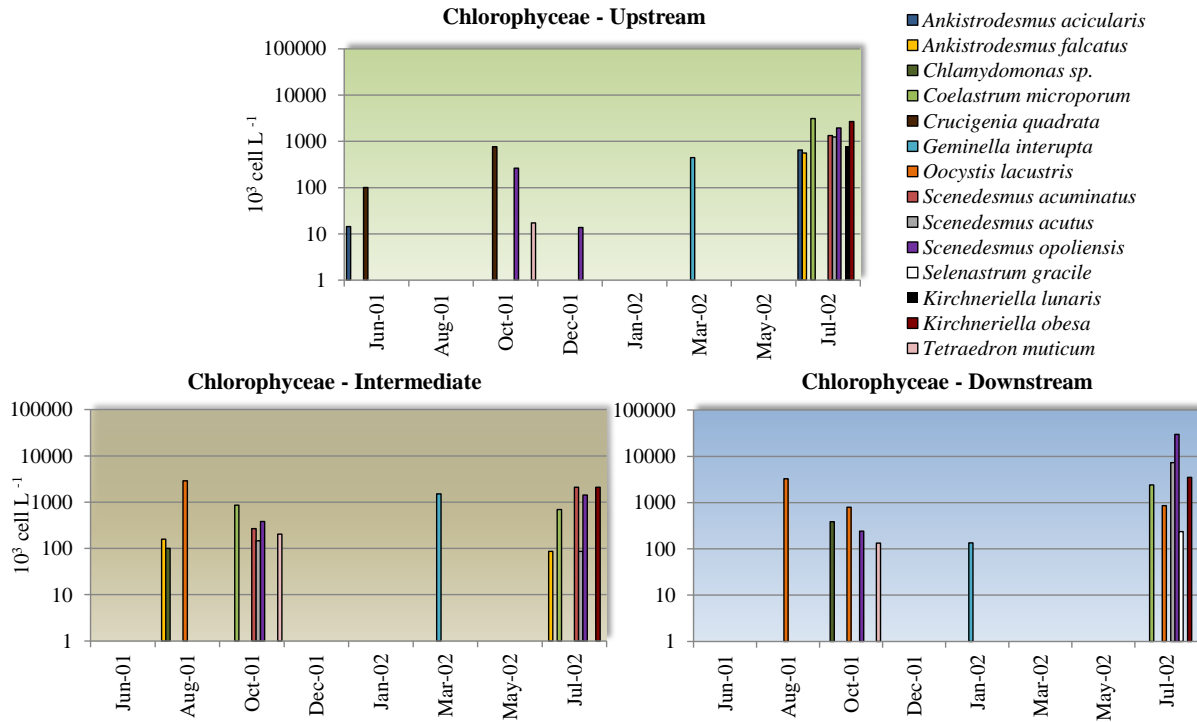


Figure 3.11 - Evolution of Chlorophyceae taxa abundance in Salgados sampling stations.

In the upstream station, Euglenophyceae was observed from August 2001 to January 2002 and in July 2002. During this period, the lowest abundance was registered in December 2001 ( $24 \times 10^3 \text{ cell L}^{-1}$ ), nevertheless it represented 14% of total phytoplankton abundance. The higher abundances were determined in August 2001 ( $1811 \times 10^3 \text{ cell L}^{-1}$ ) and July 2002 ( $1660 \times 10^3 \text{ cell L}^{-1}$ ), but in these months Euglenophyceae only accounted 3% and less than 1% of total phytoplankton. Two species were identified in August 2001, *Euglena oblonga* ( $1423 \times 10^3 \text{ cell L}^{-1}$ ; 2%) and *Euglena acus* ( $388 \times 10^3 \text{ cell L}^{-1}$ ;  $\approx$  1%) (Figure 3.12). In July 2002, *Euglena oblonga* ( $1056 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) and *Phacus acuminatus* ( $604 \times 10^3 \text{ cell L}^{-1}$ ; < 1%) were the only species observed and just as in August 2001, *Euglena oblonga* was the most abundant species. During December 2001, three taxa occurred, *Euglena oblonga* ( $5 \times$

$10^3$  cell L<sup>-1</sup>; 3%), *Phacus acuminatus* ( $16 \times 10^3$  cell L<sup>-1</sup>; 9%) and *Trachelomonas* sp. ( $3 \times 10^3$  cell L<sup>-1</sup>; 2%).

Euglenophyceae were absent from the intermediate station in August 2001 and July 2002. During January and May 2002, abundances were low ( $3 \times 10^3$  cell L<sup>-1</sup> and  $4 \times 10^3$  cell L<sup>-1</sup>), but in October 2001 the maximum of  $5881 \times 10^3$  cell L<sup>-1</sup> was achieved, representing 31% of the total phytoplankton. Two species occurred in October 2001, *Euglena oblonga* ( $5648 \times 10^3$  cell L<sup>-1</sup>; 30%) and *Phacus acuminatus* ( $233 \times 10^3$  cell L<sup>-1</sup>; 1%).

The occurrence of Euglenophyceae downstream was less regular than in the other stations, being present in August and October 2001, March and July 2002. During these months, the lowest abundances were determined in March 2002 ( $56 \times 10^3$  cell L<sup>-1</sup>; 1%) and July 2002 ( $65 \times 10^3$  cell L<sup>-1</sup>; < 1%). The maximum abundance was observed in August 2001 ( $560 \times 10^3$  cell L<sup>-1</sup>; < 1%), and in October 2001 Euglenophyceae abundance was  $168 \times 10^3$  cell L<sup>-1</sup> (< 1%). *Phacus acuminatus* was the species identified in August 2001, while in the other months *Euglena acus* occurred.

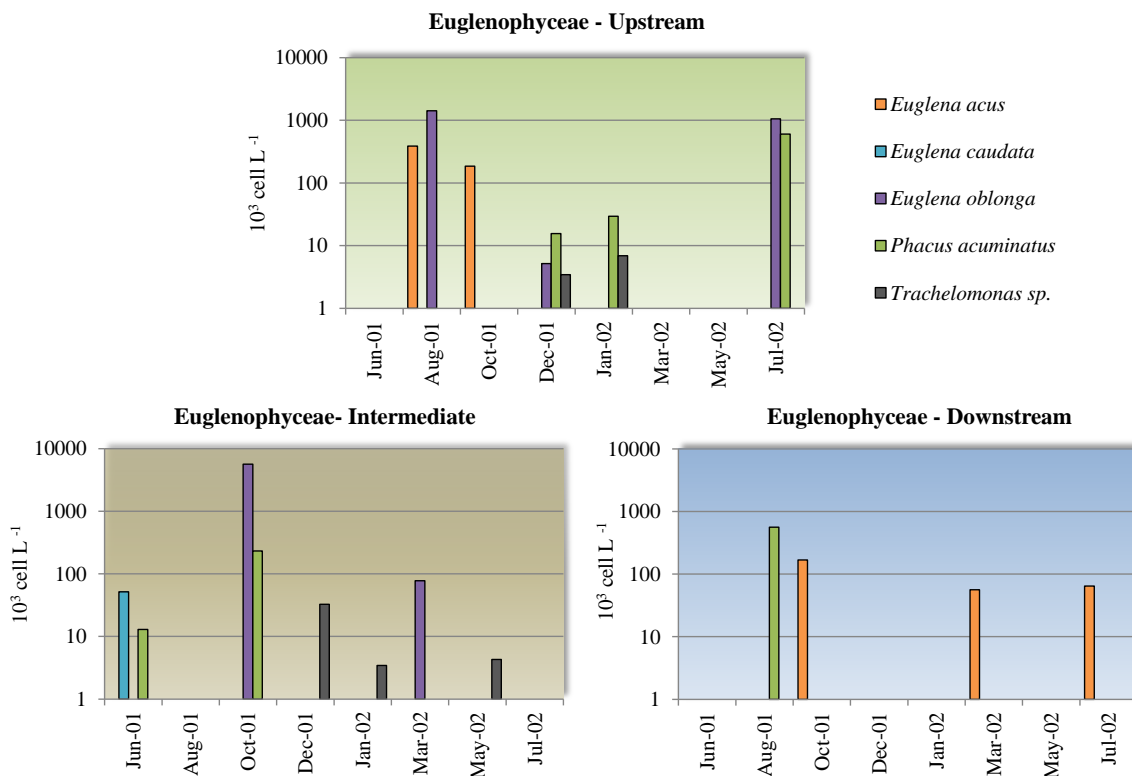


Figure 3.12 - Evolution of Euglenophyceae taxa abundance in Salgados sampling stations.

Cryptophyceae occurred in the upstream station just in June 2001 ( $107 \times 10^3$  cell L<sup>-1</sup>; 27%), May 2002 ( $259 \times 10^3$  cell L<sup>-1</sup>; 10%) and July 2002 ( $927 \times 10^3$  cell L<sup>-1</sup>; < 1%),

being mostly represented by *Rhodomonas* sp., which accounted 26%, 10% and less than 1% of the total phytoplankton abundance in these months (Figure 3.8 and 3.13). *Cryptomonas* sp. only occurred in June 2001 ( $4 \times 10^3$  cell L<sup>-1</sup>; 1%).

In the intermediate station, Cryptophyceae were observed from June to October 2001 and in January 2002. The maximum abundance was determined in August 2001 ( $1279 \times 10^3$  cell L<sup>-1</sup>; < 1%), but it was in June 2001 ( $151 \times 10^3$  cell L<sup>-1</sup>) that this class accounted a greater percentage of total phytoplankton (30%). In the two months, *Rhodomonas* sp. was the only *taxa* identified.

The presence of Cryptophyceae downstream was registered from June to October 2001 and in May 2002, with lower abundances occurring in October 2001 ( $148 \times 10^3$  cell L<sup>-1</sup>; < 1%) and May 2002 ( $138 \times 10^3$  cell L<sup>-1</sup>; 7%) and the maximum abundance being observed in August 2001 ( $1035 \times 10^3$  cell L<sup>-1</sup>; < 1%). Nevertheless, it was in June 2001 that Cryptophyceae ( $332 \times 10^3$  cell L<sup>-1</sup>) accounted the greater percentage of the phytoplankton community (47%). *Rhodomonas* sp. were present in the four months. In June 2001, besides *Rhodomonas* sp. ( $306 \times 10^3$  cell L<sup>-1</sup>; 43%), *Cryptomonas* sp. ( $26 \times 10^3$  cell L<sup>-1</sup>; 4%) were also identified.

Dinophyceae were absent upstream from December 2001 to March 2002 and during the other months, abundances oscillated from  $2 \times 10^3$  cell L<sup>-1</sup> in October 2001 (< 1% of total phytoplankton) to  $3018 \times 10^3$  cell L<sup>-1</sup> in August 2001 (4%).

*Gymnodinium* sp. was the only *taxa* identified. In June 2001, *Gymnodinium* sp. abundance ( $101 \times 10^3$  cell L<sup>-1</sup>) represented 25% of total phytoplankton and in August 2001 ( $3018 \times 10^3$  cell L<sup>-1</sup>) and July 2002 ( $841 \times 10^3$  cell L<sup>-1</sup>), although abundances were higher it only accounted 4 % and less than 1% of total phytoplankton, respectively (Figure 3.13).

In the intermediate station, Dinophyceae (*Gymnodinium* sp.) were observed only in October 2001 ( $24 \times 10^3$  cell L<sup>-1</sup>; < 1%) and July 2002 ( $4807 \times 10^3$  cell L<sup>-1</sup>; 6%).

Downstream, Dinophyceae (*Gymnodinium* sp.) were identified just in August 2001 ( $388 \times 10^3$  cell L<sup>-1</sup>; < 1%) and July 2002 ( $539 \times 10^3$  cell L<sup>-1</sup>; 1%).

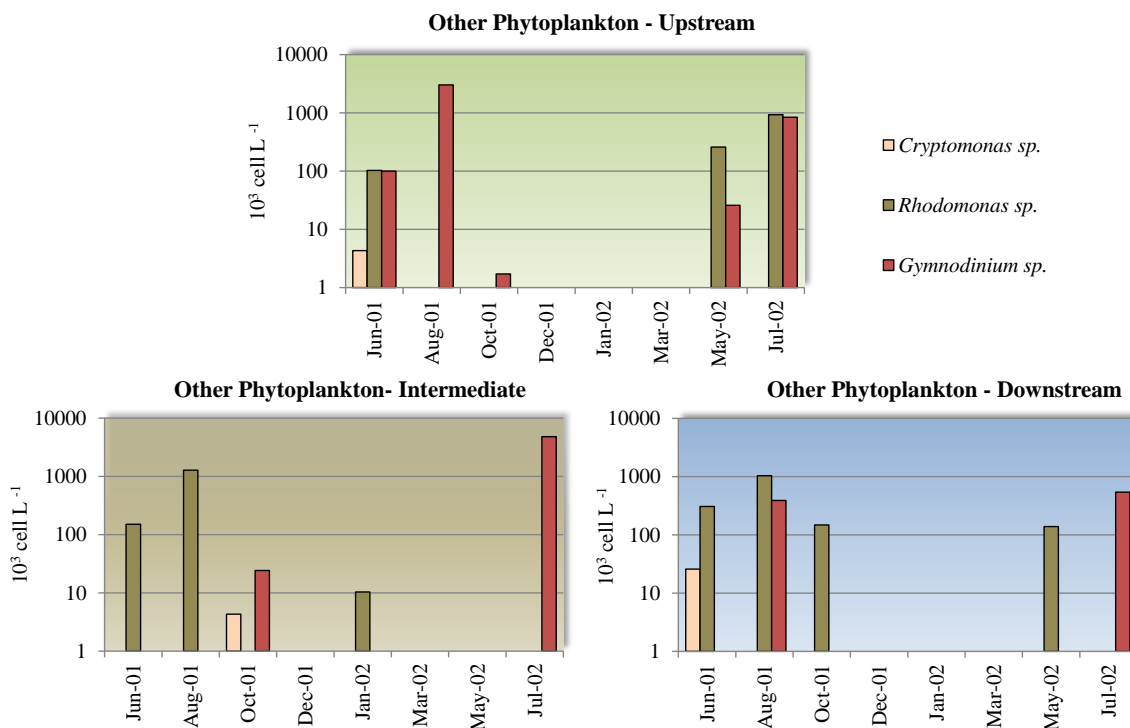


Figure 3.13 - Evolution of Cryptophyceae and Dinophyceae *taxa* abundances in Salgados sampling stations.

Some of the phytoplankton classes presented significant correlations between their abundances. Cyanophyceae abundance was positively correlated to Bacillariophyceae ( $\rho = 0.645$ ;  $p = 0.001$ ), Chlorophyceae ( $\rho = 0.523$ ;  $p = 0.009$ ) and Dinophyceae abundance ( $\rho = 0.478$ ;  $p = 0.018$ ).

Bacillariophyceae abundance, besides being correlated with Cyanophyceae, also showed positive correlations with Chlorophyceae ( $\rho = 0.613$ ;  $p = 0.001$ ), Dinophyceae ( $\rho = 0.559$ ;  $p = 0.005$ ) and Euglenophyceae ( $\rho = 0.517$ ;  $p = 0.010$ ).

Positive correlations were determined between Chlorophyceae abundance and Bacillariophyceae, Cyanophyceae, Dinophyceae abundance ( $\rho = 0.485$ ;  $p = 0.016$ ) and a negative correlation was found with pico-nano flagellate algae ( $\rho = -0.414$ ;  $p = 0.044$ ).

Cryptophyceae abundance did not present significant correlations with the abundances of the other phytoplankton classes ( $p > 0.05$ ).

Dinophyceae abundance was positively correlated to Cyanophyceae, Bacillariophyceae and Chlorophyceae abundances.

The highest annual mean values of taxonomic richness, total phytoplankton abundance, Bacillariophyceae, Cyanophyceae and pico-nano flagellate algae abundances were found upstream, while the lowest mean abundance of Cryptophyceae was observed in this station (Table 3.4). The intermediate station showed the greatest means of Dinophyceae and Euglenophyceae abundances, and also the smallest mean values of evenness and Bacillariophyceae abundance. The upstream and intermediate stations presented the higher mean values of Shannon-Wiener diversity. In the downstream station was determined the lowest means for taxonomic richness, Shannon-Wiener diversity, total phytoplankton abundance, Dinophyceae, Euglenophyceae, Cyanophyceae and pico-nano flagellate algae abundances. Chlorophyceae and Cryptophyceae abundances presented the highest mean values downstream.

Table 3.4 – Annual mean values and standard deviation of phytoplankton parameters in Salgados sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
<b>Phytoplankton classes</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total phytoplankton abundance	92850.63 ± 214738.98	33241.04 ± 5668874	27470.78 ± 43979.88
Taxonomic richness	8.13 ± 4.94	7.63 ± 4.34	6.00 ± 3.55
Shannon-Wiener diversity	1.34 ± 0.80	1.34 ± 0.79	1.13 ± 0.48
Evenness	0.52 ± 0.34	0.48 ± 0.25	0.51 ± 0.22
Chlorophyceae abundance	1741.42 ± 4285.56	1629.18 ± 2293.29	6158.97 ± 15455.95
Bacillariophyceae abundance	1428.91 ± 1887.95	639.10 ± 789.30	865.95 ± 1216.22
Cryptophyceae abundance	161.68 ± 322.64	180.58 ± 446.90	206.62 ± 354.64
Dinophyceae abundance	498.41 ± 1058.21	603.92 ± 1698.42	115.87 ± 218.31
Euglenophyceae abundance	464.56 ± 787.76	757.95 ± 2070.16	106.17 ± 192.56
Cyanophyceae abundance	86749.16 ± 4850.14	28985.84 ± 54437.50	19620.00 ± 40914.93
Pico-nano flagellate algae abundance	1806.49 ± 4850.14	444.48 ± 893.00	396.81 ± 735.82

Despite the distinct annual means each phytoplankton parameter showed in the three stations, the statistical tests applied did not reveal significant differences among the mean values of the stations (Appendix I.H). The high standard deviation values determined in each station indicate that there was a great variation in parameters during the studied period and a possible explanation is that the statistical tests could not find significant differences since the variation within each station was greater than the variation among stations.

Chlorophyll *a* concentration varied from 13.23 µg L<sup>-1</sup> (March 2002, downstream) to 661.66 µg L<sup>-1</sup> (March 2002, upstream). The upstream station was the one with higher concentrations of chlorophyll *a*. The maximum phaeo-pigments concentration (62.64 µg

L<sup>-1</sup>) was found in the downstream station, during July 2002. Margalef's pigment diversity index fluctuated between 1.75 (December 2001, intermediate) and 3.29 (May 2002, upstream) (Figure 2.19). The one-way analysis of variance and the Kruskal-Wallis test did not reveal significant statistical differences of means or mean ranks for photosynthetic pigments concentrations (chlorophyll *a* and phaeo-pigments) among the studied stations, neither for Margalef's pigment diversity index (Appendix I.C).

A positive correlation was determined between chlorophyll *a* concentration and Bacillariophyceae abundance ( $r = 0.445$ ;  $p = 0.029$ ). Phaeo-pigments concentration was positively correlated to Shannon-Wiener diversity ( $r = 0.443$ ;  $p = 0.030$ ) and Chlorophyceae abundance ( $r = 0.434$ ;  $p = 0.034$ ). Margalef's pigment diversity index and taxonomic richness showed a positive correlation ( $r = 0.411$ ;  $p = 0.046$ ).

### 3.3.2.2. Environmental parameters and phytoplankton communities

The bivariate linear correlations performed between phytoplankton and environmental parameters did not reveal any linear association between total phytoplankton abundance and environmental parameters ( $p > 0.05$ ). For taxonomic richness a negative correlation was determined with nitrates concentration, while positive correlations were found with temperature, orthophosphates and total phosphorus concentrations. Shannon-Wiener diversity and evenness were both negatively correlated to total dissolved inorganic nitrogen and N:P ratio. Shannon-Wiener diversity also presented negative correlations with nitrates and nitrites concentrations (Table 3.5).

Cyanophyceae abundance showed a positive correlation with ammonia concentration and Chlorophyceae abundance revealed a negative correlation with nitrates concentration. Dinophyceae abundance was negatively correlated to nitrates concentration, just as with cumulative rainfall and positively correlated with temperature and total phosphorus concentration. Bacillariophyceae abundance had a negative correlation with cumulative rainfall and a positive correlation with pH. Cryptophyceae abundance was negatively correlated with pH, dissolved oxygen concentration and positively correlated with temperature.

The strongest and most significant correlations were found between: 1) Cryptophyceae abundance vs. pH, dissolved oxygen concentration and temperature; 2) Dinophyceae

abundance vs. temperature and cumulative rainfall; 3) Shannon-Wiener diversity vs. N:P ratio; 4) Taxonomic richness vs. total phosphorus concentration.

Table 3.5 - Significant correlations between phytoplankton and environmental parameters in Salgados lagoon. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Phytoplankton parameters</i>	<i>Environmental parameters</i>	<i>Results</i>
Taxonomic richness	Temperature	Rho = 0.511; $p = 0.011$ *
	Nitrates concentration	Rho = -0.459; $p = 0.024$ *
	Orthophosphates concentration	Rho = 0.481; $p = 0.017$ *
	Total Phosphorus concentration	Rho = 0.532; $p = 0.007$ **
Shannon-Wiener diversity	Nitrates concentration	Rho = -0.441; $p = 0.031$ *
	Nitrites concentration	Rho = -0.456; $p = 0.025$ *
	Total dissolved inorganic nitrogen	R = -0.514; $p = 0.010$ *
	N: P ratio	Rho = -0.604; $p = 0.002$ **
Evenness	Total dissolved inorganic nitrogen	R = -0.509; $p = 0.011$ *
	N: P ratio	Rho = -0.522; $p = 0.009$ **
Chlorophyceae abundance	Nitrates concentration	Rho = -0.423; $p = 0.039$ *
Bacillariophyceae abundance	Cumulative rainfall	Rho = -0.441; $p = 0.031$ *
	pH	R = 0.499; $p = 0.013$ *
Cryptophyceae abundance	Temperature	Rho = 0.530; $p = 0.008$ **
	pH	Rho = -0.681; $p < 0.001$ **
	Dissolved oxygen concentration	Rho = -0.582; $p = 0.003$ **
Dinophyceae abundance	Cumulative rainfall	Rho = -0.535; $p = 0.007$ **
	Temperature	Rho = 0.586; $p = 0.003$ **
	Nitrates concentration	Rho = -0.459; $p = 0.024$ *
	Total phosphorus concentration	Rho = 0.422; $p = 0.040$ *
Cyanophyceae abundance	Ammonia concentration	R = 0.442; $p = 0.031$ *

Initially, a Canonical Correspondence Analysis (CCA) was performed with the environmental variables that presented longest arrows and therefore had a major contribution for the two first axes in the PCA (Figure 2.20), namely orthophosphates concentration, pH, temperature, cumulative rainfall, N:P ratio and total dissolved inorganic nitrogen concentration (DIN). Nevertheless, the Monte Carlo permutation test revealed that the relation between these environmental variables and the abundance of phytoplankton groups was not significant (Monte Carlo test:  $p = 0.608$ ). This result could mean that there were other environmental variables with greater contribution to the third or fourth axes of PCA that might explain better the relations with phytoplankton groups.

Thereby, a new CCA was done with five environmental variables that were forward selected and presented significant relations with the phytoplankton groups (Monte Carlo test:  $p = 0.004$ ). The environmental variables considered were cumulative rainfall,

temperature, dissolved oxygen concentration, nitrates concentration and N:P ratio (Figure 3.14).

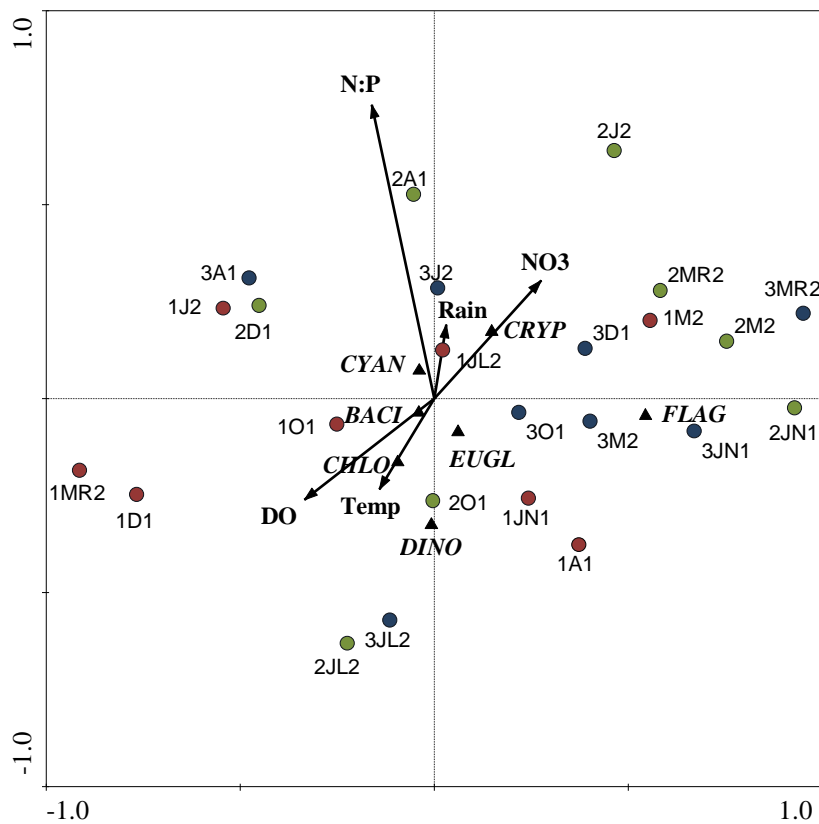


Figure 3.14 - Canonical correspondence analysis performed with the phytoplankton groups (total density *per* station) from Salgados sampling stations. Cumulative percentage variance explained by axes: Species - I = 19.1 % and I + II = 30.9 %; and Species-environment relation - I = 50.4 % and I + II = 81.7 %. Monte Carlo test of all canonical axes  $p = 0.004$

*Station codes:* First character corresponds to the sampling station (1- upstream, 2- intermediate, 3- downstream) and subsequent ones to month and year of survey (1- 2001, 2-2002).

*Environmental variables:* Rain- cumulative rainfall in 10 days previous to sampling; Temp- water temperature; DO- dissolved oxygen concentration; NO<sub>3</sub>- nitrates concentration; N: P- DIN and TP ratio

*Phytoplankton:* CRYP – Cryptophyceae; CHLO – Chlorophyceae; EUGL – Euglenophyceae; DINO – Dinophyceae; BACI – Bacillariophyceae; CYAN – Cyanophyceae; FLAG – Pico-nano flagellate algae (< 20 μm).

Cryptophyceae abundance was related to stations and months with higher nitrates concentration, N:P ratio and lower dissolved oxygen concentration.

Pico-nano flagellate algae were mainly associated to higher concentrations of nitrates, lower dissolved oxygen concentration, temperature and N:P ratio values. Euglenophyceae had similar relations with the same environmental variables as pico-nano flagellate algae, but nitrates concentration did not influence much their abundance.

Samples with higher abundances of Dinophyceae corresponded to those with lower values of N: P ratio, cumulative rainfall, nitrates concentration and higher values of temperature and dissolved oxygen.

Chlorophyceae abundances showed the opposite tendency of Cryptophyceae, being positively associated with temperature, dissolved oxygen concentration and negatively associated to nitrates concentration, cumulative rainfall and N:P ratio.

The Bacillariophyceae plot near the origin of the ordination diagram means that the group was present in the majority of the samples and as it is positioned slightly to the left and down, abundances show some increase with higher dissolved oxygen and lower nitrates concentration.

Cyanophyceae seems to be mostly influenced by N:P ratio and cumulative rainfall, with higher abundances corresponding to higher values of these environmental parameters.

Most of the CCA results are concordant with the linear bivariate associations previously determined.

### **3.3.2.3 Phytoplankton communities during isolation and connection of the lagoon with the sea**

Salgados sampling in January 2002 was made when the lagoon was opened and with direct influence from the sea. In December 2001 the lagoon was isolated and so phytoplankton communities from these two periods were compared. Two previous openings occurred, one in the middle of October (before sampling) and another between December and January samplings.

In all stations there was a decrease in Shannon-Wiener diversity, evenness, Bacillariophyceae abundance and it was observed an increase in total phytoplankton abundance and Cyanophyceae abundance (Table 3.6). The augment of total phytoplankton abundance was mostly due to the Cyanophyceae abundance, as this class represented 98.40% (downstream) to 99.51% (upstream) of total phytoplankton. Dinophyceae and pico-nano flagellate algae were absent from all stations in December 2001 and January 2002. The same happened with Cryptophyceae in the upstream and downstream stations and with Chlorophyceae in the intermediate station.

During the period of connection with the sea, the upstream station presented the greatest abundance of total phytoplankton, taxonomic richness, Cyanophyceae, Bacillariophyceae and Euglenophyceae abundances. Cryptophyceae was only found in the intermediate station, where total phytoplankton and Cyanophyceae abundances showed the minimum values. Shannon-Wiener diversity and evenness registered lower values in the upstream and intermediate stations. Downstream, Shannon-Wiener diversity, evenness and Chlorophyceae abundance registered the highest values, while Bacillariophyceae had the minimum abundances. Taxonomic richness was lower in the intermediate and downstream stations.

Table 3.6 – Phytoplankton communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.

	<i>Upstream station</i>		<i>Intermediate station</i>		<i>Downstream station</i>	
	Jan-02	Variation	Jan-02	Variation	Jan-02	Variation
<b>Phytoplankton communities</b>						
Total phytoplankton abundance ( $10^3$ cell L <sup>-1</sup> )	22788	<b>22619</b>	4164	<b>2885</b>	10014	<b>9585</b>
Taxonomic richness	5	<b>0</b>	4	<b>0</b>	4	<b>1</b>
Shannon-Wiener diversity (bits)	0.13	<b>-1.04</b>	0.12	<b>-1.27</b>	0.57	<b>-0.76</b>
Evenness	0.05	<b>-0.45</b>	0.06	<b>-0.63</b>	0.29	<b>-0.55</b>
Chlorophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	2	<b>-12</b>	0	<b>0</b>	135	<b>135</b>
Bacillariophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	74	<b>-57</b>	43	<b>-721</b>	26	<b>-191</b>
Cryptophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	0	<b>0</b>	10	<b>10</b>	0	<b>0</b>
Dinophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	0	<b>0</b>	0	<b>0</b>	0	<b>0</b>
Euglenophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	36	<b>12</b>	3	<b>-29</b>	0	<b>0</b>
Cyanophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	22676	<b>22676</b>	4108	<b>3625</b>	9854	<b>9642</b>
Pico-nano flagellate abundance ( $10^3$ cell L <sup>-1</sup> )	0	<b>0</b>	0	<b>0</b>	0	<b>0</b>

### 3.3.3. Comparison of the two coastal lagoons

#### 3.3.3.1. Phytoplankton communities

During the studied period, the number of *taxa* identified in the Foz de Almargem (29) was lower than and the number determined for Salgados lagoon (40).

Phytoplankton community in Foz de Almargem was mostly composed by Bacillariophyceae (58.6%) and Dinophyceae *taxa* (17.2%). Chlorophyceae and

Cryptophyceae *taxa* represented 6.9% each of the total *taxa* from the lagoon, while Euglenophyceae and Cyanophyceae only accounted 3.5 % each.

In Salgados lagoon, the majority of phytoplankton *taxa* belonged to Chlorophyceae (35.0%), Cyanophyceae (25.0%) and Bacillariophyceae (17.5%). Euglenophyceae *taxa* accounted 12.5% of the total *taxa* from the lagoon. Cryptophyceae and Dinophyceae *taxa* only represented 5.0% and 2.5% respectively.

Some *taxa* were found in both lagoons, five Bacillariophyceae (*Cyclotella* spp., *Cymbella* sp., *Gomphonema* sp., *Navicula* sp., *Nitzschia* sp.), two Cryptophyceae (*Cryptomonas* sp., *Rhodomonas* sp.), one Dinophyceae (*Gymnodinium* sp.) and one Cyanophyceae (*Anabaena flos-aquae*). Pico-nano flagellate algae were also present in the two lagoons.

The evolution of phytoplankton richness in the two lagoons followed a similar trend during some months, increasing in August 2001, March 2002, July 2002 and decreasing in May 2002. From October 2001 till January 2002, the lagoons presented opposite tendencies (Figure 3.15). In Foz de Almargem, the lowest number of *taxa* (9) occurred in October 2001 and May 2002, and the higher values were observed in August 2001 (18 *taxa*), December 2001 (20 *taxa*) and July 2002 (18 *taxa*).

Salgados lagoon showed the lowest richness in December 2001 (7 *taxa*) and May 2002 (8 *taxa*), while in October 2001 and July 2002 were registered 22 and 24 *taxa*, respectively. Along the year, Salgados lagoon presented a greater variation in the number of *taxa* (17) compared to Foz de Almargem (11).

The seasonal tendencies of total phytoplankton mean abundances in the lagoons were distinct most of the time. Just in December 2001 and May 2002 both lagoons showed a decrease in total phytoplankton and then an increase in July 2002.

The monthly mean values determined in Foz de Almargem were much lower than the ones obtained in Salgados lagoon. In Foz de Almargem, the highest mean abundance was  $681.42 \times 10^3$  cell L<sup>-1</sup> (March 2002) and the lowest was  $93.70 \times 10^3$  cell L<sup>-1</sup> (May 2002). In Salgados lagoon, the lower mean abundances were found in June 2001 ( $536.77 \times 10^3$  cell L<sup>-1</sup>) and December 2001 ( $625.75 \times 10^3$  cell L<sup>-1</sup>), whereas the higher values occurred in August 2001 ( $118722.18 \times 10^3$  cell L<sup>-1</sup>) and July 2002 ( $249724.15 \times 10^3$  cell L<sup>-1</sup>).

Shannon-Wiener diversity index and evenness presented higher mean values in Foz de Almargem, except in June 2001, October 2001 and March 2002. These months

corresponded to lower values of diversity and evenness in Foz de Almargem, with minimum values being registered in March 2002 ( $H' = 0.49$ ;  $E = 0.15$ ). The greatest mean values of diversity and evenness were found in July 2002 ( $H' = 2.57$ ;  $E = 0.80$ ). In Salgados lagoon, the lowest diversity and evenness were determined in January 2002 ( $H' = 0.27$ ;  $E = 0.13$ ) and the highest values were recorded in June 2001 ( $H' = 1.77$ ;  $E = 0.81$ ).

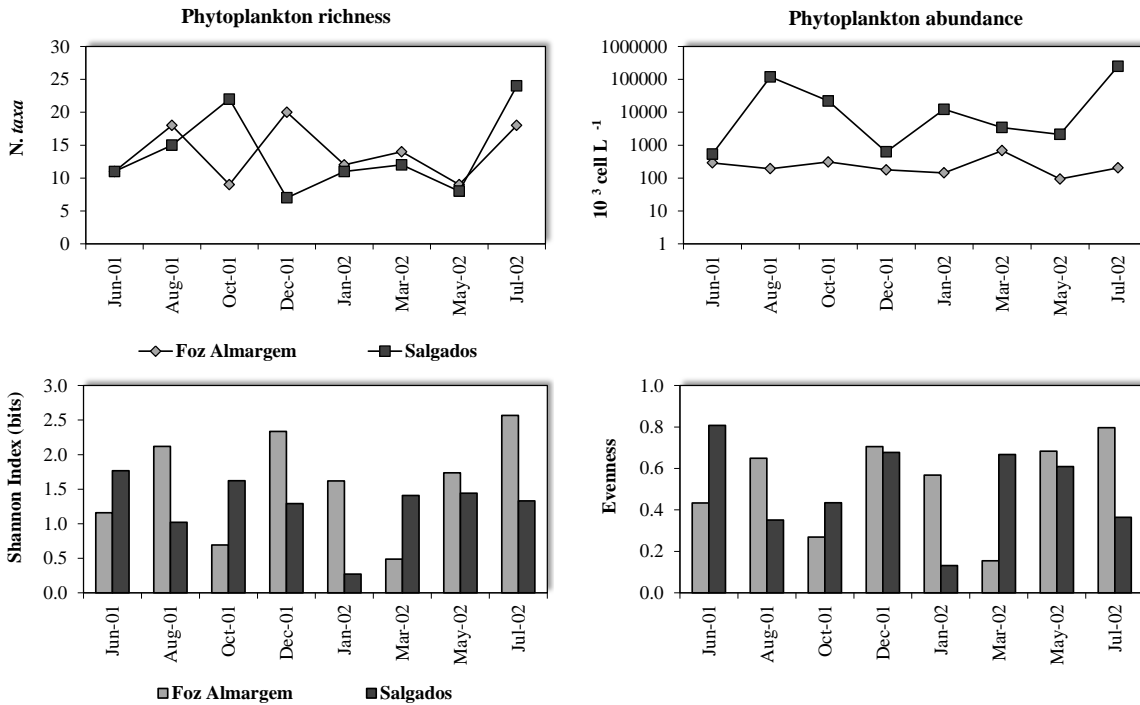


Figure 3.15 - Evolution of phytoplankton richness, total phytoplankton abundance, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) monthly mean values in Foz de Almargem and Salgados lagoons.

During the studied period, Foz de Almargem and Salgados lagoons were dominated by different groups of phytoplankton, except in December 2001, when Bacillariophyceae was the most abundant class in both lagoons (Figure 3.16).

In Foz de Almargem, most of the time, Dinophyceae was the class with greater mean abundance, namely in June 2001 ( $282 \times 10^3 \text{ cell L}^{-1}$ ; 98%), October 2001 ( $289 \times 10^3 \text{ cell L}^{-1}$ ; 93%), January 2002 ( $91 \times 10^3 \text{ cell L}^{-1}$ ; 64%), March 2002 ( $639 \times 10^3 \text{ cell L}^{-1}$ ; 94%) and May 2002 ( $60 \times 10^3 \text{ cell L}^{-1}$ ; 64%). In August 2001, the most abundant group was pico-nano flagellate algae ( $89 \times 10^3 \text{ cell L}^{-1}$ ; 46%), followed by Bacillariophyceae ( $41 \times 10^3 \text{ cell L}^{-1}$ ; 21%), Dinophyceae ( $36 \times 10^3 \text{ cell L}^{-1}$ ; 19%) and Cryptophyceae ( $25 \times 10^3 \text{ cell L}^{-1}$ ; 13%). In December 2001 and July 2002, Bacillariophyceae dominated the

phytoplankton communities, with mean abundances of  $118 \times 10^3 \text{ cell L}^{-1}$  (66%) and  $126 \times 10^3 \text{ cell L}^{-1}$  (62%), respectively. Dinophyceae was the second major group represented in these months, accounting 18% of the mean abundance in December 2001 ( $33 \times 10^3 \text{ cell L}^{-1}$ ) and 37% in July 2002 ( $76 \times 10^3 \text{ cell L}^{-1}$ ). In January 2002, besides Dinophyceae, the community was mainly composed by Bacillariophyceae ( $22 \times 10^3 \text{ cell L}^{-1}$ ; 16%), Cyanophyceae ( $13 \times 10^3 \text{ cell L}^{-1}$ ; 9%), pico-nano-flagellate algae ( $12 \times 10^3 \text{ cell L}^{-1}$ ; 8%) and Cryptophyceae ( $5 \times 10^3 \text{ cell L}^{-1}$ ; 3%). In May 2002, the phytoplankton groups were the same as in January, except for Cyanophyceae: Dinophyceae ( $60 \times 10^3 \text{ cell L}^{-1}$ ; 64%), pico-nano-flagellate algae ( $16 \times 10^3 \text{ cell L}^{-1}$ ; 17%), Bacillariophyceae ( $15 \times 10^3 \text{ cell L}^{-1}$ ; 16%) and Cryptophyceae ( $3 \times 10^3 \text{ cell L}^{-1}$ ; 4%).

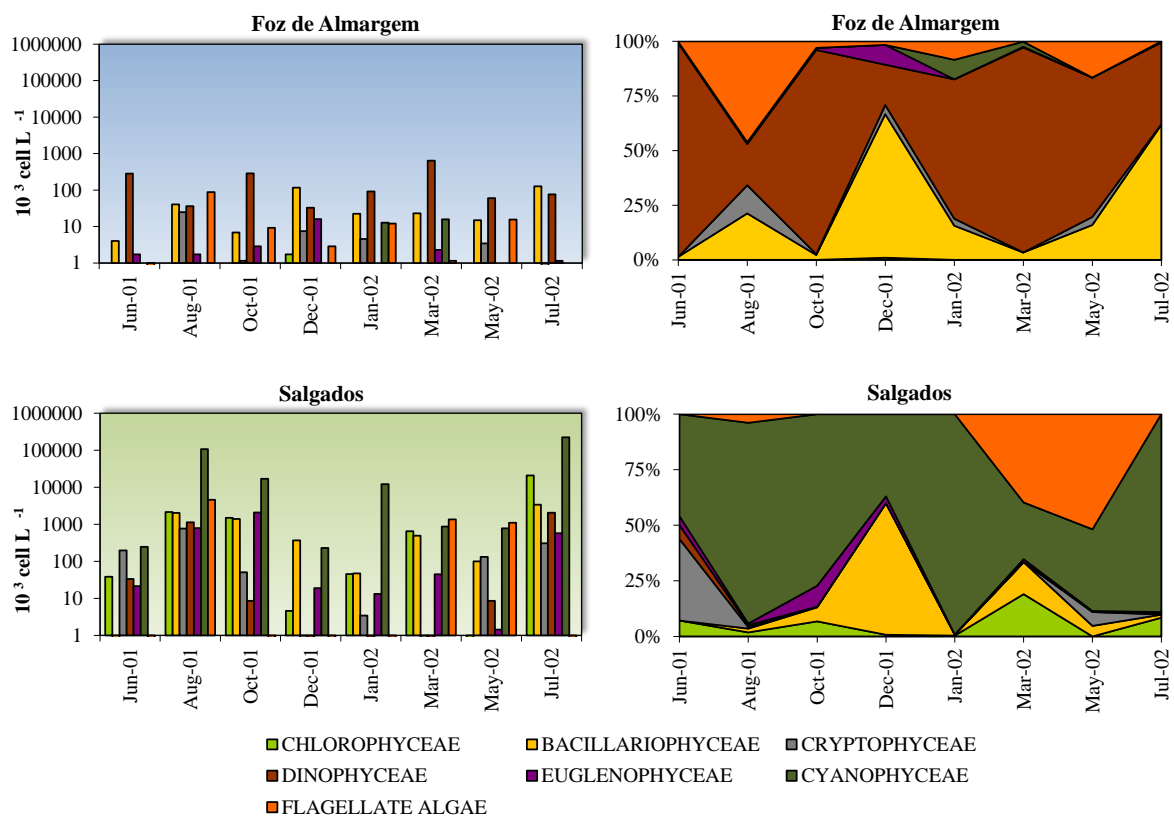


Figure 3.16 - Evolution of phytoplankton mean abundance *per* class and relative frequency of each class in Foz de Almagem and Salgados lagoons.

In Salgados lagoon, besides the fact that the monthly mean abundances of the taxonomic groups were generally higher than in Foz de Almagem, phytoplankton communities along time were mostly dominated by Cyanophyceae, with the exception of December 2001, March and May 2002.

In June 2001, the two major classes were Cyanophyceae ( $246 \times 10^3 \text{ cell L}^{-1}$ ; 46%) and Cryptophyceae ( $197 \times 10^3 \text{ cell L}^{-1}$ ; 37%). Cyanophyceae mean abundances were much higher in August 2001 ( $107249 \times 10^3 \text{ cell L}^{-1}$ ; 90%), October 2001 ( $16998 \times 10^3 \text{ cell L}^{-1}$ ; 77%), January 2002 ( $12212 \times 10^3 \text{ cell L}^{-1}$ ; 99%) and July 2002 ( $222354 \times 10^3 \text{ cell L}^{-1}$ ; 89%), being the predominant class during these months. In December 2001, Bacillariophyceae dominated the phytoplankton community ( $371 \times 10^3 \text{ cell L}^{-1}$ ; 59%), followed by Cyanophyceae ( $231 \times 10^3 \text{ cell L}^{-1}$ ; 37%). During March 2002 and May 2002, pico-nano-flagellate algae was the group with greater mean abundances, with  $1362 \times 10^3 \text{ cell L}^{-1}$  (40%) and  $1099 \times 10^3 \text{ cell L}^{-1}$  (52%) respectively. Cyanophyceae was the second major class in these months, accounting 25% of the mean abundance in March 2002 ( $876 \times 10^3 \text{ cell L}^{-1}$ ) and 37% in May 2002 ( $780 \times 10^3 \text{ cell L}^{-1}$ ). In March 2002, Chlorophyceae ( $652 \times 10^3 \text{ cell L}^{-1}$ ) and Bacillariophyceae ( $494 \times 10^3 \text{ cell L}^{-1}$ ) represented 19% and 14% of the phytoplankton community.

Besides pico-nano flagellate algae, nine *taxa* (genus or species) occurred in both lagoons: the Bacillariophyceae *Cyclotella* spp., *Cymbella* sp., *Gomphonema* sp., *Navicula* spp. and *Nitzschia* sp.; the Cryptophyceae *Cryptomonas* sp. and *Rhodomonas* sp.; the Dinophyceae *Gymnodinium* sp. and the Cyanophyceae *Anabaena flos-aquae*.

During the studied period, *Cyclotella* spp. and *Navicula* spp. were the Bacillariophyceae *taxa* with greater mean abundances in Foz de Almargem and Salgados lagoons (Figure 3.17).

*Cyclotella* spp. showed a regular presence in the two lagoons, occurring since August 2001 till July 2002. In Foz de Almargem, the lowest and the highest mean abundances were observed in May 2002 ( $1000 \text{ cell L}^{-1}$ ) and December 2001 ( $28667 \text{ cell L}^{-1}$ ), while in Salgados lagoon the lowest value was registered in January 2002 ( $22994 \text{ cell L}^{-1}$ ) and the higher values were determined in July 2002 ( $2580 \times 10^3 \text{ cell L}^{-1}$ ) and August 2001 ( $1940 \times 10^3 \text{ cell L}^{-1}$ ).

*Navicula* spp. presence was less regular and it occurred simultaneously in the two lagoons just from March to July 2002. In Foz de Almargem, *Navicula* spp. was observed also in June 2001, August 2001 and January 2002, presenting the lowest mean abundance in August 2001 ( $1000 \text{ cell L}^{-1}$ ) and the highest in July 2002 ( $9667 \text{ cell L}^{-1}$ ). Salgados lowest mean abundance for *Navicula* spp. was recorded in March 2002 ( $15809 \text{ cell L}^{-1}$ ) and the highest value was observed in October 2001 ( $1131.317 \times 10^3 \text{ cell L}^{-1}$ ).

The occurrence of *Cymbella* sp. and *Nitzschia* sp. was more irregular in Salgados lagoon, with *Cymbella* sp. being registered only in January 2002 (24719 cell L<sup>-1</sup>) and *Nitzschia* sp. in March 2002 (8623 cell L<sup>-1</sup>) and July 2002 (7186 cell L<sup>-1</sup>). In Foz de Almargem, *Cymbella* sp. was also identified in January 2002 (667 cell L<sup>-1</sup>), besides August 2001 and July 2002, while *Nitzschia* sp. presence was noticed in June and August 2001, May and July 2002 (667 cell L<sup>-1</sup>).

*Gomphonema* sp. only occurred in Foz de Almargem during August 2001 (667 cell L<sup>-1</sup>) and in Salgados lagoon it was observed just in March 2002 (145151 cell L<sup>-1</sup>).

The Dinophyceae *Gymnodinium* sp. occurred in both lagoons during June and August 2001, with mean abundances of 5667 cell L<sup>-1</sup> and 6333 cell L<sup>-1</sup> in Foz de Almargem, while in Salgados lagoon mean abundances were 33533 cell L<sup>-1</sup> and 1135 x 10<sup>3</sup> cell L<sup>-1</sup> during these months. In Foz de Almargem, *Gymnodinium* sp. was found once more in December 2001 (667 cell L<sup>-1</sup>), whereas in Salgados lagoon it was also registered in October 2001 (8623 cell L<sup>-1</sup>), May and July 2002 (2062 x10<sup>3</sup> cell L<sup>-1</sup>).

*Cryptomonas* sp. presence in Foz de Almargem was recorded from August 2001 (12667 cell L<sup>-1</sup>) to December 2001 and in May 2002, but in Salgados lagoon it only occurred in June 2001 (1068 cell L<sup>-1</sup>) and October 2001. Therefore, October 2001 was the only month during which *Cryptomonas* sp. was simultaneously present in the two lagoons and coincided with the lowest mean abundances in Foz de Almargem (1000 cell L<sup>-1</sup>) and in Salgados (1437 cell L<sup>-1</sup>).

*Rhodomonas* sp. was observed in Salgados during most of the studied period, being absent in December 2001 and March 2002. Mean abundances went from 3449 cell L<sup>-1</sup> (January 2002) to 771256 cell L<sup>-1</sup> (August 2001). In Foz de Almargem, *Rhodomonas* sp. highest mean abundance was also determined in August 2001 (12333 cell L<sup>-1</sup>), but the lowest abundance was registered in July 2002 (667 cell L<sup>-1</sup>). Besides these months, *Rhodomonas* sp. was also present in Foz de Almargem during December 2001 and January 2002 (4333 cell L<sup>-1</sup>).

*Anabaena flos-aqua* was identified in different periods of the year in Foz de Almargem (January 2002: 12667 cell L<sup>-1</sup>; March 2002: 15667 cell L<sup>-1</sup>) and in Salgados lagoon (June 2001: 127906 cell L<sup>-1</sup>; August 2001: 488628 cell L<sup>-1</sup>).

Most of the *taxa* that were common to the two lagoons presented higher monthly mean abundances in Salgados lagoon, except for *Cryptomonas* sp.

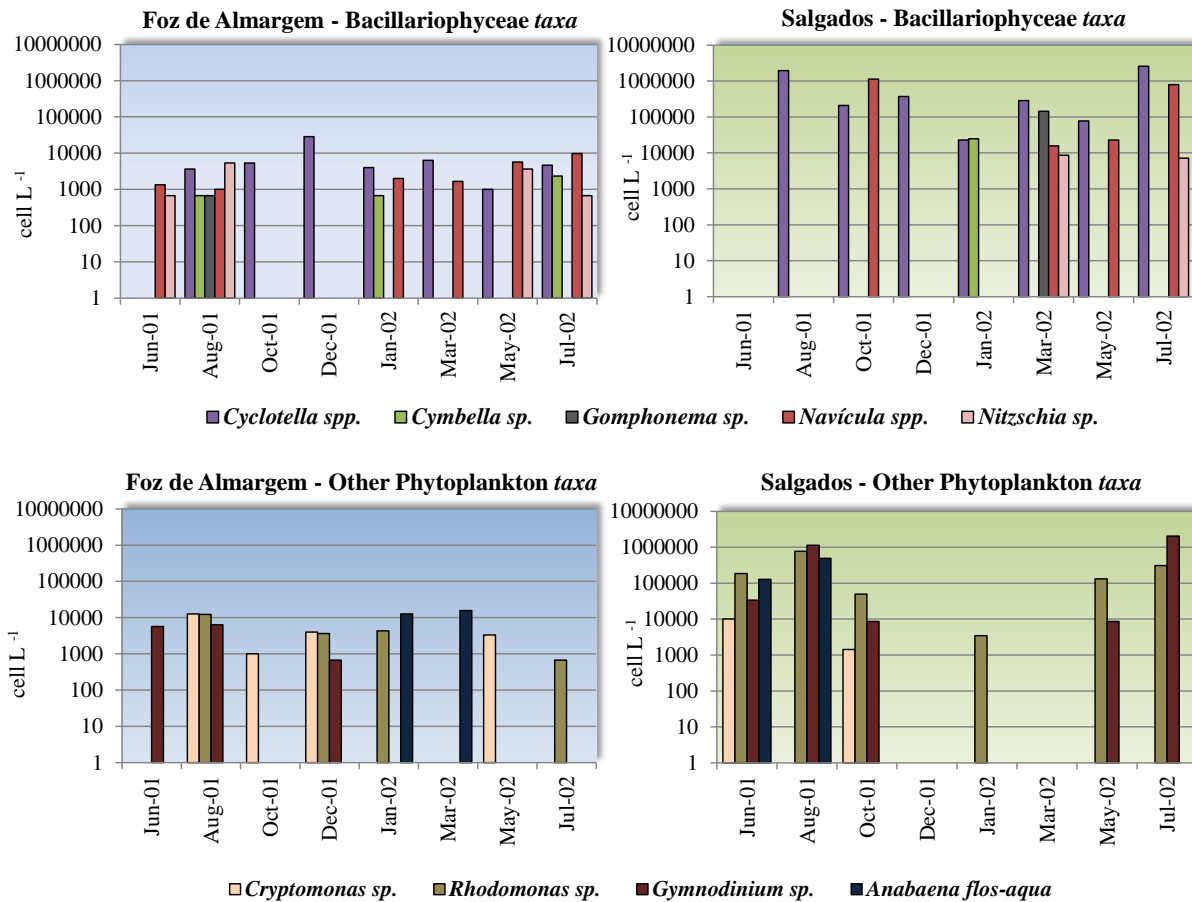


Figure 3.17 - Monthly mean abundances of phytoplankton taxa that occurred in both lagoons.

Taxonomic richness, Shannon-Winner diversity and evenness annual mean values were slightly higher in Foz de Almargem, but all other phytoplankton parameters presented much higher values in Salgados lagoon (Table 3.7).

The statistical tests used to compare the annual mean values of phytoplankton parameters from Foz de Almargem and Salgados lagoons determined that the differences in taxonomic richness, Shannon-Winner diversity, evenness and pico-nano flagellate algae abundances were not statistically significant ( $p > 0.05$ ). Nevertheless, the two lagoons presented significant differences ( $0.05 \geq p > 0.01$ ) between the annual mean values of Cryptophyceae abundance and highly significant differences ( $p \leq 0.01$ ) for all other phytoplankton parameters, namely total phytoplankton, Chlorophyceae, Bacillariophyceae, Dinophyceae, Euglenophyceae and Cyanophyceae abundances (Appendix I.I).

Table 3.7 – Annual mean values and standard deviation of phytoplankton parameters in Foz de Almargem and Salgados lagoons.

	<i>Foz de Almargem</i>	<i>Salgados</i>
<b>Phytoplankton parameters</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total phytoplankton abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	261.25 ± 239.87	51187.48 ± 128501.01
Taxonomic richness	7.92 ± 2.24	7.25 ± 4.22
Shannon-Wiener diversity (bits)	1.59 ± 0.81	1.27 ± 0.68
Evenness	0.53 ± 0.25	0.51 ± 0.26
Chlorophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	0.22 ± 0.58	3176.53 ± 9194.45
Bacillariophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	44.41 ± 74.11	977.99 ± 1356.33
Cryptophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	5.25 ± 9.38	182.96 ± 362.07
Dinophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	188.34 ± 244.01	406.07 ± 1130.98
Euglenophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	3.23 ± 5.78	442.89 ± 1256.42
Cyanophyceae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	3.57 ± 10.24	45118.46 ± 124596.56
Pico-nano flagellate algae abundance (10 <sup>3</sup> cell L <sup>-1</sup> )	16.24 ± 43.98	882.59 ± 2830.66

### 3.3.3.2. Environmental parameters and phytoplankton communities

Phytoplankton parameters in Foz de Almargem presented a lower number of significant correlations with environmental parameters than in Salgados lagoon and most of the correlations determined for Foz de Almargem were different from those found in Salgados lagoon (Table 3.8). Just Bacillariophyceae and Dinophyceae abundances presented correlations with cumulative rainfall in both lagoons. This might have happened due to differences in *taxa* composition and abundances of the major taxonomic groups, but also because the range of some environmental parameters was quite distinct in the two lagoons.

In Foz de Almargem, phytoplankton parameters were mostly correlated with cumulative rainfall, pH and a few parameters presented correlations with water level, dissolved oxygen concentration, orthophosphates, total phosphorus and nitrites concentrations.

Phytoplankton parameters in Salgados lagoon were majorly correlated with temperature, nitrogen compounds concentrations (nitrates, total dissolved inorganic nitrogen and ammonia), phosphorus compounds concentrations (orthophosphates and total phosphorus), N: P ratio and for some parameters were also determined correlations with cumulative rainfall, pH and dissolved oxygen concentration.

Table 3.8- Resume of the significant correlations between phytoplankton communities and environmental parameters in Foz de Almargem and Salgados lagoons. (+) Positive correlation; (-) Negative correlation; (\*) Correlation significant at the 0.05 level; (\*\*) Correlation significant at the 0.01 level.

<i>Phytoplankton parameters</i>	<i>Environmental parameters</i>	<i>Foz de Almargem</i>	<i>Salgados</i>
Taxonomic richness	Cumulative rainfall	(-) **	
	Temperature		(+) *
	Nitrates concentration		(-) *
	Orthophosphates concentration		(+) *
	Total Phosphorus concentration		(+) **
Shannon-Wiener diversity	Cumulative rainfall	(-) **	
	pH	(+) **	
	Nitrates concentration		(-) *
	Nitrites concentration		(-) *
	Total dissolved inorganic nitrogen		(-) *
	N: P ratio		(-) **
Evenness	Cumulative rainfall	(-) **	
	pH	(+) *	
	Total dissolved inorganic nitrogen		(-) *
	N: P ratio		(-) **
Chlorophyceae abundance	Nitrates concentration		(-) *
Bacillariophyceae abundance	Cumulative rainfall	(-) **	(-) *
	pH		(+) *
Cryptophyceae abundance	Water level	(-) **	
	Cumulative rainfall	(-) *	
	Temperature		(+) **
	pH		(-) **
	Dissolved oxygen concentration		(-) **
	Nitrites concentration	(-) *	
Dinophyceae abundance	Cumulative rainfall	(+) **	(-) **
	Temperature		(+) **
	pH	(+) *	
	Nitrates concentration		(-) *
	Total phosphorus concentration		(+) *
Euglenophyceae abundance	Dissolved oxygen concentration	(+) *	
	Orthophosphates concentration	(-) *	
	Total phosphorus concentration	(-) *	
Cyanophyceae abundance	Ammonia concentration		(+) *

The Canonical Correspondence Analysis performed with phytoplankton groups and five environmental parameters (orthophosphates, N: P ratio concentration, temperature, nitrates and nitrites concentrations) showed that the lagoons had distinct characteristics and relations between phytoplankton groups abundances and these environmental parameters, once samples from the two lagoons were plotted separately along axis I (Figure 3.18).

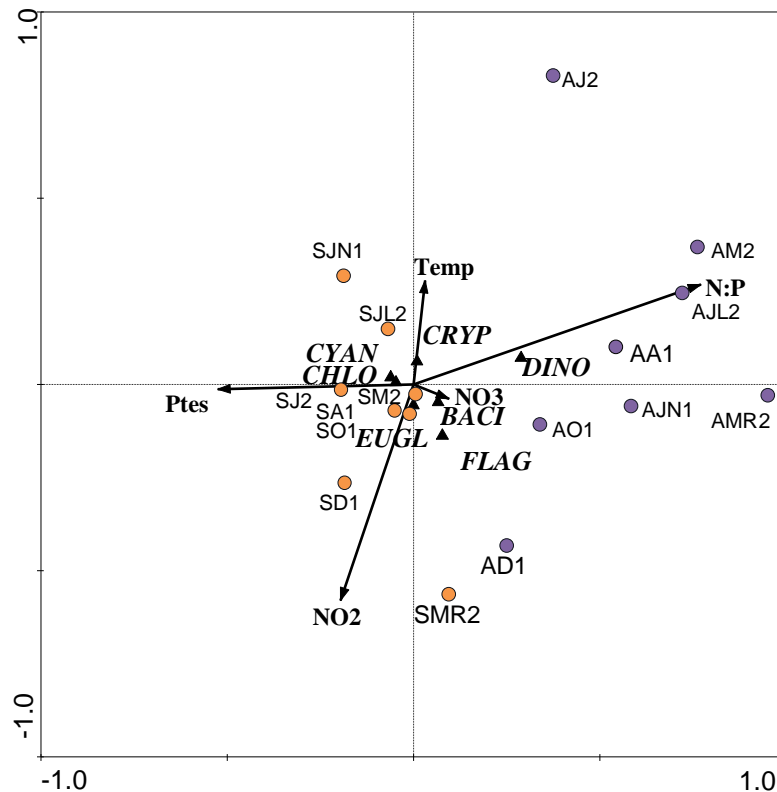


Figure 3.18 - Canonical correspondence analysis performed with the phytoplankton groups (mean density) from Foz de Almargem and Salgados lagoon. Cumulative percentage variance explained by axes: Species - I = 36.5 % and I + II = 45.8 %; and Species-environment relation - I = 69.9 % and I + II = 87.7 %. Monte Carlo test of all canonical axes  $p = 0.008$ .

*Station codes:* First character corresponds to lagoon (S- Salgados, A-Foz de Almargem) and subsequent ones to month and year of survey (1- 2001, 2-2002).

*Environmental variables:* TEMP – water temperature; N:P - DIN and TP ratio; NO<sub>3</sub>- nitrates concentration; NO<sub>2</sub>- nitrites concentration; PTES- orthophosphates concentration.

*Phytoplankton:* CRYP – Cryptophyceae; CHLO – Chlorophyceae; EUGL – Euglenophyceae; DINO – Dinophyceae; BACI – Bacillariophyceae; CYAN – Cyanophyceae; FLAG – Pico-nano flagellate algae (< 20  $\mu\text{m}$ ).

All samples from Foz de Almargem were disposed in the right side of the first axis, presenting higher abundances of Dinophyceae, Bacillariophyceae, pico-nano flagellate algae and lower abundances of Cyanophyceae and Chlorophyceae. The Dinophyceae and Bacillariophyceae abundances were positively related with N:P ratio, nitrates concentration and negatively associated to orthophosphates concentration, whereas Cyanophyceae and Chlorophyceae abundances had the opposite relation with these parameters. On the left side of the first axis were located only samples from Salgados lagoon, which presented higher abundances of Cyanophyceae and Chlorophyceae.

Samples from the two lagoons were displayed along the second axis according to the abundance of Cryptophyceae, which was positively associated to temperature and

negatively related with nitrites concentration (upper side); and the abundances of picoplankton flagellate algae and Euglenophyceae, that increased with higher concentrations of nitrites and lower temperatures (bottom side).

The Monte Carlo permutation test determined that the abundance of phytoplankton groups and these environmental variables was significant ( $p = 0.008$ ).

For some of the *taxa* that occurred in both lagoons, significant linear associations were found with the environmental parameters (Table 3.9).

Table 3.9 - Significant correlations between environmental parameters and the abundances of phytoplankton *taxa* that occurred in both lagoons. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Phytoplankton taxa</i>	<i>Environmental parameters</i>	<i>Results</i>
<i>Cyclotella</i> spp.	pH	Rho = 0.420; $p = 0.003$ **
	Salinity	Rho = -0.442; $p = 0.002$ **
	Orthophosphates concentration	Rho = 0.431; $p = 0.002$ **
	Total Phosphorus concentration	Rho = 0.428; $p = 0.002$ **
	N: P ratio	Rho = -0.383; $p = 0.007$ **
	Total solids in suspension	Rho = 0.349; $p = 0.015$ *
<i>Navicula</i> spp.	Water level	Rho = 0.437; $p = 0.002$ **
	Salinity	Rho = -0.344; $p = 0.017$ *
	Total solids in suspension	Rho = -0.328; $p = 0.023$ *
<i>Cryptomonas</i> sp.	Nitrites concentration	Rho = -0.298; $p = 0.040$ *
<i>Rhodomonas</i> sp.	Cumulative rainfall	Rho = -0.312; $p = 0.031$ *
	Dissolved oxygen concentration	Rho = -0.287; $p = 0.048$ *
	Orthophosphates concentration	Rho = 0.300; $p = 0.038$ *
<i>Gymnodinium</i> sp.	Cumulative rainfall	Rho = -0.357; $p = 0.013$ *
	Nitrates concentration	Rho = -0.328; $p = 0.023$ *
	Nitrites concentration	Rho = -0.371; $p = 0.009$ **

The Bacillariophyceae *Cyclotella* spp. and *Navicula* spp. abundances presented negative correlations with salinity and although the two *taxa* were correlated with total solids in suspension, *Cyclotella* spp. had a positive association whereas *Navicula* spp. had a negative association. Besides these environmental parameters, *Navicula* spp. was positively correlated with water level, and *Cyclotella* spp. showed positive correlations with pH, orthophosphates, total phosphorus concentrations and a negative correlation with N: P ratio.

A negative correlation was found between *Cryptomonas* sp. and nitrites concentration.

*Gymnodinium* sp. was also negatively correlated with nitrites concentration, as well as with nitrates concentration and cumulative rainfall.

For *Rhodomonas* sp., negative correlations were observed with cumulative rainfall and dissolved oxygen concentration. There was also a positive association between *Rhodomonas* sp. abundance and orthophosphates concentration.

The strongest and most significant linear associations were determined between *Cyclotella* spp. and salinity (-), orthophosphates (+), total phosphorus concentration (+), pH (+), N: P ratio (-); *Navicula* spp. and water level (+); *Gymnodinium* sp. and nitrites concentration (-).

### **3.3.3.3. Comparison of phytoplankton communities during isolation and connection of the lagoons with the sea**

Foz de Almargem and Salgados lagoons were isolated from the sea during December 2001 samplings and in January 2002, the two lagoons were in connection with the sea. Thereby, mean phytoplankton data from these two periods were compared.

Both lagoons presented a decrease in Shannon-Wiener diversity, evenness, Bacillariophyceae abundances, Euglenophyceae abundances and an increase in Cyanophyceae abundances (Table 3.10). Total phytoplankton abundance, taxonomic richness, Chlorophyceae and Cryptophyceae abundances showed a decrease in Foz de Almargem lagoon and an increase in Salgados lagoon. It was also observed an augment in Dinophyceae and pico-nano flagellate algae abundances in Foz de Almargem lagoon. The greatest variation within phytoplankton groups was registered in Salgados Cyanophyceae abundances.

When the lagoons were in connection with the sea, the higher values of Shannon-Wiener diversity, evenness, Cryptophyceae, Dinophyceae and pico-nano flagellate algae abundances occurred in Foz de Almargem lagoon and no specimens of Chlorophyceae or Euglenophyceae were found here. Salgados lagoon registered the greatest values of total phytoplankton abundance, Chlorophyceae, Bacillariophyceae, Euglenophyceae and Cyanophyceae abundances. Dinophyceae and pico-nano flagellate algae were absent from Salgados lagoon in December 2001 and January 2002.

Taxonomic richness was slightly higher in Foz de Almargem lagoon.

Table 3.10 – Phytoplankton parameters in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).

Phytoplankton parameters	<i>Foz de Almagem lagoon</i>		<i>Salgados lagoon</i>	
	Jan-02	Variation	Jan-02	Variation
Total phytoplankton abundance ( $10^3$ cell L <sup>-1</sup> )	143	<b>-34</b>	12322	<b>11697</b>
Taxonomic richness	12	<b>-7</b>	10	<b>3</b>
Shannon-Wiener diversity (bits)	1.62	<b>-0.72</b>	0.27	<b>-1.02</b>
Evenness	0.57	<b>-0.14</b>	0.13	<b>-0.55</b>
Chlorophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	0	<b>-2</b>	45	<b>41</b>
Bacillariophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	22	<b>-94</b>	48	<b>-323</b>
Cryptophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	5	<b>-3</b>	3	<b>3</b>
Dinophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	91	<b>59</b>	0	<b>0</b>
Euglenophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	0	<b>-16</b>	13	<b>-6</b>
Cyanophyceae abundance ( $10^3$ cell L <sup>-1</sup> )	13	<b>13</b>	12212	<b>11981</b>
Pico-nano flagellate algae abundance ( $10^3$ cell L <sup>-1</sup> )	12	<b>9</b>	0	<b>0</b>

For the majority of phytoplankton parameters in both lagoons, the variation between the values obtained in December 2001 and January 2002 was related to the variation registered in some environmental parameters.

In Foz de Almagem, when the lagoon was in connection with the sea, there was an increase in cumulative rainfall, pH, nitrates, orthophosphates and total phosphorus concentrations, just as a decrease in dissolved oxygen concentration (Table 2.6).

According to the significant correlations determined between phytoplankton groups and environmental parameters (Table 3.2), the decreases in Bacillariophyceae and Cryptophyceae abundances might have been associated to the increase of cumulative rainfall, as these parameters were negatively correlated. Besides cumulative rainfall, Cryptophyceae abundance also presented a negative correlation with nitrates concentrations, which increased in January 2002.

Dinophyceae abundance augment could be explained by the positive correlations with cumulative rainfall and pH.

The diminishment in Euglenophyceae abundance might have been related to the increase of orthophosphates and total phosphorus concentrations and the decrease in dissolved oxygen concentration, once this group abundance showed negative correlations with the first parameters and a positive correlation with the last one.

When Salgados lagoon was connected to the sea, environmental parameters as cumulative rainfall, water temperature, nitrogen compounds concentrations and N: P ratio presented an increase in their values, while dissolved oxygen concentration and pH decreased (Table 2.14).

Bacillariophyceae abundance reduction might have been associated to the increase of cumulative rainfall and the decrease in pH, as Bacillariophyceae abundance was negatively correlated with cumulative rainfall and positively correlated with pH.

The augment in Cryptophyceae abundance matched with the increase of temperature and the decrease of pH and dissolved oxygen concentration, as this group was positively correlated with temperature and negatively correlated with pH and dissolved oxygen concentration.

The positive correlation between Cyanophyceae abundance and ammonia concentration was explicit in the increase of both parameters, when the lagoon was opened to the sea.

#### **3.3.3.4. Phytoplankton communities according to salinity preferences**

Phytoplankton community in Foz de Almargem lagoon was mainly composed by freshwater/brackish/marine *taxa* (9 *taxa*; 31%), brackish/marine *taxa* (8 *taxa*; 27%) and freshwater/brackish *taxa* (8 *taxa*; 27%). Freshwater *taxa* (4) represented 14% of the *taxa* identified in the lagoon.

Most of the time, brackish/marine *taxa* was the category with greatest richness (Figure 3.19), namely in June 2001 (7 *taxa*; 70%), October 2001 (5 *taxa*; 56%), December 2001 (7 *taxa*; 37%), March 2002 (6 *taxa*; 43%) and July (6 *taxa*; 33%). In August 2001, the number of brackish/marine *taxa* was the same as in March and July 2002 (6 *taxa*; 35%). Brackish/marine category integrated three Bacillariophyceae, four Dinophyceae and one Euglenophyceae *taxa*.

In terms of abundance, brackish/marine *taxa* presented the highest values (March 2002:  $651 \times 10^3$  cell L<sup>-1</sup>; October 2001:  $292 \times 10^3$  cell L<sup>-1</sup>), dominating the phytoplankton community in October 2001 (95%), December 2001 ( $89 \times 10^3$  cell L<sup>-1</sup>; 50%), March 2002 (96%) and May 2002 ( $52 \times 10^3$  cell L<sup>-1</sup>; 56%). In July 2002, it was also one of the main categories ( $86 \times 10^3$  cell L<sup>-1</sup>; 42%), jointly with freshwater/brackish/marine *taxa* ( $82 \times 10^3$  cell L<sup>-1</sup>; 40%). In October 2001, the most relevant brackish/marine *taxon* was the dinoflagellate *Protoperidinium* sp. ( $283667$  cell L<sup>-1</sup>; 92% of the abundance in this month), while in December 2001, the diatom *Leptocylindrus danicus* had the highest abundance ( $53333$  cell L<sup>-1</sup>; 30%), followed by the Euglenophyceae *Eutreptiella* sp. ( $16000$  cell L<sup>-1</sup>; 9%) and the diatom *Scrippsiella trochoidea* ( $11333$  cell L<sup>-1</sup>; 6%). The dinoflagellate *Prorocentrum minimum* was the brackish/marine *taxon* with greater

abundances in March (637000 cell L<sup>-1</sup>; 94%), May (52333 cell L<sup>-1</sup>; 56%) and July 2002 (58333 cell L<sup>-1</sup>; 29%).

Freshwater/brackish/marine *taxa* registered a greater richness in December 2001 (6 *taxa*; 32%), January (5 *taxa*; 42%) and July 2002 (5 *taxa*; 28%), being the category with the highest number of *taxa* in January 2002, when the lagoon was in connection with the sea. This category was composed by one Chlorophyceae, six Bacillariophyceae, one Dinophyceae and pico-nano flagellate algae.

Concerning abundance, freshwater/brackish/marine *taxa* was the second major category, showing the greatest values in June 2001 (244 x 10<sup>3</sup> cell L<sup>-1</sup>; 85%), August 2001 (95 x 10<sup>3</sup> cell L<sup>-1</sup>; 50%) and January 2002 (106 x 10<sup>3</sup> cell L<sup>-1</sup>; 75%). During June 2001 and January 2002, for the dinoflagellate *Gymnodinium* sp. were determined abundances of 243667 cell L<sup>-1</sup> and 90333 cell L<sup>-1</sup>, representing 84% and 64 % of the phytoplankton abundances in each month. In August 2001, pico-nano flagellate algae presented an abundance of 88333 cell L<sup>-1</sup>, accounting 46% of the phytoplankton abundance.

The richness of fresh/brackish *taxa* varied from two *taxa* in June 2001 (20%) and October 2001 (22%) to seven *taxa* in August 2001 (41%) and six *taxa* in July 2002 (33%). During May 2002, five *taxa* were observed, accomplishing 56% of the *taxa* identified in this month. Fresh/brackish *taxa* included eight Bacillariophyceae and two Cryptophyceae *taxa*.

The months with higher abundances of freshwater/brackish *taxa* were August 2001 (61 x 10<sup>3</sup> cell L<sup>-1</sup>; 32%) and December 2001 (37 x 10<sup>3</sup> cell L<sup>-1</sup>; 21%), May (18 x 10<sup>3</sup> cell L<sup>-1</sup>; 19%) and July 2002 (36 x 10<sup>3</sup> cell L<sup>-1</sup>; 18%). In August 2001, the diatom *Cocconeis* sp. (25667 cell L<sup>-1</sup>; 13%) and the Cryptophyceae *Cryptomonas* sp. (12667 cell L<sup>-1</sup>; 7%) and *Rhodomonas* sp. (12333 cell L<sup>-1</sup>; 6%) were the most abundant freshwater/brackish *taxa*. During December 2001, the diatom *Cyclotella* spp. (28667 cell L<sup>-1</sup>; 16%), *Cryptomonas* sp. (4000 cell L<sup>-1</sup>; 2%) and *Rhodomonas* sp. (3667 cell L<sup>-1</sup>; 2%) were the freshwater/brackish *taxa* with higher abundances. In May 2002, the most relevant *taxa* were the diatoms *Navicula* spp. (5667 cell L<sup>-1</sup>; 6%), *Cocconeis* sp. (4000 cell L<sup>-1</sup>; 4%), *Nitzschia* sp. (3667 cell L<sup>-1</sup>; 4%) and the Cryptophyceae *Cryptomonas* sp. (3333 cell L<sup>-1</sup>; 4%).

Freshwater *taxa* were observed from December 2001 to March 2002 and in July 2001. In December 2001 three *taxa* were identified (16% of the *taxa* in this month), the Chlorophyceae *Cosmarinum* sp. (667 cell L<sup>-1</sup>; < 1%) and the diatoms *Asterionellopsis formosa* (7000 cell L<sup>-1</sup>; 4%) and *Aulacoseira granulate* (2333 cell L<sup>-1</sup>; 1%), presenting a

mean abundance of  $10 \times 10^3$  cell L<sup>-1</sup> (6% of the abundance in this month). In January and March 2002, none of these *taxa* were observed, just the Cyanophyceae *Anabaena flos-aqua* (8% and 7% of the *taxa*), with mean abundances of 12667 cell L<sup>-1</sup> (9%) and 15667 cell L<sup>-1</sup> (2%). During July 2002, *Asterionellopsis formosa* was the only freshwater species identified (6%), showing a mean abundance of 667 cell L<sup>-1</sup> (less than 1% of the abundance).

Most of phytoplankton *taxa* from Salgados lagoon were characteristic from freshwater *habitats* (23 *taxa*; 58%) and from freshwater/brackish *habitats* (11 *taxa*; 28%). Six of the 40 *taxa* (15%) occur in freshwater/brackish/marine *habitats*. No *taxa* from brackish/marine *habitats* were found in Salgados lagoon.

During the studied period, freshwater *taxa* presented the highest richness, except in March (3 *taxa*; 27% of the *taxa* in this month) and May 2002 (2 *taxa*; 22%), when a lower number of *taxa* was observed. The maximum values were registered in October 2001 (14 *taxa*; 64%) and July 2002 (15 *taxa*; 63%). The freshwater *taxa* category included four Cyanophyceae *taxa* and all Chlorophyceae and Euglenophyceae *taxa*.

Regarding abundance, the freshwater *taxa* category only dominated the phytoplankton community in June ( $295 \times 10^3$  cell L<sup>-1</sup>; 55%) and October 2001 ( $19956 \times 10^3$  cell L<sup>-1</sup>; 91%), although high abundances were determined also in August 2001 ( $11397 \times 10^3$  cell L<sup>-1</sup>; 10%) and July 2002 ( $23764 \times 10^3$  cell L<sup>-1</sup>; 10%). The Cyanophyceae *Anabaena flos-aqua* (127906 cell L<sup>-1</sup>; 24%), *Planktothrix* sp. (93414 cell L<sup>-1</sup>; 17%), the Chlorophyceae *Crucigenia quadrata* (33533 cell L<sup>-1</sup>; 6%) and the Euglenophyceae *Euglena caudate* (17246 cell L<sup>-1</sup>; 3%) were the freshwater *taxa* with greater relevance in June 2001, while in August 2001 the major *taxa* were *Planktothrix* sp. ( $7976130$  cell L<sup>-1</sup>; 7%) and the Chlorophyceae *Oocystis lacustris* ( $2055111$  cell L<sup>-1</sup>; 2%). In October 2001, the most abundant *taxa* were *Anabaena spiroides* ( $14346974$  cell L<sup>-1</sup>; 65%) and *Euglena oblonga* ( $1882654$  cell L<sup>-1</sup>; 9%). Other freshwater *taxa* presented lower abundances, but some *taxa* were also important, namely the Cyanophyceae *Planktothrix* sp. ( $1074981$  cell L<sup>-1</sup>; 5%) and *Chroococcus limneticus* ( $965758$  cell L<sup>-1</sup>; 4%) and the Chlorophyceae *Scenedesmus opoliensis* ( $296051$  cell L<sup>-1</sup>; 1%), *Coelastrum microporum* ( $287428$  cell L<sup>-1</sup>; 1%), *Oocystis lacustris* ( $265871$  cell L<sup>-1</sup>; 1%) and *Crucigenia quadrata* ( $255811$  cell L<sup>-1</sup>; 1%).

Freshwater/brackish *taxa* was the category with the highest richness in March (6 *taxa*; 55%) and May 2002 (5 *taxa*; 56%), presenting its maximum number of *taxa* in July

2002 (7 *taxa*; 29%) and the minimum in June 2001 (2 *taxa*; 20%) and December 2001 (2 *taxa*; 29%). This category comprised five Cyanophyceae, four Bacillariophyceae and the two Cryptophyceae *taxa*.

The abundances of freshwater/brackish *taxa* dominated the phytoplankton community in August ( $101496 \times 10^3$  cell L<sup>-1</sup>; 86%) and December 2001 ( $557 \times 10^3$  cell L<sup>-1</sup>; 89%), January ( $11859 \times 10^3$  cell L<sup>-1</sup>; 96%) and July 2002 ( $223898 \times 10^3$  cell L<sup>-1</sup>; 90%), also accounting the second most important percentages in June 2001 ( $209 \times 10^3$  cell L<sup>-1</sup>; 39%), March ( $1330 \times 10^3$  cell L<sup>-1</sup>; 39%) and May 2002 ( $950 \times 10^3$  cell L<sup>-1</sup>; 45%).

In June 2001, the Cryptophyceae *Rhodomonas* sp. ( $186828$  cell L<sup>-1</sup>; 35%), *Cryptomonas* sp. ( $10060$  cell L<sup>-1</sup>; 2%) and the Cyanophyceae *Lyngbya* sp. ( $11658$  cell L<sup>-1</sup>; 2%) were the main freshwater/brackish *taxa*. During August 2001, *Cryptomonas* sp. as well as *Lyngbya* sp. were absent and the most abundant *taxa* were the Cyanophyceae *Microcystis aeruginosa* ( $98784247$  cell L<sup>-1</sup>; 83%) and the Bacillariophyceae *Cyclotella* spp. ( $1940140$  cell L<sup>-1</sup>; 2%). *Cyclotella* spp. became the major *taxa* in December 2001 (59%), although its abundance was lower ( $370782$  cell L<sup>-1</sup>) than in October 2001. In January 2002, when the lagoon was connected to the sea, *Microcystis aeruginosa* dominated the phytoplankton community with an abundance of  $11833029$  cell L<sup>-1</sup> (96%). Besides *Microcystis aeruginosa*, two other freshwater/brackish *taxa* occurred in January 2002, *Cyclotella* spp. ( $22994$  cell L<sup>-1</sup>; < 1%) and *Rhodomonas* sp. ( $3449$  cell L<sup>-1</sup>; < 1%). During March and May 2002, the freshwater/brackish *taxa* with greater abundances were unidentified coccoid Cyanophyceae ( $855375$  cell L<sup>-1</sup> and  $596413$  cell L<sup>-1</sup>), which represented 25% and 28% of the abundance in each month. *Cyclotella* spp. was also registered in these months, with abundances of  $284554$  cell L<sup>-1</sup> (8%) and  $77606$  cell L<sup>-1</sup> (4%), respectively. In July 2002, there was bloom of *Microcystis aeruginosa* ( $217450334$  cell L<sup>-1</sup>), that accounted 87% of total abundance. During this month, *Cyclotella* spp. and the Cyanophyceae *Merismopedia punctate* reached their maximum abundances,  $2579667$  cell L<sup>-1</sup> and  $612023$  cell L<sup>-1</sup> respectively, nevertheless its contribution to the total abundance was very low (1% and < 1%).

Freshwater/brackish/marine *taxa* was the category with lower richness along time, with no *taxa* identified in December 2001 and oscillating between one taxon (10%) in January 2002 and four *taxa* in October 2001 (18%). This category included three Bacillariophyceae *taxa*, one Dinophyceae and pico-nano flagellate algae.

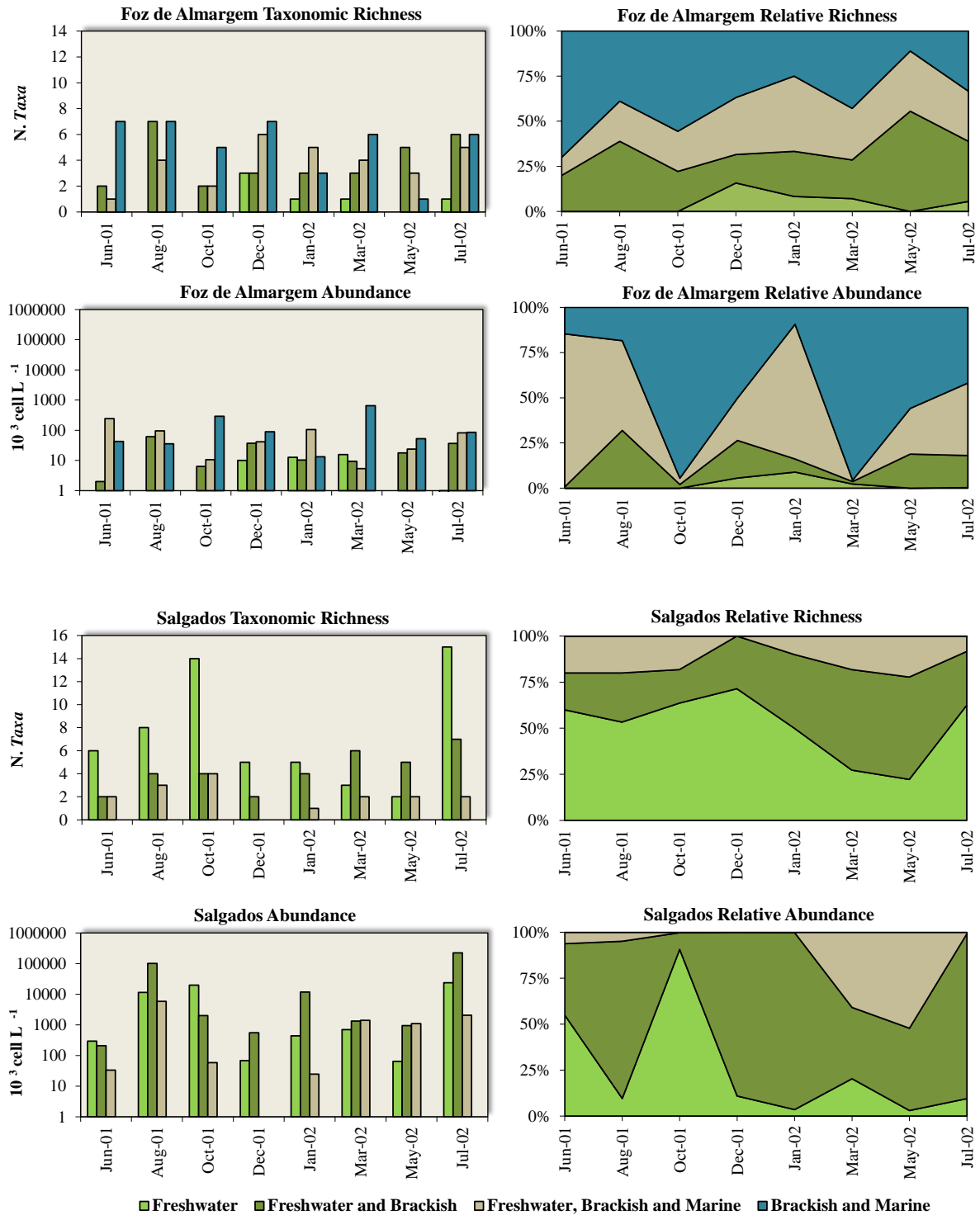


Figure 3.19 – Evolution of taxonomic richness and phytoplankton abundance according to *taxa* salinity preferences in Foz de Almagem and Salgados lagoons.

During the studied period, freshwater/brackish/marine *taxa* was the category with lower abundances, except in March ( $1403 \times 10^3 \text{ cell L}^{-1}$ ; 41%) and May 2002 ( $1108 \times 10^3 \text{ cell L}^{-1}$ ; 52%). The maximum abundance was achieved in August 2001 ( $5830 \times 10^3 \text{ cell L}^{-1}$ ) and in July 2002 ( $2062 \times 10^3 \text{ cell L}^{-1}$ ), abundance was also higher than in March and

May 2002, nevertheless it only represented 5% and approximately 1% of the abundance in these months.

Pico-nano flagellate algae presented the highest abundances in August 2001 (4598850 cell L<sup>-1</sup>; 4%), March (1362480 cell L<sup>-1</sup>; 40%) and May 2002 (1099412 cell L<sup>-1</sup>; 52%). *Gymnodinium* sp. was the major *taxa* in July 2002 (2062297 cell L<sup>-1</sup>; ≈ 1%) and in August 2001 registered an abundance of 1135341 cell L<sup>-1</sup> (≈ 1%).

### 3.3.3.5. Potentially harmful phytoplankton

In Foz de Almargem were identified four *taxa* that can be potentially harmful due to the production of biotoxins. Three of the *taxa* belong to Dinophyceae (*Gymnodinium* sp., *Prorocentrum minimum*, *Scrippsiella trochoidea*) and the fourth *taxon* is a Cyanophyceae (*Anabaena flos-aqua*).

*Gymnodinium* sp. was observed during all the studied period, with mean abundances ranging from 1000 cell L<sup>-1</sup> (March 2002) to 243667 cell L<sup>-1</sup> (June 2001) (Figure 3.20).

*Prorocentrum minimum* occurred during the all period, except in October 2001. The lowest mean abundance was determined in January 2002 (1333 cell L<sup>-1</sup>) and the maximum occurred in March 2002 (637000 cell L<sup>-1</sup>), nevertheless high values were also found in August 2001 (22333 cell L<sup>-1</sup>), May 2002 (52333 cell L<sup>-1</sup>) and July 2002 (58333 cell L<sup>-1</sup>).

*Scrippsiella trochoidea* was absent in January and May 2002, presenting mean abundances that varied between 667 cell L<sup>-1</sup> (March 2002) and 14333 cell L<sup>-1</sup> (June 2001).

*Anabaena flos-aqua* was identified only in January and March 2002, with mean abundances of 12667 cell L<sup>-1</sup> and 15667 cell L<sup>-1</sup>, respectively.

*Gymnodinium* sp. and *Prorocentrum minimum* were the potentially harmful *taxa* with higher mean abundances and a more regular presence in Foz de Almargem lagoon.

October 2001 and May 2002 were the months with lower number of potentially harmful *taxa* (two), while in March 2002 was registered the presence of the four *taxa*.

Considering the sum of mean abundances *per* month, October 2001 presented the lowest abundance of potentially harmful *taxa* (5000 cell L<sup>-1</sup>) and March 2002 (654333 cell L<sup>-1</sup>), June 2001 (266667 cell L<sup>-1</sup>) and January 2002 (104333 cell L<sup>-1</sup>) were the months with greater abundances of these *taxa*.

In Salgados lagoon, the number of potentially harmful *taxa* (six) was greater than in Foz de Almargem and most of the *taxa* presented higher mean abundances. Just one *taxon* belonged to Dinophyceae (*Gymnodinium* sp.) and the remaining *taxa* were Cyanophyceae (*Anabaena flos-aqua*, *Anabaena spiroides*, *Lyngbya* sp., *Microcystis aeruginosa*, *Planktothrix* sp.).

*Gymnodinium* sp. was registered from June to October 2001, in May and July 2002. The lower mean abundances were determined in October 2001 and May 2002 (8623 cell L<sup>-1</sup>), while the higher values were found in August 2001 (1135341 cell L<sup>-1</sup>) and July 2002 (2062297 cell L<sup>-1</sup>). The occurrence of *Gymnodinium* sp. in Salgados lagoon was less regular than in Foz de Almargem, but mean abundances were much higher.

*Anabaena flos-aqua* was observed only in June and August 2001, with mean abundances of 127906 cell L<sup>-1</sup> and 488628 cell L<sup>-1</sup>, respectively. Abundances were higher than in Foz de Almargem and the period of occurrence was different.

Another *Anabaena* species was identified in Salgados lagoon, *Anabaena spiroides*, but its presence was registered in different months than *Anabaena flos-aqua*, October 2001 (maximum: 14346974 cell L<sup>-1</sup>), December 2001 (minimum: 44882 cell L<sup>-1</sup>) and July 2002 (837888 cell L<sup>-1</sup>).

*Lyngbya* sp. was also present in October 2001 (maximum: 610785 cell L<sup>-1</sup>) and July 2002 (561021 cell L<sup>-1</sup>), besides June 2001 (minimum: 11658 cell L<sup>-1</sup>).

*Microcystis aeruginosa* was the species with higher mean abundances, although it occurred only in August 2001 (98784247 cell L<sup>-1</sup>), January 2002 (minimum: 11833029 cell L<sup>-1</sup>) and July 2002 (maximum: 217450334 cell L<sup>-1</sup>).

*Planktothrix* sp. was the *taxon* which presented a more regular occurrence in Salgados lagoon, being absent only in December 2001 and March 2002. Lower mean abundances were determined in January 2001 (68780 cell L<sup>-1</sup>) and May 2002 (minimum: 63234 cell L<sup>-1</sup>), while the higher mean abundances were observed in August 2001 (maximum: 7976130 cell L<sup>-1</sup>), October 2001 (1074981 cell L<sup>-1</sup>) and July 2002 (1311478 cell L<sup>-1</sup>).

In March 2002, no potentially harmful *taxa* were identified in Salgados lagoon; in December 2001 and May 2002 was registered the presence of one *taxon* in each month, *Anabaena spiroides* and *Planktothrix* sp. respectively; while in July 2002 were found five *taxa* (*Gymnodinium* sp., *Anabaena spiroides*, *Lyngbya* sp., *Microcystis aeruginosa* and *Planktothrix* sp.). From June till October 2001, the number of potentially harmful *taxa* was also high (four *taxa*).

If the sum of mean abundances *per* month is considered, December 2001 and May 2002 were also the months with lower abundances of potentially harmful *taxa* (44882 cell L<sup>-1</sup> and 71857 cell L<sup>-1</sup>) and the higher abundances were determined in August 2001 (108384346 cell L<sup>-1</sup>) and July 2002 (222223019 cell L<sup>-1</sup>).

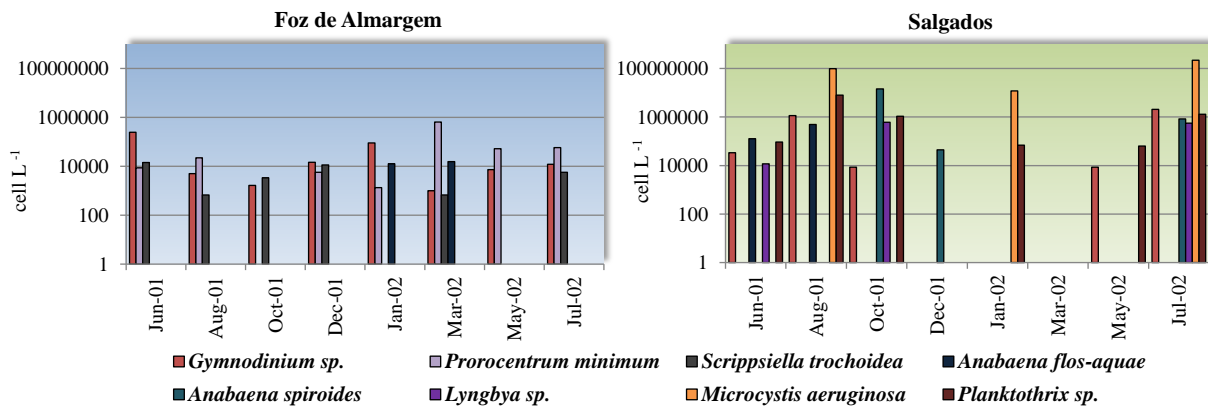


Figure 3.20 - Monthly mean abundances of potentially harmful *taxa* in Foz de Almargem and Salgados lagoons.

The Cyanophyceae *Anabaena flos-aqua*, *Anabaena spiroides* and *Planktothrix sp.* are generally associated to freshwater *habitats*, while *Microcystis aeruginosa* and *Lyngbya* species can be found in freshwater and brackish environments. The Dinophyceae *Prorocentrum minimum* and *Scrippsiella trochoidea* usually occur in brackish and marine *habitats*. *Gymnodinium* may inhabit freshwater, brackish and marine environments.

In Foz de Almargem, during January and March 2002 was registered a potentially harmful species typical from freshwater *habitats*, *Anabaena flos-aqua* (Figure 3.21). Most of the time, brackish/marine *taxa* presented higher abundances, with the maximum value being determined in March 2002 (637667 cell L<sup>-1</sup>). Nevertheless, in June 2001 and January 2002, higher abundances corresponded to *Gymnodinium* species, that can live in different types of *habitats*, from freshwater to marine.

In Salgados lagoon, none of the potentially harmful *taxa* were from brackish and marine environments. Freshwater and brackish *taxa* were represented by *Microcystis aeruginosa* and *Lyngbya sp.* *Microcystis aeruginosa* was responsible for the highest abundances of potentially harmful *taxa* in August 2001, January 2002 and July 2002, whereas *Lyngbya sp.* was the freshwater/brackish *taxon* present in June and October 2001, being also observed in July 2002.

In June, October and December 2001, just as in May 2002, higher abundances were determined for freshwater *taxa* (*Anabaena flos-aqua*, *Anabaena spiroides*, *Planktothrix* sp.). In December 2001, just a freshwater *taxon* was present, *Anabaena spiroides*. *Gymnodinium* sp., the only freshwater/brackish/marine *taxon* found in Salgados lagoon, presented lower abundances than the other potentially harmful *taxa* and was absent from December 2001 till March 2002.

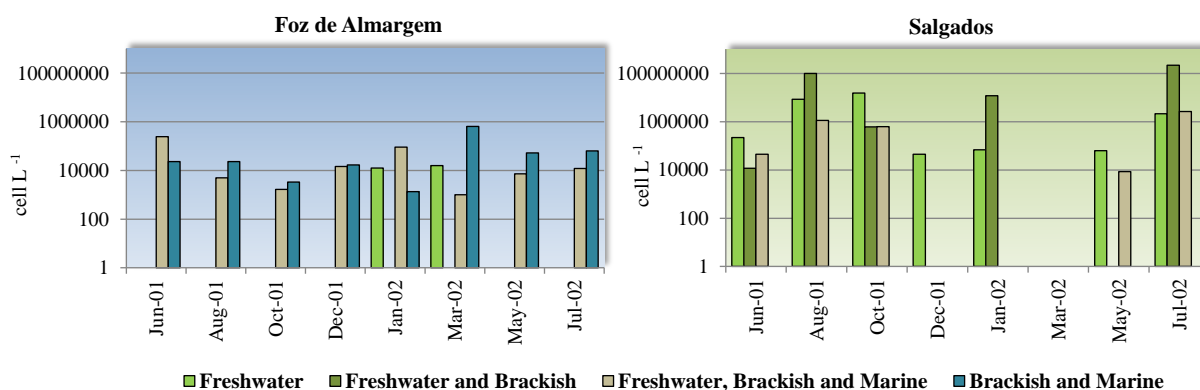


Figure 3.21 - Monthly mean abundances of potentially harmful *taxa* in Foz de Almargem and Salgados lagoons, according to *taxa* occurrence in terms of *habitats*.

Most of the potentially harmful *taxa* presented significant correlations with orthophosphates and total phosphorus concentrations (Table 3.11). The Dinophyceae species *Prorocentrum minimum* and *Scrippsiella trochoidea* were negatively correlated with these nutrients, while the Cyanophyceae *Anabaena spiroides*, *Lyngbya* sp., *Microcystis aeruginosa* and *Planktothrix* sp. had positive correlations.

Significant correlations were also found between the majority of *taxa* and at least one of the nitrogen compounds. Dinophyceae *taxa* showed negative correlations with nitrates (*Gymnodinium* sp., *Scrippsiella trochoidea*), nitrites (*Gymnodinium* sp.) and ammonia (*Prorocentrum minimum*) concentrations. Some Cyanophyceae *taxa* were positively correlated with ammonia concentration (*Anabaena spiroides*, *Microcystis aeruginosa* and *Planktothrix* sp.) and total dissolved inorganic nitrogen (*Microcystis aeruginosa* and *Planktothrix* sp.).

To what concerns the N: P ratio, *Prorocentrum minimum* and *Scrippsiella trochoidea* presented positive correlations, while *Anabaena spiroides* and *Lyngbya* sp. showed negative correlations.

Negative linear associations were also found between some other environmental parameters and potentially harmful *taxa*, namely *Gymnodinium* sp. and cumulative

rainfall; *Prorocentrum minimum* and total solids in suspension; *Scrippsiella trochoidea* and water level; *Anabaena spiroides* and salinity; *Planktothrix* sp. and salinity.

On the other hand, positive correlations were observed between *Anabaena spiroides*, water level and pH; *Planktothrix* sp. and total solids in suspension.

Table 3.11 - Significant correlations between environmental parameters and potentially harmful *taxa* from Foz de Almargem and Salgados lagoons. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Phytoplankton taxa</i>	<i>Environmental parameters</i>	<i>Results</i>
<i>Gymnodinium</i> sp.	Cumulative rainfall	Rho = -0.357; <i>p</i> = 0.013 *
	Nitrates concentration	Rho = -0.328; <i>p</i> = 0.023 *
	Nitrites concentration	Rho = -0.371; <i>p</i> = 0.009 **
<i>Prorocentrum minimum</i>	Ammonia concentration	Rho = -0.350; <i>p</i> = 0.015 *
	Orthophosphates concentration	Rho = -0.663; <i>p</i> < 0.001 **
	Total Phosphorus concentration	Rho = -0.603; <i>p</i> < 0.001 **
	N: P ratio	Rho = 0.677; <i>p</i> < 0.001 **
	Total solids in suspension	Rho = -0.543; <i>p</i> < 0.001 **
<i>Scrippsiella trochoidea</i>	Water level	Rho = -0.298; <i>p</i> = 0.040 *
	Nitrates concentration	Rho = -0.308; <i>p</i> = 0.033 *
	Orthophosphates concentration	Rho = -0.430; <i>p</i> = 0.002 **
	Total Phosphorus concentration	Rho = -0.450; <i>p</i> = 0.001 **
	N: P ratio	Rho = 0.339; <i>p</i> = 0.018 *
<i>Anabaena spiroides</i>	Water level	Rho = 0.301; <i>p</i> = 0.037 *
	pH	Rho = 0.354; <i>p</i> = 0.014 *
	Salinity	Rho = -0.487; <i>p</i> < 0.001 **
	Ammonia concentration	Rho = 0.306; <i>p</i> = 0.034 *
	Orthophosphates concentration	Rho = 0.361; <i>p</i> = 0.012 *
	Total Phosphorus concentration	Rho = 0.375; <i>p</i> = 0.009 **
	N: P ratio	Rho = -0.401; <i>p</i> = 0.005 **
<i>Lyngbya</i> sp.	Orthophosphates concentration	Rho = 0.312; <i>p</i> = 0.031 *
	Total Phosphorus concentration	Rho = 0.328; <i>p</i> = 0.023 *
	N: P ratio	Rho = -0.369; <i>p</i> = 0.010 **
<i>Microcystis aeruginosa</i>	Ammonia concentration	Rho = 0.420; <i>p</i> = 0.003 **
	Total dissolved inorganic nitrogen concentration	Rho = 0.394; <i>p</i> = 0.006 **
	Orthophosphates concentration	Rho = 0.481; <i>p</i> = 0.001 **
	Total Phosphorus concentration	Rho = 0.479; <i>p</i> = 0.001 **
<i>Planktothrix</i> sp.	Salinity	Rho = -0.305; <i>p</i> = 0.035 *
	Ammonia concentration	Rho = 0.519; <i>p</i> < 0.001 **
	Total dissolved inorganic nitrogen concentration	Rho = 0.297; <i>p</i> = 0.041 *
	Orthophosphates concentration	Rho = 0.523; <i>p</i> < 0.001 **
	Total Phosphorus concentration	Rho = 0.533; <i>p</i> < 0.001 **
	N: P ratio	Rho = -0.293; <i>p</i> = 0.043 *
	Total solids in suspension	Rho = 0.378; <i>p</i> = 0.008 **

The strongest and more relevant correlations were determined between:

- *Gymnodinium* sp. and nitrites concentration (-);
- *Prorocentrum minimum* and N: P ratio (+), orthophosphates concentration (-), total phosphorus concentration (-), total solids in suspension (-);
- *Scrippsiella trochoidea* and total phosphorus concentration (-), orthophosphates concentration (-);
- *Anabaena spiroides* and salinity (-), N: P ratio (-), total phosphorus concentration (+);
- *Lyngbya* sp. and N: P ratio (-);
- *Microcystis aeruginosa* and orthophosphates concentration (+), total phosphorus concentration (+), ammonia concentration (+), total dissolved inorganic nitrogen (+);
- *Planktothrix* sp. and total phosphorus concentration (+), orthophosphates concentration (+), ammonia concentration (+), total solids in suspension (+).

### 3.3.3.6. Phytoplankton communities and water quality

During the studied period, the highest phytoplankton abundance in Foz de Almagem (March 2002:  $681.42 \times 10^3$  cell L<sup>-1</sup>) did not reach the bloom thresholds defined for semi-enclosed lagoons by Pereira Coutinho *et al.* (2012) in the open period ( $2.5 \times 10^6$  cell L<sup>-1</sup>) and the closed period ( $6 \times 10^6$  cell L<sup>-1</sup>).

In Salgados lagoon, when the lagoon was in connection with the sea phytoplankton abundance (January 2002:  $12.322 \times 10^6$  cell L<sup>-1</sup>) was above the threshold of  $2.5 \times 10^6$  cell L<sup>-1</sup> and during the period of isolation abundances were higher than  $6 \times 10^6$  cell L<sup>-1</sup> in August 2001 ( $118.722 \times 10^6$  cell L<sup>-1</sup>), October 2001 ( $21.976 \times 10^6$  cell L<sup>-1</sup>) and July 2002 ( $249.724 \times 10^6$  cell L<sup>-1</sup>).

According to the water quality assessment based on phytoplankton blooms frequency, proposed by Pereira Coutinho *et al.* (2012), during the sampling periods lagoons were closed (June-December 2001; March-July 2002), water quality was high in Foz de Almagem (blooms percentage < 33.3%) and moderate (41.7%-50%) in Salgados lagoon (blooms percentage = 42.9%). Blooms frequency was determined considering seven sampling periods with the lagoons isolated from the sea. Water quality when the

lagoons were opened (January 2002) could not be determined as there were not enough data.

The classification of water quality in Foz de Almargem according to the criteria defined by Pereira Coutinho *et al.* (2012) is the same (high water quality), whether it is based on the frequency of phytoplankton blooms or on the 90<sup>th</sup> percentile of chlorophyll *a*.

For Salgados lagoon, the classification of water quality differs if the frequency of phytoplankton blooms is considered (moderate water quality) and if it is based on the 90<sup>th</sup> percentile of chlorophyll *a* (bad water quality). Given the occurrence of six potentially harmful *taxa* when the lagoon was closed and the high abundances registered during some of the blooms (*e.g.* August 2001: *Microcystis aeruginosa*, *Planktothrix* sp., *Gymnodinium* sp.; October 2001: *Anabaena spiroides*; *Planktothrix* sp.; July 2002: *Microcystis aeruginosa*, *Planktothrix* sp., *Gymnodinium* sp.), it seems more appropriate to classify the water quality as bad.

Brito *et al.* (2012) defined a threshold of  $1.5 \times 10^6$  cell L<sup>-1</sup> for phytoplankton blooms during the growing season (February to October). This value is lower than the threshold considered by Pereira Coutinho *et al.* (2012) when the lagoons are closed to the sea and therefore in Foz de Almargem there was no phytoplankton bloom during the studied period. In Salgados lagoon, total phytoplankton presented monthly mean values higher than  $1.5 \times 10^6$  cell L<sup>-1</sup> in August and October 2001, March, May and July 2002; and as referred before, the blooms in August, October 2001 and July 2002 included high abundances of potentially harmful *taxa*. Brito *et al.* (2012) did not propose water quality categories based on blooms frequency.

## **4. BENTHIC MACROINVERTEBRATE COMMUNITIES**

### **4.1. Specific Aims**

The study of benthic macroinvertebrate communities was planned to achieve four specific aims:

1. Characterize seasonal variation of benthic macroinvertebrate communities; compare spatial patterns along the gradient of distance to the sea and between lagoons.
2. Study the relations between benthic macroinvertebrate communities and environmental parameters in the two lagoons, namely hydrological, water and sediment parameters.
3. Evaluate the effect of lagoons opening in benthic macroinvertebrate communities.
4. Evaluate the relation between benthic macroinvertebrate communities, water quality and trophic state in the lagoons.

### **4.2. Material and Methods**

#### **4.2.1. Field sampling and laboratory procedures**

Field work was done in the same periods as sediment monitoring (June 2001 - July 2002).

In Foz de Almagem sampling took place in the three stations previously described (E1- Upstream; E2- Intermediate; E3- Downstream). In March 2002, no samples were collected as the water level was in its maximum and sampling stations were not reachable.

In Salgados only two sampling stations were considered, upstream (E1) and downstream (E3). The intermediate station (E2) was difficult to access due to the channel depth and high density of emergent vegetation.

Three core samples of sediments ( $\approx 0.01 \text{ m}^2$  each) were taken at the sampling stations, with a 12 cm internal diameter corer and to a depth of approximately 20 cm. Samples

were washed through a 1 mm sieving bag and the remaining sediments and macrofauna were preserved in a solution of 70% ethanol coloured with Rose Bengal. In laboratory, each sample was processed individually; organisms were counted and identified to the species level or the lowest taxonomic level possible. Identification was undertaken with reference to various authors, namely Fauvel (1975), Hayward and Ryland (1995), Naylor (1972), Poppe and Goto (1993), Rield (1983), Tachet *et al.* (2002).

#### 4.2.2. Data analysis

The characterization of benthic macroinvertebrate communities was based on taxonomic composition, *taxa* density and richness, diversity - Shannon-Wiener diversity index – (Shannon and Weaver, 1963) and evenness (Pielou, 1966).

Macroinvertebrate density in each station was determined for a total area of 0.03 m<sup>2</sup>. Richness included all different *taxa* found in the three samples.

*Taxa* were classified in terms of trophic groups, following the same criteria as Gamito (2008) and references therein:

- Suspension feeders (plankton and detritus);
- Deposit feeders (detritus and microphytobenthos);
- Suspension and deposit feeders (species which have the two feeding strategies depending on food availability);
- Herbivorous (macroalgae and macrophytes);
- Carnivorous, parasites, omnivorous and scavengers; most carnivores are also omnivores and scavengers, thereby were grouped together.

For each station, constancy and fidelity indexes were determined according to the formulae and approach used by Cancela da Fonseca (1989). The constancy of a *taxon* in each lagoon, during the sampled months was also calculated.

The constancy (C) and fidelity (F) of a *taxon* to a station was calculated by the formulae:

$$C = \frac{\text{Number of samples collected in a specific station containing the } \textit{taxon}}{\text{Total number of samples in that station}} \times 100$$

$$F = \frac{\text{Constancy of the } \textit{taxon} \text{ in that station}}{\text{Sum of the constancies of the } \textit{taxon} \text{ in all stations}} \times 100$$

*Taxa* were classified into categories of constancy and fidelity (Cancela da Fonseca, 1989).

Constancy categories:

- 1) Constant *taxon* ( $C \geq 50$ )
- 2) Accessory *taxon* ( $10 \leq C < 50$ )
- 3) Accidental *taxon* ( $C < 10$ )

Fidelity categories:

- 1) Exclusive *taxon* ( $F > 90$ )
- 2) Elective *taxon* ( $67 < F \leq 90$ )
- 3) Preferential *taxon* ( $50 < F \leq 67$ )
- 4) Accessory *taxon* ( $20 < F \leq 50$ )
- 5) Accidental /rare *taxon* ( $F \leq 20$ )

*Taxa* were also classified according to the biotic classes of salinity preference and tolerance for macroinvertebrates, defined by Wolf *et al.* (2009):

- Limnic - freshwater *taxa*, do not tolerate even low salinity;
- Limnic, tolerates salt - freshwater *taxa*, tolerate salinity below 5‰;
- Euryhaline-limnic - freshwater *taxa*, tolerate salinity up to 10‰ (even higher salinity for a short time);
- Brackish - brackish water *taxa*, permanently living and reproducing in brackish waters, tolerate varying salinity between 0.5 and 30‰; according to Remane and Schlieper (1971), these *taxa* originate from fresh or marine waters, but are now confined to brackish waters;
- Euryhaline marine - marine *taxa* with a wide affinity for salinity, tolerate salinity between 0.5 and 35‰;
- Holeuryhaline - *taxa* with marine origin, but tolerate the entire range of salinity from freshwater to seawater.

Benthic macroinvertebrate parameters from a period of lagoons isolation (December 2001) were compared with the data collected when the lagoons were in connection with the sea (January 2002).

The variation among sampling stations within each lagoon was first analysed and afterwards, the mean values from the lagoons were compared.

The One-Way ANOVA and the Student T test were the statistical tests applied to analyse differences in data among stations and differences between the lagoons, when data presented normality and homogeneity of variances. Previously, a logarithmic transformation of data was done ( $\ln x + 1$ ) and if data distribution did not present normality or variances were not homogeneous, the non-parametric tests of Kruskal-

Wallis and the Mann-Whitney U were chosen. The LSD Fisher multiple comparison test was used to determine which of the three stations differed significantly (Maroco, 2010). The confidence level used in all statistical tests was 95 % ( $\alpha = 0.05$ ).

Canonical correspondence analyses (CCA) were applied to data of each sampling station and to the mean values of each lagoon, in order to see how the benthic macroinvertebrate communities were associated with the environmental variables studied. Benthic macroinvertebrate data was presented as the density of the main *taxa*. Data were previously root transformed to reduce the importance of abundant *taxa* and increase the relevance of rare *taxa*. Environmental variables were centred and standardized as they presented different measurement units. Just the environmental that the PCA previously performed for each lagoon and for the mean values of the lagoons showed not to be highly correlated were included in CCA (see Chapter 2).

Correlations between environmental parameters, benthic macroinvertebrate densities and indexes were also evaluated by Pearson or Spearman coefficients, according to data normality after transformation ( $\ln x+1$ ).

The evaluation of water quality based on benthic macroinvertebrate communities could be made using the Benthic Assessment Tool (BAT) (Teixeira *et al.*, 2009) or the M-AMBI index (Muxica *et al.*, 2007). However, these multimetric indices require the use of specific reference conditions defined for each water body typology (Bettencourt *et al.*, 2004; Borja *et al.* 2004; Muxica *et al.*, 2007; Teixeira *et al.*, 2008). M-AMBI has been applied to leaky and restricted lagoons but not to choked lagoons such as Foz de Almargem or Salgados. BAT was applied to Ria Formosa coastal lagoon (Gamito *et al.*, 2012), where the reference conditions were defined according to the different *habitats*: channels, muddy or sandy banks, and seagrasses beds. However, since no reference conditions were defined for choked lagoons yet, only a general analysis of the ecological groups was done, according to their sensitivity to an increasing stress gradient (*i.e.* increasing organic matter enrichment).

These groups have been summarized by Grall and Glémarec (1997), as defined below.

- *Group I.* Species very sensitive to organic enrichment and present under unpolluted conditions (initial state). They include the specialist carnivores and some deposit-feeding tubicolous polychaetes.

- *Group II.* Species indifferent to enrichment, always present in low densities with non-significant variations with time (from initial state, to slight unbalance). These include suspension feeders, less selective carnivores and scavengers.
- *Group III.* Species tolerant to excess organic matter enrichment. These species may occur under normal conditions, but their populations are stimulated by organic enrichment (slight unbalance situations). They are surface deposit-feeding species, as tubicolous spionids.
- *Group IV.* Second-order opportunistic species (slight to pronounced unbalanced situations). Mainly small sized polychaetes: subsurface deposit-feeders, such as cirratulids.
- *Group V.* First-order opportunistic species (pronounced unbalanced situations). These are deposit-feeders, which proliferate in reduced sediments.

The classification of each *taxon* was obtained from Borja *et al.* (2000), actualized with the species ecological group list available at <http://ambi.azti.es/>.

The softwares used for data analysis were CANOCO (Ter Braak, version 4.54, 1988-2005 Biometris) and SPSS (version 19, IBM SPSS Statistics).

## 4.3. Results and discussion

### 4.3.1. Foz de Almargem coastal lagoon

#### 4.3.1.1. Benthic macroinvertebrate communities

In Foz de Almargem were identified ten *taxa* of benthic macroinvertebrates (Appendix II.C), which included Polychaeta (3), Oligochaeta (1), Crustacea Isopoda (1), Mollusca Gastropoda (2), Mollusca Bivalvia (2) and Insecta Diptera (1).

The Mollusca Gastropoda *Hydrobia ulvae* represented 80% of the total benthic macro invertebrates identified during the studied period and was registered in all samplings. Five *taxa* were present at least in 50% of the samplings (*Lekanesphaera hookeri*: 86%; *Hediste diversicolor*: 76%; *Chironomus* sp.: 76%; *Abra segmentum*: 52%; *Cerastoderma glaucum*: 52%) and two other *taxa* occurred in more than 25% of the samplings (*Ventrosia ventrosa*: 48%; *Capitella capitata*: 33%).

Taxonomic richness varied from 2 (intermediate station in June 2001) to 9 (upstream station in August 2001) (Figure 4.1). In August 2001, intermediate and downstream stations had the same *taxa* richness (7), although species composition was not the same. Similar *taxa* richness was also found in the upstream and intermediate stations during January 2002 (3 *taxa*), and in the upstream and downstream stations during July 2002 (4 *taxa*). In May 2002, all stations presented the same *taxa* richness (6 *taxa*) and composition. The greater variation in the number of *taxa* (6) was determined in the upstream station.

Taxonomic richness was positively correlated with Polychaeta density ( $\rho = 0.821$ ;  $p < 0.001$ ) and Bivalvia density ( $\rho = 0.659$ ;  $p = 0.001$ ).

Benthic macroinvertebrate densities presented values from 324 ind m<sup>-2</sup> (intermediate station in June 2001) to 77552 ind m<sup>-2</sup> (downstream station in January 2002), when the lagoon was connected to the sea. During most of the studied period densities were under 30000 ind m<sup>-2</sup>. The intermediate station registered the lowest variation in time (20737 ind m<sup>-2</sup>), while the downstream station showed the greatest variation (76106 ind m<sup>-2</sup>).

Macroinvertebrate density was positively correlated with Gastropoda density ( $\rho = 0.927$ ;  $p < 0.001$ ).

The Shannon-Wiener diversity index ranged from 0.07 bits (downstream station in January 2002) to 1.74 bits (intermediate station in July 2002) and the major seasonal variations were determined in the intermediate (1.31 bits) and downstream (1.30 bits) stations. The upstream station presented higher diversity except in October 2001 and July 2002, when the intermediate station registered greater values.

Shannon-Wiener diversity was positively correlated with Insecta density ( $\rho = 0.731$ ;  $p < 0.001$ ), Bivalvia density ( $\rho = 0.531$ ;  $p = 0.013$ ) and it was negatively correlated with Gastropoda density ( $r = -0.536$ ;  $p = 0.012$ ).

Evenness was minimum downstream (0.03) and maximum upstream (0.81) in January 2002. The upstream and intermediate stations had lower seasonal variation (0.53 and 0.58), whereas the downstream station showed the greatest variation along time (0.65). A negative correlation was determined between evenness and Gastropoda density ( $r = -0.790$ ;  $p < 0.001$ ), and a positive correlation was found with Insecta density ( $\rho = 0.500$ ;  $p = 0.021$ ).

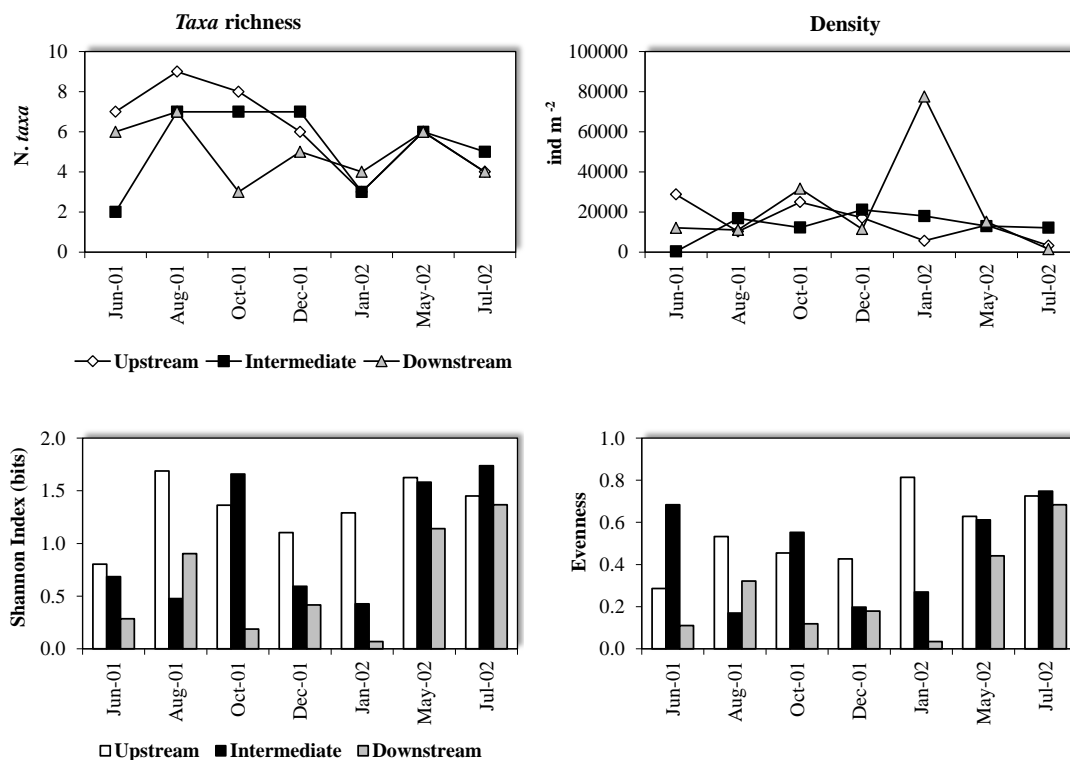


Figure 4.1 - Evolution of total benthos density, taxa richness, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Foz de Almagem sampling stations.

Most of the time, the three stations were dominated by Mollusca, except from January to July 2002 in the upstream station and during July 2002 in the intermediate and downstream stations (Figure 4.2). The maximum densities of Mollusca in the stations occurred in June 2001 (Upstream: 28171 ind m<sup>-2</sup>), December 2001 (Intermediate: 19794 ind m<sup>-2</sup>) and January 2002 (Downstream: 77021 ind m<sup>-2</sup>). Crustacea were absent from all stations in June 2001 and Oligochaeta were found only in May 2002.

In the upstream station, Mollusca densities went from 973 ind m<sup>-2</sup> (July 2002) to 28171 ind m<sup>-2</sup> (June 2001). Until December 2001, Mollusca accounted 74 % (October 2001: 18496 ind m<sup>-2</sup>) to 98% (June 2001: 28171 ind m<sup>-2</sup>) of total benthic macroinvertebrates. In January 2002, there was an increase in Crustacea density and the community was mostly represented by Crustacea (2714 ind m<sup>-2</sup>; 48%) and Mollusca (2537 ind m<sup>-2</sup>; 45%). In May 2002, Crustacea reached the maximum density (7876 ind m<sup>-2</sup>; 59%) and Mollusca density (4749 ind m<sup>-2</sup>) comprised 36% of benthic macroinvertebrates. Both Mollusca (973 ind m<sup>-2</sup>; 31%) and Crustacea (383 ind m<sup>-2</sup>; 12 %) densities decreased in

July 2002 and there was an increase of Insecta (1829 ind m<sup>-2</sup>; 57 %). Besides Oligochaeta, no Polychaeta *taxa* were collected in January and July 2002.

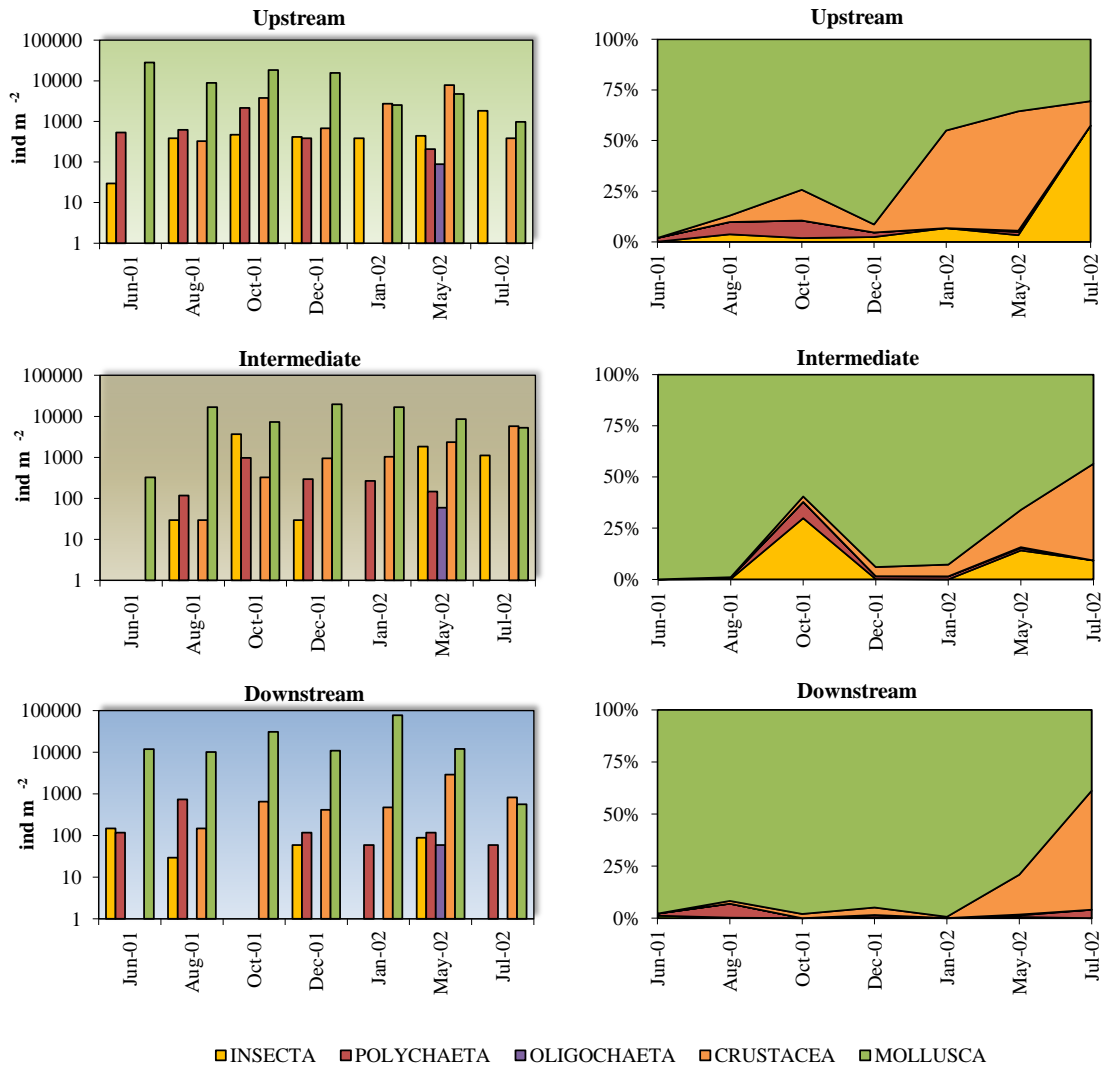


Figure 4.2 - Seasonal variation of the main taxonomic benthos groups densities and relative frequency of each group in Foz de Almagem sampling stations.

In the intermediate station, the range of Mollusca density was lower than upstream varying from 324 ind m<sup>-2</sup> in June 2001 to 19794 ind m<sup>-2</sup> in December 2001. Nevertheless, it was the major group all over the studied period, with the exception of July 2002 (5280 ind m<sup>-2</sup>; 44 %), when Crustacea presented the maximum density of 5723 ind m<sup>-2</sup> (47%). October 2001 was the month with highest densities of Insecta (3658 ind m<sup>-2</sup>) and Polychaeta (973 ind m<sup>-2</sup>), accounting 30% and 8% respectively of the total density. No Insecta were found in June 2001 and January 2002, while Polychaeta *taxa* were absent in June 2001 and July 2002.

Downstream, Mollusca densities were much higher than the densities of other taxonomic groups, just in July 2002 Crustacea showed a greater density (826 ind m<sup>-2</sup>; 57%). Compared to the upstream and intermediate stations, Mollusca minimum (July 2002: 560 ind m<sup>-2</sup>; 38%) and maximum densities (January 2002: 77021 ind m<sup>-2</sup>; 99%) were greater downstream. Contrary to what happened in the other stations, Insecta and Polychaeta were absent in October 2001 in the downstream station. Similarly to the intermediate station, no Insecta were collected downstream in January 2002.

Class Insecta was represented just by the Diptera *Chironomus* sp.

In terms of constancy, *Chironomus* sp. was classified as a constant *taxon* in all stations as it was present in more than 50% of samples of each station, nevertheless constancy decreased from the upstream to downstream station ( $C_{\text{Upstream}} = 100\%$ ;  $C_{\text{Intermediate}} = 71,4\%$ ;  $C_{\text{Downstream}} = 57,1\%$ ). To what concerns fidelity, it was considered an accessory *taxon* ( $20 < F < 50$ ) in all stations, but with fidelity values decreasing from the upstream to downstream station ( $F_{\text{Upstream}} = 43,7\%$ ;  $F_{\text{Intermediate}} = 31,2\%$ ;  $F_{\text{Downstream}} = 25,0\%$ ).

During some of the samplings *Chironomus* sp. was absent (June 2001: intermediate; October 2001: downstream; January 2002: intermediate and downstream; July 2002: downstream) and it reached its maximum density (3658 ind m<sup>-2</sup>) in October 2001 (intermediate station) (Figure 4.2). The intermediate station presented the greatest mean density (952 ind m<sup>-2</sup>), followed by the upstream station (565 ind m<sup>-2</sup>) and the downstream station (46 ind m<sup>-2</sup>).

*Lekanesphaera hookeri* was the only Crustacea Isopoda found in Foz de Almargem soft-bottoms. It was classified as constant *taxon* presenting similar constancy in the three stations ( $C = 85,7\%$ ) and as accessory *taxon*, with all stations showing the same fidelity ( $F = 33,3\%$ ).

In June 2001 none of the stations presented *Lekanosphaera hookeri*, but after that, densities ranged from 29 ind m<sup>-2</sup> (August 2001: intermediate) to 7876 ind m<sup>-2</sup> (May 2002: upstream), with mean densities increasing along the gradient of distance to the sea (Upstream: 2250 ind m<sup>-2</sup>; Intermediate: 1488 ind m<sup>-2</sup>; Downstream: 771 ind m<sup>-2</sup>).

Class Polychaeta was represented by two species, *Capitella capitata* and *Hediste diversicolor*. No Polychaeta were collected during the samplings of June 2001 (Intermediate), October 2001 (Downstream), January (Upstream) nor July (Upstream)

and Intermediate) and the maximum density was registered at the upstream station in October 2001 (2153 ind m<sup>-2</sup>).

*Capitella capitata* was considered a constant *taxon* in the upstream station ( $C_{\text{Upstream}} = 57,1\%$ ) and an accessory *taxon* ( $10 \leq C < 50$ ) in the intermediate and downstream stations ( $C_{\text{Intermediate}} = 14,3\%$ ;  $C_{\text{Downstream}} = 28,6\%$ ). Regarding fidelity, it was the only preferential *taxon* ( $50 < F \leq 67$ ) at the upstream station ( $F_{\text{Upstream}} = 57,1\%$ ), being classified as accidental or rare *taxon* ( $F \leq 20$ ) in the intermediate station ( $F_{\text{Intermediate}} = 14,3\%$ ) and as an accessory *taxon* in the downstream station ( $F_{\text{Downstream}} = 28,6\%$ ). The species was collected in the upstream station from June till December 2001 and the maximum density (2035 ind m<sup>-2</sup>) was reached in October (Figure 4.3). In the intermediate station, it was present only in October (944 ind m<sup>-2</sup>) and in the downstream station it was identified in June (29 ind m<sup>-2</sup>) and August 2001 (678 ind m<sup>-2</sup>). No specimens were found in the samples taken from January to July 2002. Mean densities decreased from the upstream station (383 ind m<sup>-2</sup>) to the intermediate (135 ind m<sup>-2</sup>) and the downstream (101 ind m<sup>-2</sup>) stations.

*Hediste diversicolor* was classified as a constant *taxon* in all sampling stations ( $C_{\text{Upstream}} = C_{\text{Intermediate}} = 71,4\%$ ;  $C_{\text{Downstream}} = 85,7\%$ ) and an accessory *taxon* ( $F_{\text{Upstream}} = F_{\text{Intermediate}} = 31,3\%$ ;  $F_{\text{Downstream}} = 37,5\%$ ), with intermediate and downstream stations showing the same constancy and fidelity values. The species was absent from some samples (June 2001: intermediate; October 2001: downstream; January 2002: upstream; July 2002: upstream and intermediate) and presented its maximum density in June 2001 (upstream station: 442 ind m<sup>-2</sup>). Just as *Capitella capitata*, mean densities decreased with station proximity to the sea (Upstream: 160 ind m<sup>-2</sup>; Intermediate: 122 ind m<sup>-2</sup>; Downstream: 72 ind m<sup>-2</sup>).

Four Mollusca species were identified in Foz de Almargem lagoon, belonging to class Gastropoda (*Hydrobia ulvae* and *Ventrosia ventrosa*) and to class Bivalvia (*Cerastoderma glaucum* and *Abra segmentum*). The minimum and maximum Gastropoda densities were observed downstream (July 2002: 501 ind m<sup>-2</sup>; January 2002: 76991 ind m<sup>-2</sup>). The highest Bivalvia density was registered upstream (August 2001: 6490 ind m<sup>-2</sup>), but in December 2001 and January 2002 no specimens were found in this station. In January 2002, Bivalvia were also absent from the intermediate station and in October and December 2001 there were no observations downstream.

*Hydrobia ulvae* was constant in the three stations, presenting a constancy of 100%. Species fidelity at each station was 33.3%, meaning that the species was accessory. Densities went from 265 ind m<sup>-2</sup> in June 2001 (intermediate) to 76991 ind m<sup>-2</sup> in January 2002 (downstream) (Figure 4.4) and mean densities increased along the gradient of proximity to the sea (Upstream: 9279 ind m<sup>-2</sup>; Intermediate: 10114 ind m<sup>-2</sup>; Downstream: 21601 ind m<sup>-2</sup>).

*Ventrosia ventrosa* was constant upstream ( $C_{\text{Upstream}} = 57.1\%$ ) and accessory at the intermediate and downstream stations ( $C_{\text{Intermediate}} = C_{\text{Downstream}} = 42.9\%$ ). In terms of fidelity, the species was accessory in all stations ( $F_{\text{Upstream}} = 40.0\%$ ;  $F_{\text{Intermediate}} = F_{\text{Downstream}} = 30.0\%$ ). From June to December 2001, the absence of the species was noticed in June (intermediate) and in August (downstream). After that, no specimens were found in the sampling stations. The maximum density was registered in December 2001 at the upstream station (2124 ind m<sup>-2</sup>). Mean densities decreased with station proximity to the sea (Upstream: 607 ind m<sup>-2</sup>; Intermediate: 63 ind m<sup>-2</sup>; Downstream: 38 ind m<sup>-2</sup>).

*Cerastoderma glaucum* was considered as constant *taxon* in the upstream and intermediate stations ( $C_{\text{Upstream}} = C_{\text{Intermediate}} = 57.1\%$ ), being accessory downstream ( $C_{\text{Downstream}} = 42.9\%$ ). The *taxon* was accessory in all stations, presenting the same fidelity values in the upstream and intermediate stations ( $F_{\text{Upstream}} = F_{\text{Intermediate}} = 36.4\%$ ;  $F_{\text{Downstream}} = 27.3\%$ ). During June 2001, August 2001 and July 2002, the species was present in all stations. In January and May 2002, no specimens were found alive in the lagoon, just empty shells. The remaining samples showed an irregular presence of *Cerastoderma glaucum*. The maximum density (6401 ind m<sup>-2</sup>) was determined upstream in August 2001. Mean densities decreased along the gradient of proximity to the sea (Upstream: 1172 ind m<sup>-2</sup>; Intermediate: 240 ind m<sup>-2</sup>; Downstream: 110 ind m<sup>-2</sup>).

*Abra segmentum* was classified as accessory *taxon* in the upstream and downstream stations ( $C_{\text{Upstream}} = C_{\text{Downstream}} = 42.9\%$ ) and as constant *taxon* in the intermediate station ( $C_{\text{Intermediate}} = 71.4\%$ ). Fidelity values defined it as an accessory *taxon* in all stations, although the intermediate station presented a higher value ( $F_{\text{Upstream}} = F_{\text{Downstream}} = 27.3\%$ ;  $F_{\text{Intermediate}} = 45.5\%$ ). During August 2001 and May 2002 the species was present in all stations, registering its maximum at the upstream station in May (1829 ind m<sup>-2</sup>). Mean densities decreased with station proximity to the sea (Upstream: 295 ind m<sup>-2</sup>; Intermediate: 236 ind m<sup>-2</sup>; Downstream: 143 ind m<sup>-2</sup>).

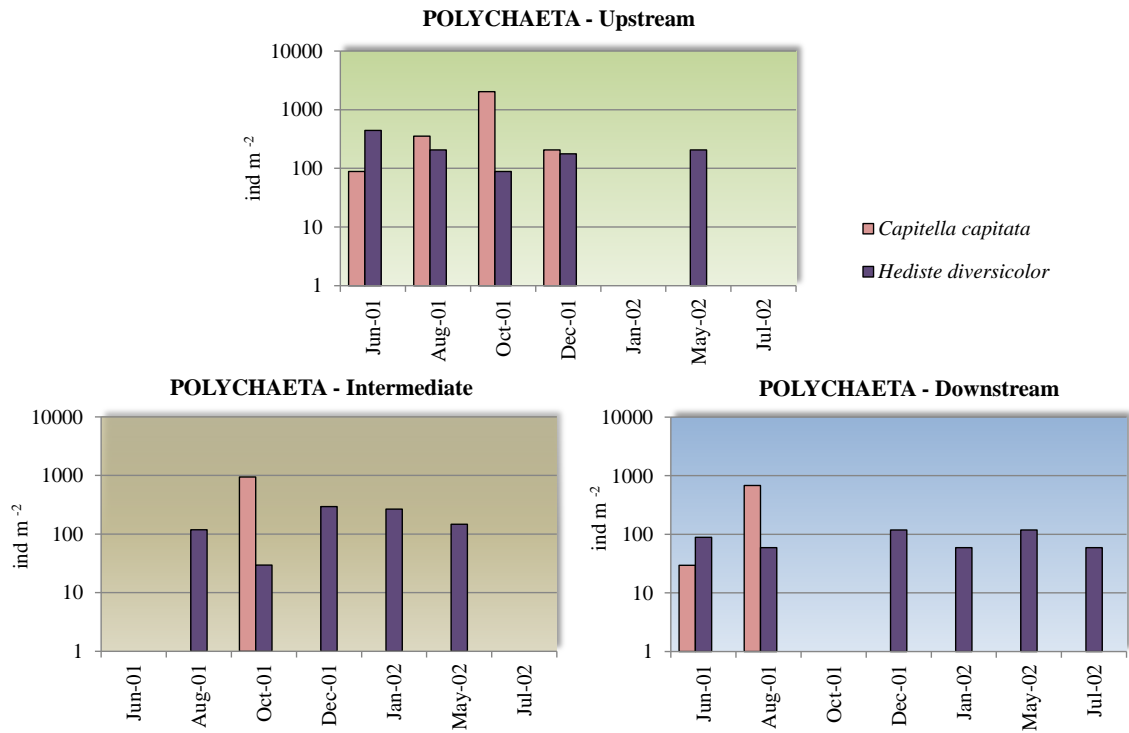


Figure 4.3 - Seasonal variation of Polychaeta *taxa* densities in Foz de Almargem sampling stations.

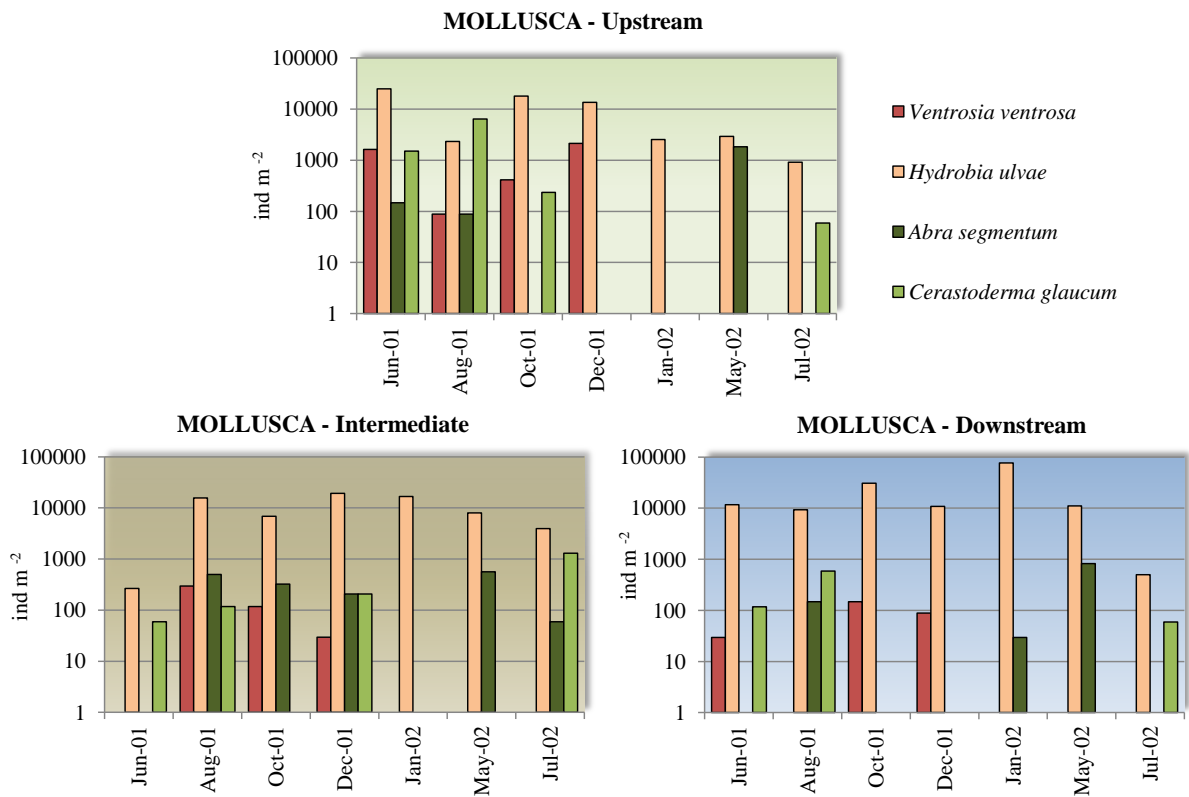


Figure 4.4 - Seasonal variation of Mollusca *taxa* densities (Gastropoda *Ventrosia ventrosa* and *Hydrobia ulvae*; Bivalvia *Abra segmentum* and *Cerastoderma glaucum*) in Foz de Almargem sampling stations.

Most of the benthic macroinvertebrate *taxa* did not present significant correlations between their densities, only *Ventrosia ventrosa* was positively correlated with *Hydrobia ulvae* ( $\rho = 0.493$ ;  $p = 0.023$ ) and with *Capitella capitata* ( $\rho = 0.556$ ;  $p = 0.009$ ).

In terms of constancy, there was a decrease of the percentage of constant *taxa* in the studied stations with the proximity to the sea, as the upstream station presented a greater percentage of constant *taxa* (78%), followed by the intermediate (60%) and the downstream (44%) stations (Figure 4.5). The relative frequency of accessory *taxa* presented the opposite tendency, increasing from the upstream station (22%) to the intermediate station (40%) and to the downstream station (56%).

To what concerns fidelity, most of the *taxa* in the upstream station were accessory (89%) and a smaller percentage were preferential (11%). In the intermediate station, the relative frequency of accessory *taxa* was lower (80%) than upstream, but still represented the majority of the *taxa*. Exclusive *taxa* and accidental (or rare) *taxa*, accounted 10% of the *taxa* found in the intermediate station. Downstream all *taxa* were accessory.

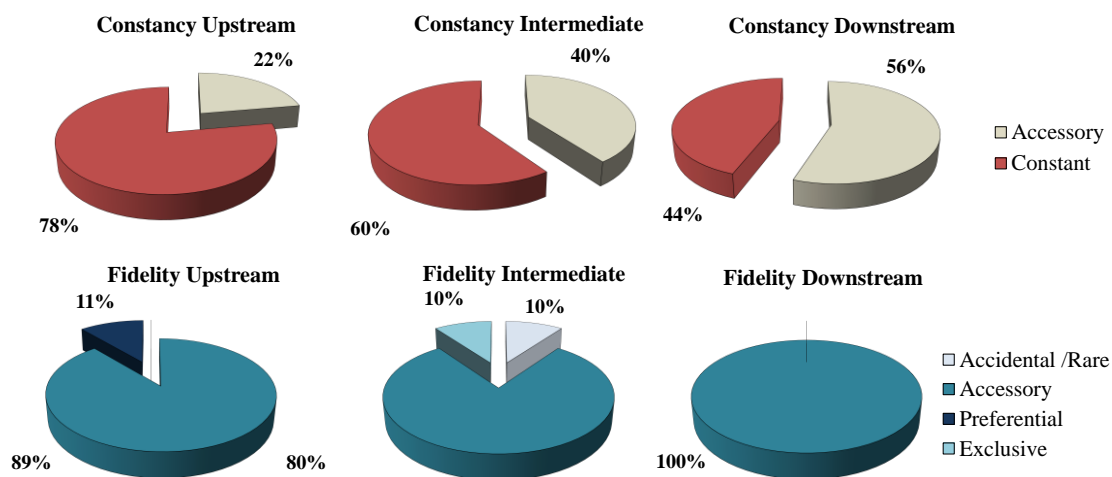


Figure 4.5 – Relative frequency of *taxa* in terms of constancy and fidelity in Foz de Almargem sampling stations.

The upstream station presented higher annual means of taxonomic richness, Shannon-Wiener diversity, evenness, Polychaeta density (*Capitella capitata* and *Hediste*

*diversicolor*), Oligochaeta density, Isopoda density (*Lekanesphaera hookeri*), Gastropoda *Ventrosia ventrosa* density and Bivalvia densities (*Abra segmentum* and *Cerastoderma glaucum*) (Table 4.1).

In the intermediate station, total benthic macroinvertebrate showed the lowest mean density; nevertheless Insecta (*Chironomus* sp.) registered the greatest mean density.

The highest annual means for total benthic macroinvertebrate density and the Gastropoda *Hydrobia ulvae* were determined in the downstream station. This station also presented the lowest means for taxonomic richness, Shannon-Wiener diversity, evenness, Insecta density (*Chironomus* sp.), Polychaeta densities (*Capitella capitata* and *Hediste diversicolor*), Isopoda density (*Lekanesphaera hookeri*), Gastropoda *Ventrosia ventrosa* density and Bivalvia densities (*Abra segmentum* and *Cerastoderma glaucum*).

Table 4.1 – Annual mean values and standard deviation of benthic macroinvertebrate parameters in Foz de Almargem sampling stations.

	<i>Upstream Station</i>	<i>Intermediate Station</i>	<i>Downstream Station</i>
<b>Benthic macroinvertebrate communities</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total benthos density (ind m <sup>-2</sup> )	14737 ± 9514	13359 ± 6652	22891 ± 25732
Taxonomic richness	6.14 ± 2.12	5.29 ± 2.06	5.00 ± 1.41
Shannon-Wiener diversity (bits)	1.33 ± 0.31	1.02 ± 0.60	0.62 ± 0.51
Evenness	0.55 ± 0.18	0.46 ± 0.24	0.27 ± 0.23
<b>INSECTA Diptera density (ind m<sup>-2</sup>)</b>	<b>565 ± 577</b>	<b>952 ± 1392</b>	<b>46 ± 56</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	565 ± 577	952 ± 1392	46 ± 56
<b>POLYCHAETA density (ind m<sup>-2</sup>)</b>	<b>556 ± 745</b>	<b>257 ± 336</b>	<b>173 ± 253</b>
<i>Capitella capitata</i> density (ind m <sup>-2</sup> )	383 ± 740	135 ± 357	101 ± 255
<i>Hediste diversicolor</i> density (ind m <sup>-2</sup> )	160 ± 153	122 ± 122	72 ± 41
<b>OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	<b>13 ± 33</b>	<b>8 ± 22</b>	<b>8 ± 22</b>
<b>ISOPODA density (ind m<sup>-2</sup>)</b>	<b>2250 ± 2855</b>	<b>1488 ± 2037</b>	<b>771 ± 976</b>
<i>Lekanesphaera hookeri</i> density (ind m <sup>-2</sup> )	2250 ± 2855	1488 ± 2037	771 ± 976
<b>GASTROPODA density (inds m<sup>-2</sup>)</b>	<b>9888 ± 10154</b>	<b>10177 ± 7231</b>	<b>21639 ± 26057</b>
<i>Ventrosia ventrosa</i> density (inds m <sup>-2</sup> )	607 ± 889	63 ± 111	38 ± 58
<i>Hydrobia ulvae</i> density (inds m <sup>-2</sup> )	9279 ± 9481	10114 ± 7194	21601 ± 26056
<b>BIVALVIA density (inds m<sup>-2</sup>)</b>	<b>1466 ± 2352</b>	<b>476 ± 453</b>	<b>253 ± 364</b>
<i>Abra segmentum</i> density (inds m <sup>-2</sup> )	295 ± 679	236 ± 234	143 ± 306
<i>Cerastoderma glaucum</i> density (inds m <sup>-2</sup> )	1172 ± 2370	240 ± 473	110 ± 217

Statistical tests applied to compare benthic macroinvertebrate parameters among stations determined that Shannon-Wiener diversity, evenness and Insecta densities presented significant differences in the mean values (Appendix I.J), specifically

between the upstream and downstream stations in the case of Shannon-Wiener diversity and evenness (upstream > downstream); and between Insecta density from the intermediate and downstream stations (intermediate > downstream).

All stations presented *taxa* from the five trophic groups considered (Figure 4.6) and during the studied period, deposit feeders were the most abundant macroinvertebrates with a few exceptions, particularly in the upstream station. The intermediate and the downstream stations were more similar in terms of trophic groups' dominance along time, being majorly composed by deposit feeders from June 2001 to May 2002; just in July 2002, herbivorous densities were greater than deposit feeders, nevertheless deposit feeders represented the second group with higher densities.

In the upstream station, deposit feeders densities ranged from 2743 ind m<sup>-2</sup> (86%) in July 2002 to 26637 ind m<sup>-2</sup> (93%) in June 2001, presenting higher densities than the other trophic groups also in October and December 2001. In August 2001, there was decrease in deposit feeders' density (3215 ind m<sup>-2</sup>; 31%) and the community was dominated by suspension feeders (6401 ind m<sup>-2</sup>; 63%). Suspension feeders were identified from June to October 2001 and in July 2002. During January 2002, when the lagoon was in connection with the sea, deposit feeders density diminished (2920 ind m<sup>-2</sup>; 52%) and there was an increment in herbivores density (2714 ind m<sup>-2</sup>; 42%). In May 2002, herbivores were the most abundant trophic group (7876 ind m<sup>-2</sup>; 59%), followed by deposit feeders (3451 ind m<sup>-2</sup>; 26%), suspension/deposit feeders (1829 ind m<sup>-2</sup>; 14%) and carnivorous/ scavengers/ omnivorous (206 ind m<sup>-2</sup>; 1%). Herbivores were absent from the upstream station in June 2001 and during the remaining studied months, herbivores densities went from 324 ind m<sup>-2</sup> (3%) in August 2001 to 7876 ind m<sup>-2</sup> (59%) in May 2002. Suspension/ deposit feeders only occurred in June and August 2001, and in May 2002 the group presented its higher density. Carnivorous/ scavengers/ omnivorous were observed from June to December 2001 and then, only in May 2002. During the period of occurrence, densities varied from 88 ind m<sup>-2</sup> (< 1%) in October 2001 to 442 ind m<sup>-2</sup> (2%) in June 2001.

In the intermediate station, the minimum (265 ind m<sup>-2</sup>; 82%) and maximum (19410 ind m<sup>-2</sup>; 92%) densities of deposit feeders were lower than upstream and occurred in different months, June and December 2001 respectively. Herbivores were observed in the same months as in the upstream station and the lowest density (29 ind m<sup>-2</sup>; < 1%) was also observed in August 2001. In July 2002, herbivores reached

the maximum density of 2360 ind m<sup>-2</sup>, accounting a greater percentage (47%) than deposit feeders. Suspension feeders also presented the maximum density in July 2002 (1298 ind m<sup>-2</sup>; 11%), but it was lower than the maximum density registered upstream in August 2001. This group was observed in the intermediate station during the studied months, except in October 2001, January and May 2002. Suspension/ deposit feeders were absent in June 2001 and January 2002, showing the greatest densities in August 2001 (501 ind m<sup>-2</sup>; 3%) and May 2002 (560 ind m<sup>-2</sup>; 4%). Carnivorous/ scavengers/ omnivorous occurred from August 2001 to May 2002, with a minimum density of 29 ind m<sup>-2</sup> (< 1%) in October and a maximum density in December 2001 (295 ind m<sup>-2</sup>; 1%).

Downstream, the maximum density of deposit feeders was determined in January 2002 (76991 ind m<sup>-2</sup>; 99%), being the highest density of all sampling stations. The minimum density was registered in July 2002 (501 ind m<sup>-2</sup>; 35%), just as in the upstream station. Herbivores occurred in the same months as in the other sampling stations and followed a similar evolution as the upstream station, with the minimum and maximum densities registered in the same months, August 2001 (147 ind m<sup>-2</sup>; 1%) and May 2002 (2891 ind m<sup>-2</sup>; 19%). Nevertheless, it was in July 2002 that herbivores accounted the greatest percentage of macroinvertebrates downstream (826 ind m<sup>-2</sup>; 57%). Suspension feeder's occurrence was less regular than in the intermediate station and August 2001 was the month with maximum density (590 ind m<sup>-2</sup>; 5%) as it happened upstream, although density was lower downstream. Suspension/ deposit feeders presented the maximum density in May 2002 (826 ind m<sup>-2</sup>; 6%), as in the upstream and intermediate stations. Carnivorous/ scavengers/ omnivorous were absent from the downstream station in August 2001 and the maximum density (118 ind m<sup>-2</sup>) that occurred in December 2001 (1%) and May 2002 (< 1%) was lower than the maximum densities determined in the two other stations.

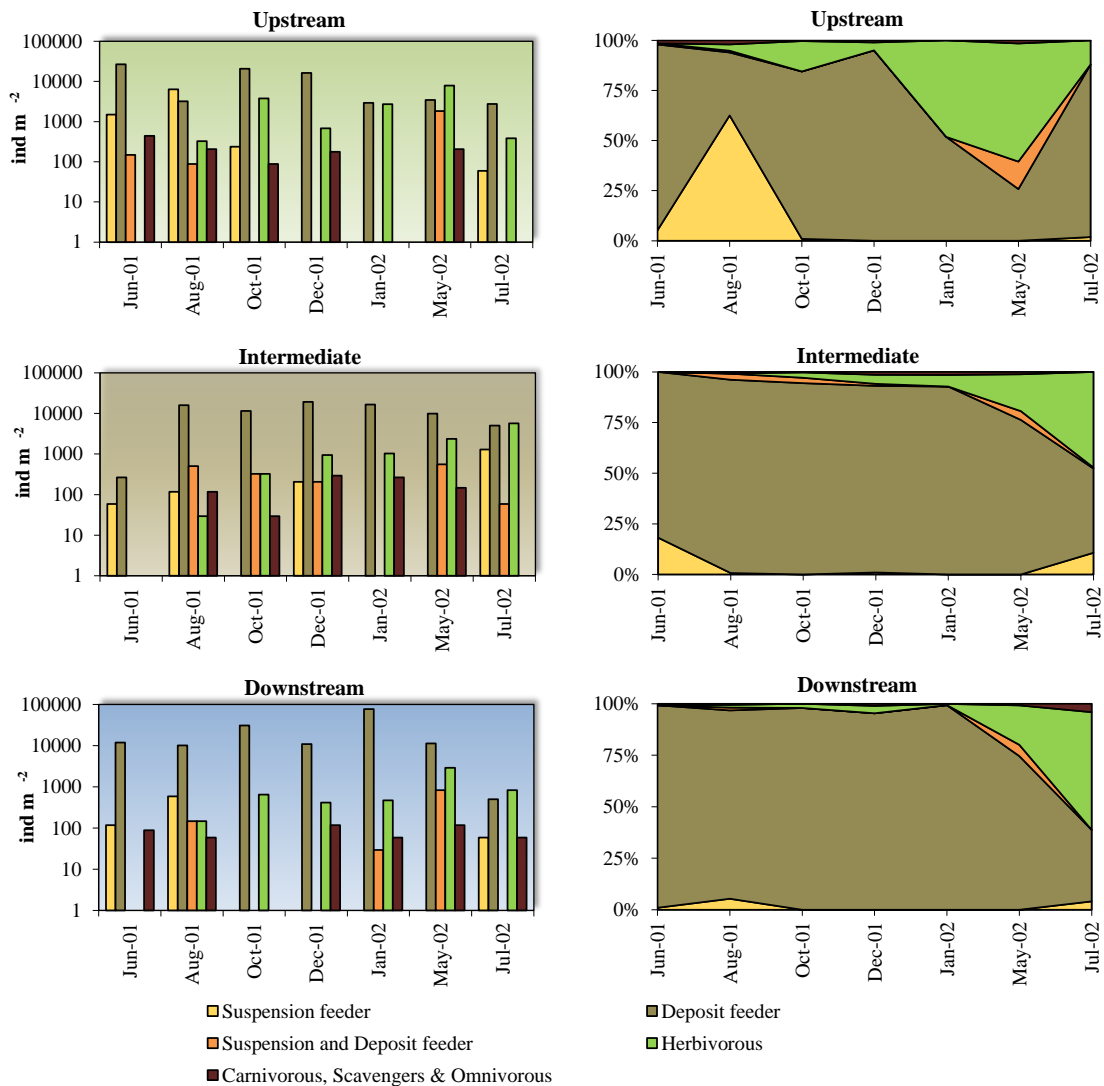


Figure 4.6 - Seasonal variation of trophic groups densities and relative frequency of each group in Foz de Almargem sampling stations.

In terms of ecological groups (according to their sensitivity to an increasing stress gradient), most *taxa* found in the three stations belong to group III - AMBI, which include *taxa* tolerant to excess organic matter enrichment (Figure 4.7). Group III ranged from 92% (October 2001) to 100% of monthly macroinvertebrate densities in the upstream and intermediate stations, with annual means of 98% and 99% respectively. Downstream, group III *taxa* comprised 93% (August 2001) to 100% and the annual mean was 99%.

Polychaeta and Oligochaeta were the only first-order opportunistic *taxa*, indicators of pronounced unbalanced situations, (Group V - AMBI) identified in the sampling stations. Their presence during the studied period was more regular in the upstream

station, being absent just in January and July 2002. During these months, no group V *taxa* were found in the other stations too.

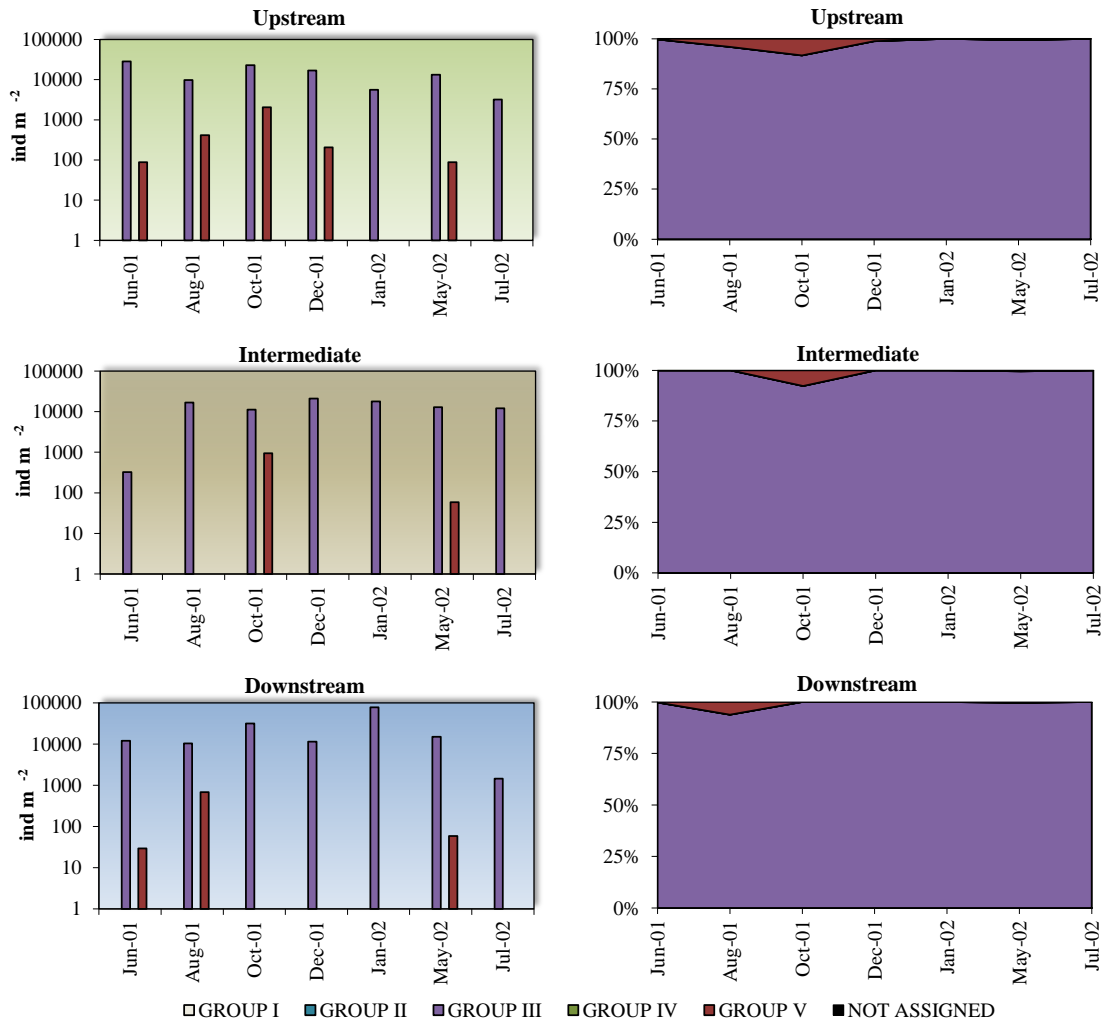


Figure 4.7 - Seasonal variation of ecological AMBI groups densities and relative frequency of each group in Foz de Almagem sampling stations.

#### 4.3.1.2. Environmental parameters and benthic macroinvertebrate communities

The analysis of bivariate linear associations between benthic macroinvertebrates and environmental parameters determined that the total density of benthic macroinvertebrates was negatively correlated with pH (Table 4.2). For taxonomic richness no significant correlation was found ( $p > 0.05$ ), but Shannon-Wiener diversity and evenness showed positive associations with chlorophyll *a* and phaeo-pigments concentrations in the sediment, just as chlorophyll *a* concentration in water. Besides

these environmental parameters, Shannon-Wiener diversity presented a negative correlation with dissolved oxygen concentration and positive correlations with phaeo-pigments concentration and pigment diversity in the water. Evenness was also positively correlated with the water level in the lagoon and negatively correlated to salinity and total solids in suspension.

Insecta density (*Chironomus* sp.) showed a negative correlation with dissolved oxygen concentration and positive correlations with chlorophyll *a* and phaeo-pigments concentrations in the sediment, and phaeo-pigments concentration in water.

For the two species of Polychaeta *Capitella capitata* and *Hediste diversicolor* were determined linear associations with different environmental parameters. *Capitella capitata* was positively correlated with total solids in suspension, salinity and negatively correlated with the water level in the lagoon and N:P ratio. *Hediste diversicolor* presented a positive correlation with clay content and negative correlations with total phosphorus and chlorophyll *a* concentration in water.

The Isopoda *Lekanesphaera hookeri* was the *taxa* correlated with a greater number of environmental parameters. Its density was negatively correlated with salinity and total solids in suspension, whereas positive correlations were determined with the water level in the lagoon, nitrogen compounds (nitrites, nitrates and total dissolved inorganic nitrogen), N:P ratio, chlorophyll *a* and phaeo-pigments concentrations in the sediment.

The Gastropoda *Ventrosia ventrosa* showed a negative association with the water level in the lagoon and positive associations with total solids in suspension and salinity.

For the Bivalvia *Abra segmentum* was found a positive correlation with water content in the sediment, while for *Cerastoderma glaucum* several correlations were determined namely, positive correlations with temperature, ammonia concentration and negative correlations with cumulative rainfall, nitrites, nitrates and N:P ratio.

The strongest and most significant linear associations were determined between: 1) *Ventrosia ventrosa* density vs. total solids in suspension; 2) *Cerastoderma glaucum* density vs. temperature and nitrates concentration; 3) *Lekanesphaera hookeri* density vs. nitrates concentration, salinity and water level in the lagoon; 4) Shannon-Wiener diversity, evenness vs. chlorophyll *a* concentration in the sediment; 5) *Capitella capitata* density vs. total solids in suspension.

Table 4.2 - Significant correlations between benthic macroinvertebrate and environmental parameters (water and sediment) from Foz de Almargem lagoon. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Benthos parameters</i>	<i>Environmental parameters</i>	<i>Results</i>
<b>Total Benthos density</b>	pH	Rho = -0.543; $p = 0.011$ *
<b>Shannon-Wiener Diversity Index (H')</b>	Chlorophyll <i>a</i> concentration in sediment (ChlaS)	R = 0.674; $p = 0.001$ **
	Phaeo-pigments concentration in sediment (PhaeS)	R = 0.515; $p = 0.017$ *
	Dissolved Oxygen (DO)	R = -0.484; $p = 0.026$ *
	Chlorophyll <i>a</i> concentration in water (ChlaW)	Rho = 0.453; $p = 0.039$ *
	Phaeo-pigments concentration in water(PhaeW)	Rho = 0.439; $p = 0.047$ *
	Pigment diversity index in water (PigDW)	Rho = 0.440; $p = 0.046$ *
<b>Evenness (J)</b>	Water level in the lagoon (WLevel)	Rho = 0.505; $p = 0.020$ *
	Chlorophyll <i>a</i> concentration in sediment (ChlaS)	R = 0.622; $p = 0.003$ **
	Phaeo-pigments concentration in sediment (PhaeS)	R = 0.457; $p = 0.037$ *
	Salinity (Sali)	R = -0.468; $p = 0.032$ *
	Chlorophyll <i>a</i> concentration in water (ChlaW)	Rho = 0.472; $p = 0.031$ *
	Total solids in suspension (TSS)	R = -0.495; $p = 0.022$ *
<b>Chironomus sp. density</b>	Chlorophyll <i>a</i> concentration in sediment (ChlaS)	Rho = 0.505; $p = 0.019$ *
	Phaeo-pigments concentration in sediment (PhaeS)	Rho = 0.476; $p = 0.029$ *
	Dissolved Oxygen (DO)	Rho = -0.436; $p = 0.048$ *
	Phaeo-pigments concentration in water(PhaeW)	Rho = 0.525; $p = 0.015$ *
<b>Capitella capitata density</b>	Water level in the lagoon (WLevel)	Rho = -0.479; $p = 0.028$ *
	Salinity (Sali)	Rho = 0.514; $p = 0.017$ *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	Rho = -0.465; $p = 0.034$ *
	Total solids in suspension (TSS)	Rho = 0.563; $p = 0.008$ **
<b>Hediste diversicolor density</b>	Clay content (% Clay)	Rho = 0.445; $p = 0.043$ *
	Chlorophyll <i>a</i> concentration in water (ChlaW)	Rho = -0.435; $p = 0.049$ *
	Total phosphorus concentration (TP)	Rho = -0.532; $p = 0.013$ *
<b>Lekanesphaera hookeri density</b>	Water level in the lagoon (WLevel)	Rho = 0.588; $p = 0.005$ **
	Chlorophyll <i>a</i> concentration in sediment (ChlaS)	Rho = 0.614; $p = 0.003$ **
	Phaeo-pigments concentration in sediment (PhaeS)	Rho = 0.439; $p = 0.046$ *
	Salinity (Sali)	Rho = -0.603; $p = 0.004$ **
	Nitrates concentration (NO <sub>3</sub> )	Rho = 0.693; $p = 0.000$ **
	Nitrites concentration (NO <sub>2</sub> )	Rho = 0.450; $p = 0.041$ *
	Total dissolved inorganic nitrogen concentration (DIN)	Rho = 0.436; $p = 0.048$ *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	Rho = 0.442; $p = 0.045$ *
Total solids in suspension (TSS)	Rho = -0.460; $p = 0.036$ *	
<b>Ventrosia ventrosa density</b>	Water level in the lagoon (WLevel)	Rho = -0.492; $p = 0.024$ *
	Salinity (Sali)	Rho = 0.437; $p = 0.048$ *
	Total solids in suspension (TSS)	Rho = 0.701; $p = 0.000$ **
<b>Abra segmentum density</b>	Water content in sediment (%WSed)	Rho = 0.527; $p = 0.014$ *
<b>Cerastoderma glaucum density</b>	Cumulative rainfall (Rain)	Rho = -0.441; $p = 0.046$ *
	Temperature (Temp)	Rho = 0.684; $p = 0.000$ **
	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.665; $p = 0.001$ **
	Nitrites concentration (NO <sub>2</sub> )	Rho = -0.502; $p = 0.020$ *
	Ammonia concentration (NH <sub>4</sub> )	Rho = 0.497; $p = 0.022$ *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	Rho = -0.498; $p = 0.022$ *

The Canonical Correspondence Analysis (CCA) performed with four of the environmental parameters that contributed more to the two first axes in the PCA of water and sediment parameters (Figure 2.20) determined a significant relation between the densities of benthic macroinvertebrate *taxa* and these environmental parameters (Monte Carlo test:  $p = 0.008$ ). The environmental parameters considered were water level in the lagoon, salinity, total solids in suspension, water content in the sediment and chlorophyll *a* concentration in the sediment (Figure 4.8).

The densities of the Insecta *Chironomus* sp., the Isopoda *Lekanesphaera hookeri*, the Bivalvia *Abra segmentum* and Oligochaeta were positively associated with the water level in the lagoon, the concentration of chlorophyll *a* in the sediment and the percentage of water in the sediment (right side of first axis). The two last parameters presented higher values in sediments with greater content of silt and clay, which correspond to most of the upstream and intermediate samples. These *taxa* also occurred more abundantly during months with lower salinity and total solids in suspension.

On the left side of axis I were plotted the Polychaeta *Capitella capitata*, the Bivalvia *Cerastoderma glaucum* and the Gastropoda *Hydrobia ulvae* and *Ventrosia ventrosa*. The higher densities of *Capitella capitata*, *Cerastoderma glaucum* and *Ventrosia ventrosa* were mainly related with higher values of salinity and total solids in suspension, but also with periods of lower water level in the lagoon.

*Hydrobia ulvae* and *Ventrosia ventrosa* densities showed a negative relation with chlorophyll *a* concentration and water content in the sediment, occurring with higher densities mainly in downstream samples.

*Hediste diversicolor* was displayed in the centre of the two axes, meaning that it was present in most of the samples and that the *taxon* was not associated with the environmental parameters selected.

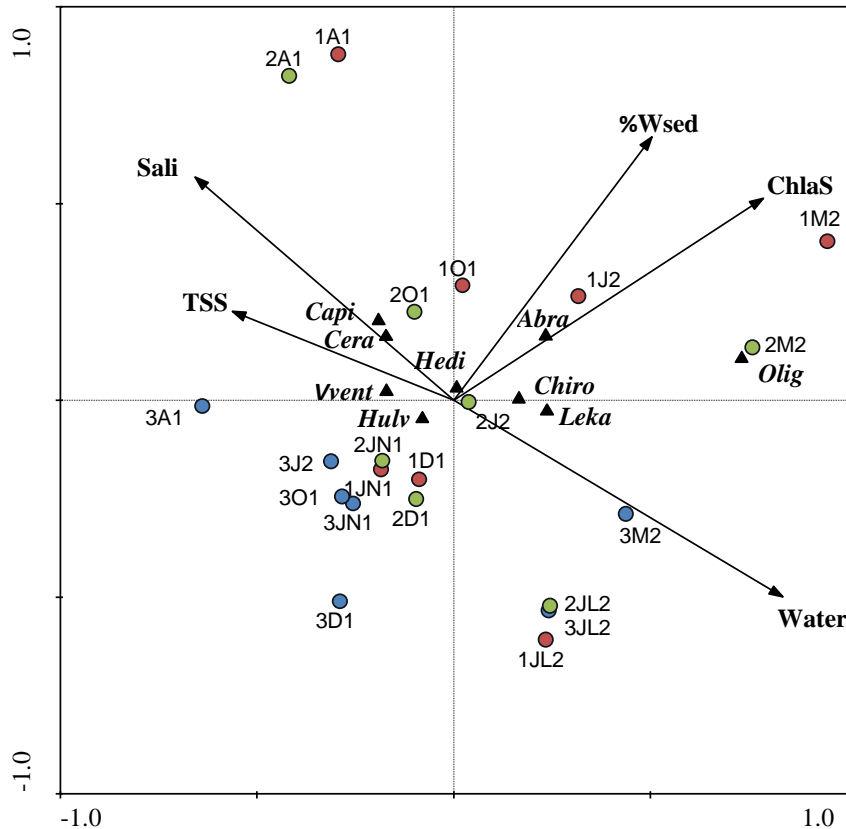


Figure 4.8 - Canonical correspondence analysis performed with the benthic macroinvertebrate *taxa* (total density *per station*) from Foz de Almargem lagoon. Cumulative percentage variance explained by axes: Species - I = 24.2% and I + II = 29.9%; and species-environment relation - I = 63.1% and I + II = 78.1%. Monte Carlo test of all canonical axes:  $p = 0.008$ .

*Station codes*: First character corresponds to the sampling station (1 - upstream, 2 - intermediate, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002).

*Environmental parameters*: Water- water level in the lagoon; Sali – Salinity; TSS – Total solids in suspension; %Wsed – Water content in the sediment; ChlaS - Chlorophyll *a* concentration in the sediment. *Benthic macroinvertebrate taxa*: Olig – Oligochaeta; Capi – Capitellidae; Chiro – *Chironomus* sp.; Abra – *Abra segmentum*; Leka – *Lekanesphaera hookeri*; Hedi – *Hediste diversicolor*; Hulv – *Hydrobia ulvae*; Vvent – *Ventrosia ventrosa*; Cera – *Cerastoderma glaucum*.

#### 4.3.1.3. Comparison of benthic macroinvertebrate communities during isolation and connection of the lagoon with the sea

The comparison of benthic communities before and during the lagoon connection with the sea was done based on data from December 2001 and January 2002. Before December, there was a first opening of the lagoon in the end of October and between December and January samplings, the lagoon opened for a second time.

All stations presented a decrease in taxonomic richness, total Insecta densities (*Chironomus* sp.), total Polychaeta densities and *Hediste diversicolor*, total Gastropoda

densities and *Ventrosia ventrosa*, registering an increase in total Isopoda densities (*Lekanesphaera hookeri*) (Table 4.3). Total benthos density suffered a reduction upstream and in the intermediate station, while downstream was observed an increment and densities in January followed the gradient of distance from the sea, being smaller upstream and greater downstream. The variation observed in total benthos was mainly caused by the decrease of Gastropoda (*Ventrosia ventrosa* and *Hydrobia ulvae*) upstream and in the intermediate station and the increase of *Hydrobia ulvae* downstream. Thereby, total Gastropoda and *Hydrobia ulvae* densities augmented with the proximity to the sea, but no individuals of *Ventrosia ventrosa* were found in the three stations. The variation and densities of the Isopoda *Lekanesphaera hookeri* in January followed the opposite tendency than total Gastropoda and *Hydrobia ulvae* densities, decreasing from upstream to downstream. In January, only three species were found in the upstream station, *Lekanesphaera hookeri* (48.17% of total benthos in this station), *Hydrobia ulvae* (45.03%) and *Chironomus* sp. (6.80%). The Gastropoda *Ventrosia ventrosa* and Polychaeta species as *Capitella capitata* and *Hediste diversicolor* were absent from this station in January. When the lagoon was in connection with the sea, the intermediate station benthic community was also composed just by three species, *Hydrobia ulvae* (92.78% of total benthos in this station), *Lekanesphaera hookeri* (5.74%) and *Hediste diversicolor* (1.48%). Four of the taxa observed in December (*Chironomus* sp., *Ventrosia ventrosa*, *Abra segmentum* e *Cerastoderma glaucum*), were not found in January. At the downstream station, four species were quantified in January, *Hydrobia ulvae* (99.28%), *Lekanesphaera hookeri* (0.61%), *Hediste diversicolor* (0.08%) and *Abra segmentum* (0.04%). Comparative to December, two taxa became absent (*Chironomus* sp. and *Ventrosia ventrosa*) and a new one registered (*Abra segmentum*). Shannon-Wiener diversity and evenness increased upstream and decreased downstream, presenting a reduction in their values along the gradient of proximity to the sea.

When the lagoon was opened to the sea, there was an increase of the number of trophic groups along the gradient of proximity with the sea. The upstream station just presented deposit feeders (52%) and herbivores (42%), with deposit feeders showing a density decrease and herbivores had an increase.

In the intermediate station, besides deposit feeders (93%) and herbivores (6%), there were also carnivorous/ scavengers/ omnivorous (2%). Herbivores increased their density, while deposit feeders and carnivorous/scavengers/omnivorous diminished.

Downstream station had the same three groups described the intermediate station (deposit feeders: 99%; herbivores: <1%; carnivorous/ scavengers/ omnivorous: < 1%) plus suspension/ deposit feeders (< 1%). An augment of densities was observed in deposit feeders, in herbivores, in suspension/ deposit feeders and there was a reduction in carnivorous/ scavengers/ omnivorous density.

In terms of AMBI ecological groups, only *taxa* tolerant to excess organic matter enrichment (Group III) were observed in January 2002. Both upstream and intermediate stations registered a decrease in group III densities, while downstream there was an increase which corresponded to the major variation of all stations. Densities augmented along the gradient of proximity with the sea.

With the opening of the lagoon, the upstream station also showed a diminishment in the densities of first-order opportunistic *taxa* associated to pronounced unbalanced situations (Group V).

Table 4.3 – Benthic macroinvertebrate communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Foz de Almargem sampling stations.

	<i>Upstream station</i>		<i>Intermediate station</i>		<i>Downstream station</i>	
	Jan-02	Variation	Jan-02	Variation	Jan-02	Variation
<b>Macroinvertebrate benthic communities</b>						
Total benthos density (ind m <sup>-2</sup> )	5634	<b>-11474</b>	17964	<b>-3097</b>	77551	<b>66077</b>
Taxonomic richness	3	<b>-3</b>	3	<b>-4</b>	4	<b>-1</b>
Shannon-Wiener diversity (bits)	1.29	<b>0.19</b>	0.43	<b>-0.17</b>	0.07	<b>-0.35</b>
Evenness	0.81	<b>0.39</b>	0.27	<b>0.07</b>	0.03	<b>-0.15</b>
<b>Total INSECTA density (ind m<sup>-2</sup>)</b>	383	<b>-29</b>	0	<b>-29</b>	0	<b>-59</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	383	<b>-29</b>	0	<b>-29</b>	0	<b>-59</b>
<b>Total POLYCHAETA density (ind m<sup>-2</sup>)</b>	0	<b>-383</b>	265	<b>-29</b>	59	<b>-59</b>
<i>Capitella capitata</i> density (ind m <sup>-2</sup> )	0	<b>-206</b>	0	<b>0</b>	0	<b>0</b>
<i>Hediste diversicolor</i> density (ind m <sup>-2</sup> )	0	<b>-117</b>	265	<b>-29</b>	59	<b>-59</b>
<b>Total OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	0	<b>0</b>	0	<b>0</b>	0	<b>0</b>
<b>Total ISOPODA density (ind m<sup>-2</sup>)</b>	2714	<b>2035</b>	1032	<b>88</b>	472	<b>59</b>
<i>Lekanesphaera hookeri</i> density (ind m <sup>-2</sup> )	2714	<b>2035</b>	1032	<b>88</b>	472	<b>59</b>
<b>Total GASTROPODA density (ind m<sup>-2</sup>)</b>	2537	<b>-13097</b>	16667	<b>-2713</b>	76991	<b>66107</b>
<i>Ventrosia ventrosa</i> density (ind m <sup>-2</sup> )	0	<b>-2124</b>	0	<b>-29</b>	0	<b>-88</b>
<i>Hydrobia ulvae</i> density (ind m <sup>-2</sup> )	2537	<b>-10973</b>	16667	<b>-2684</b>	76991	<b>66195</b>
<b>Total BIVALVIA density (ind m<sup>-2</sup>)</b>	0	<b>0</b>	0	<b>-414</b>	29	<b>29</b>
<i>Abra segmentum</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-211</b>	29	<b>29</b>
<i>Cerastoderma glaucum</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-203</b>	0	<b>0</b>
<b>TROPHIC GROUPS DENSITIES</b>						
Suspension feeders (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-206</b>	0	<b>0</b>
Deposit feeders (ind m <sup>-2</sup> )	2920	<b>-13332</b>	16667	<b>-2742</b>	76991	<b>66048</b>
Suspension/ deposit feeders (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-211</b>	29	<b>29</b>
Herbivores (ind m <sup>-2</sup> )	2714	<b>2035</b>	1032	<b>88</b>	472	<b>59</b>
Carnivorous/scavengers/omnivorous (ind m <sup>-2</sup> )	0	<b>-177</b>	265	<b>-29</b>	59	<b>-59</b>
<b>AMBI GROUPS DENSITIES</b>						
Group III (ind m <sup>-2</sup> )	5634	<b>-11268</b>	17964	<b>-3097</b>	77551	<b>66077</b>
Group V (ind m <sup>-2</sup> )	0	<b>-206</b>	0	<b>0</b>	0	<b>0</b>

### 4.3.2. Salgados coastal lagoon

#### 4.3.2.1. Benthic macroinvertebrate communities

In Salgados lagoon were identified 16 *taxa* (Appendix II.D), most of them Insecta (12) and the remaining *taxa* belonged to Oligochaeta (2), Crustacea Isopoda (1) and Crustacea Amphipoda (1).

During the studied period, the Insecta Diptera *Chironomus* sp. was the only *taxa* present in all samplings, representing 58% of the total benthic macroinvertebrates identified in the lagoon. Five *taxa* occurred at least in 25% of the samplings (Insecta Ephidridae: 44%; Crustacea Isopoda *Lekanesphaera hookeri*: 44%; Insecta Tabanidae: 31%; Oligochaeta Tubificidae: 31%; Insecta Ceratopogonidae: 25%). *Lekanesphaera hookeri* and Oligochaeta Tubificidae accounted 18% and 14% of the total macroinvertebrates identified in Salgados lagoon.

Taxonomic richness in the upstream station varied from two (August 2001) to nine (March 2002), while downstream it ranged from three (June 2001, December 2001 and January 2002) to six *taxa* (August 2001) (Figure 4.9). The evolution of the number of *taxa* was distinct in the studied stations, just in December 2001 and in March 2002 the two stations showed a similar tendency.

The lowest and the highest densities of benthic macroinvertebrate were observed downstream in January 2002 (88 ind m<sup>-2</sup>) and July 2002 (84926 ind m<sup>-2</sup>), whereas upstream densities varied between 398 ind m<sup>-2</sup> (October 2001) and 52802 ind m<sup>-2</sup> (July 2002). The two stations presented similar patterns of evolution, although the range was different. In January 2002, when the lagoon was connected to the sea, there was a marked decrease upstream and downstream, followed by an increase in the densities of the two stations till July 2002.

Total benthic macroinvertebrate density was positively and highly correlated to total Insecta density ( $r = 0.937$ ;  $p < 0.001$ ), Insecta Diptera density ( $r = 0.933$ ;  $p < 0.001$ ) and *Chironomus* sp. density ( $r = 0.932$ ;  $p < 0.001$ ).

The minimum and maximum values of Shannon-Wiener diversity index were determined upstream, in May (0.05 bits) and March 2002 (2.56 bits). Downstream, values ranged from 0.66 bits (June 2001) to 1.82 bits (July 2002). Diversity was higher downstream, except in October 2001, January and March 2002.

Shannon-Wiener diversity was negatively correlated with *Chironomus* sp. density ( $r = -0.520$ ;  $p = 0.039$ ).

The lowest evenness was calculated upstream in May 2002 (0.03) and the highest was found downstream in January 2002 (1.00). Along time, the downstream station presented greater values of evenness, with the exception of October 2001 and March 2002.

Evenness was negatively correlated with total Insecta density ( $r = -0.629$ ;  $p = 0.009$ ), Insecta Diptera density ( $r = -0.627$ ;  $p = 0.009$ ) and *Chironomus* sp. density ( $r = -0.667$ ;  $p = 0.005$ ).

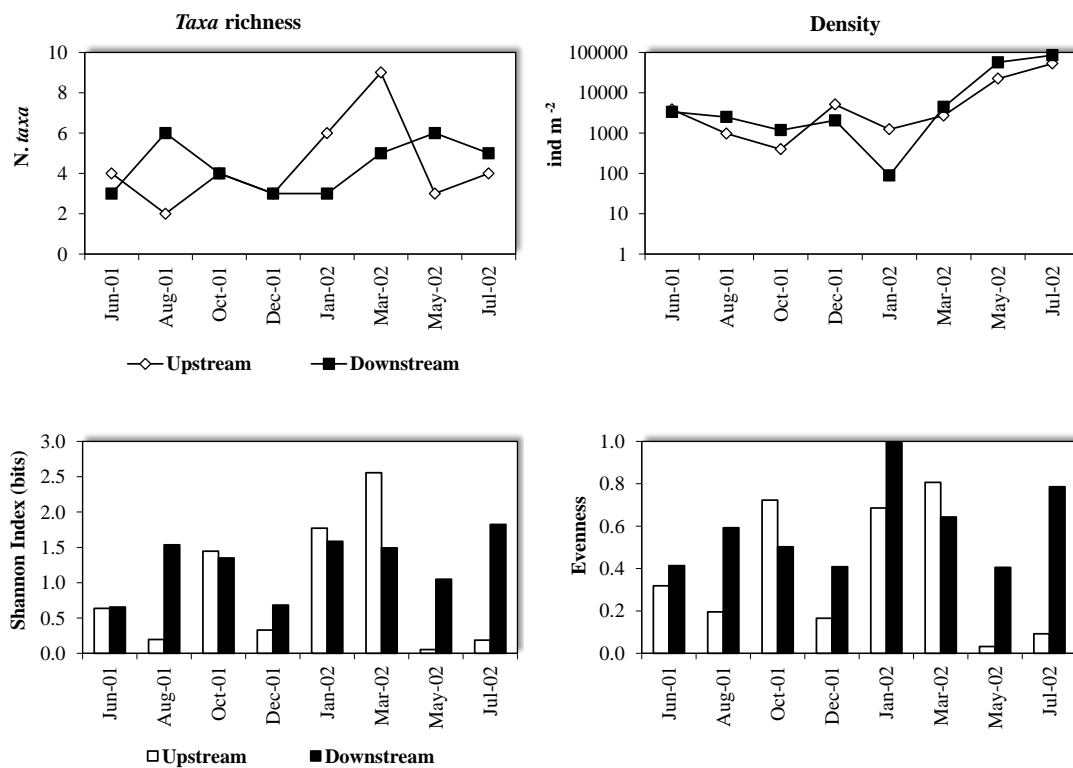


Figure 4.9 - Evolution of total benthos density, taxa richness, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) in Salgados sampling stations.

Benthic macroinvertebrate communities in the two stations were quite distinct (Figure 4.10).

The upstream station community was mainly composed by Insecta, which was the only taxonomic group found from June to December 2001 and in May 2002. Insecta densities ranged from 398 ind m<sup>-2</sup> (100%) in October 2001 to 52448 ind m<sup>-2</sup> (99%) in July 2002. Oligochaeta taxa were observed in January (324 ind m<sup>-2</sup>; 26%) and March

2002 (118 ind m<sup>-2</sup>; 4%); the presence of Crustacea taxa was registered just in July 2002 (354 ind m<sup>-2</sup>; 1%).

In the downstream community, besides Insecta, the presence of Oligochaeta and Crustacea taxa was more regular, particularly from January until July 2002. When the lagoon was opened to the sea (January 2002), densities of these groups were the lowest determined during the studied period and each group had approximately the same density value.

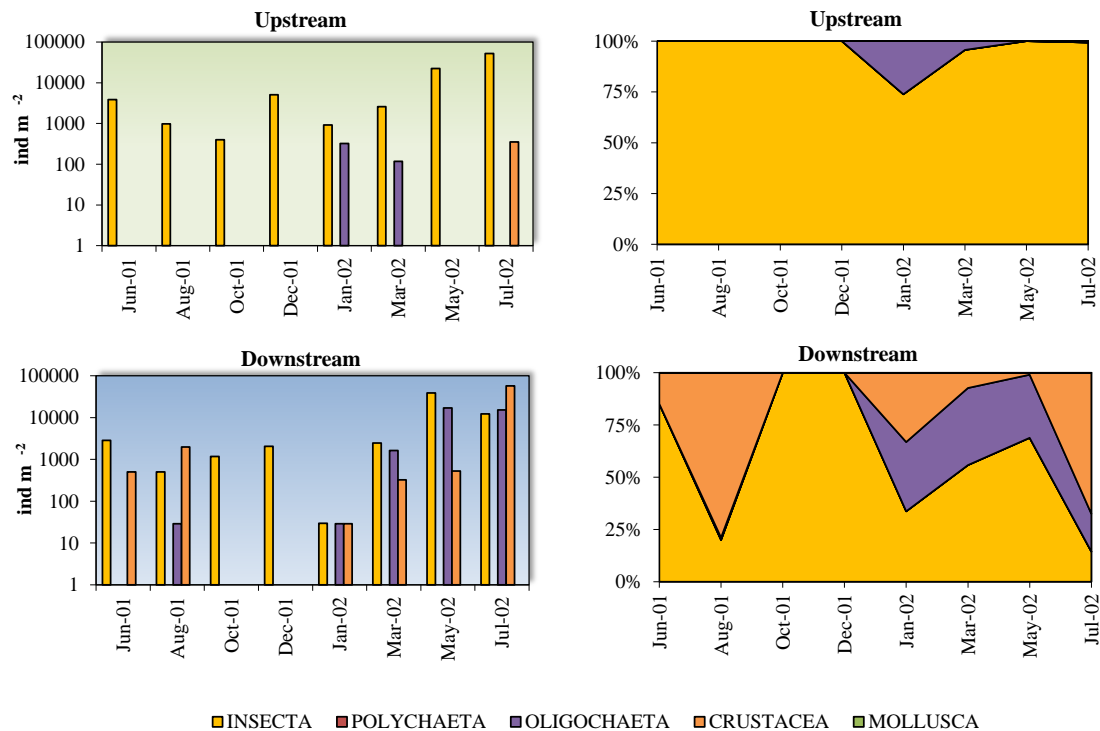


Figure 4.10 - Seasonal variation of the main taxonomic benthos groups densities and relative frequency of each group in Salgados sampling stations.

The range of Insecta densities was smaller than in the upstream station, varying from 29 ind m<sup>-2</sup> (34%) in January 2002 to 39086 ind m<sup>-2</sup> (69%) in May 2002. During June, October and December 2001, Insecta was the dominant taxonomic group, accounting 85% of macroinvertebrate densities in June (2861 ind m<sup>-2</sup>) and 100% in October (1180 ind m<sup>-2</sup>) and December 2001 (2065 ind m<sup>-2</sup>).

Oligochaeta absence was registered in June, October and December 2001. The highest density occurred in May 2002 (17139 ind m<sup>-2</sup>; 30%), but from January till May, Oligochaeta represented around 1/3 of the total densities in these months.

Crustacea were present during most of the studied period, except in October and December 2001. July 2002 was the month with greater density of Crustacea

(57316 ind m<sup>-2</sup>), comprising 67% of total macroinvertebrates density. Although Crustacea density in August 2001 (1976 ind m<sup>-2</sup>) was lower than in July 2002, this group dominated the community with 79% of total macroinvertebrate density.

During the studied period, neither Mollusca nor Polychaeta *taxa* were captured upstream and downstream.

Class Insecta was represented by *taxa* from three Orders, Diptera (*Chironomus* sp., Ephydriidae, Tabanidae, Ceratopogonidae, Empididae, Rhagionidae and Brachyceres), Coleoptera (*Berosus spinosus*) and Hemiptera (*Corixa affinis*, *Hesperocorixa sahlbergi*, *Parasigara infuscata*, *Notonecta* sp.).

In the upstream station, August 2001 was the month with lower number of Insecta *taxa* (Figure 4.11), just *Chironomus* sp. and *Hesperocorixa sahlbergi* were captured, whereas in March 2002, eight *taxa* were identified (all Diptera *taxa* and *Berosus spinosus*).

Some of the *taxa* that occurred upstream were not registered downstream, namely Empididae, Rhagionidae and *Corixa affinis*. During June 2001 and January 2002 only *Chironomus* sp. was observed downstream and the maximum number of *taxa* in this station was found in May 2002 (*Chironomus* sp., Ephydriidae, Tabanidae, Ceratopogonidae and *Parasigara infuscata*).

*Chironomus* sp. was the most abundant Insecta in the two stations, ranging from 265 ind m<sup>-2</sup> (October 2001: 67%) to 51593 ind m<sup>-2</sup> (July 2002: 98%) in the upstream station and from 29 ind m<sup>-2</sup> (January 2002: 100%) to 38525 ind m<sup>-2</sup> (May 2002: 99%) in the downstream station. It was observed in all samplings upstream and downstream, being classified in both stations as a constant *taxon* in terms of constancy ( $C_{\text{Upstream}} = C_{\text{Downstream}} = 100\%$ ) and an accessory *taxon* concerning fidelity ( $F_{\text{Upstream}} = F_{\text{Downstream}} = 50\%$ ).

Although Ephydriidae and *Berosus spinosus* were absent from some samplings, these *taxa* were also considered constant in the upstream station, presenting constancy values of 50% and 87.5% respectively. Downstream, Ephydriidae constancy was 25% and *Berosus spinosus* constancy was 12.5%, being both classified as accessory *taxa*. Regarding fidelity, Ephydriidae was preferential upstream ( $F_{\text{Upstream}} = 66.7\%$ ) and accessory downstream ( $F_{\text{Downstream}} = 33.3\%$ ), while *Berosus spinosus* was elective upstream ( $F_{\text{Upstream}} = 87.5\%$ ) and accidental or rare downstream ( $F_{\text{Downstream}} = 12.5\%$ ).

The remaining Insecta *taxa* were classified as accessory ( $10 \leq C < 50$ ) in both stations, with the exception of Empididae, Rhagionidae and *Corixa affinis* in the downstream

station. These three *taxa* were exclusive upstream ( $F = 100\%$ ) and *Notonecta* sp. was exclusive downstream ( $F = 100\%$ ). Ceratopogonidae, Brachyceres and *Parasigara infuscata* were considered accessory ( $20 < F \leq 50$ ) in the two stations; Tabanidae was accessory upstream ( $F_{\text{Upstream}} = 40\%$ ) and preferential downstream ( $F_{\text{Downstream}} = 60\%$ ); *Hesperocorixa sahlbergi* was preferential upstream ( $F_{\text{Upstream}} = 66.7\%$ ) and accessory downstream ( $F_{\text{Downstream}} = 33.3\%$ ).

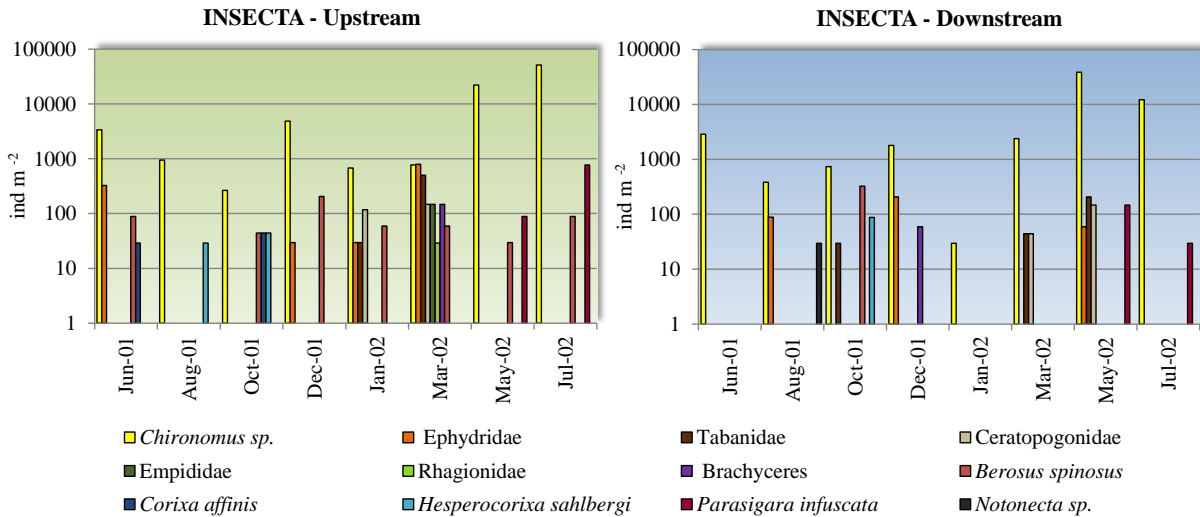


Figure 4.11 - Seasonal variation of Insecta *taxa* densities in Salgados sampling stations.

The Oligochaeta *taxa* found belonged to the Family Tubificidae and Enchytraeidae. In the upstream station, each *taxon* was observed only once (Figure 4.12), Enchytraeidae in January 2002 ( $324 \text{ ind m}^{-2}$ ) and Tubificidae in March 2002 ( $118 \text{ ind m}^{-2}$ ). Downstream, Enchytraeidae was also registered just in January 2002, when the lagoon was in connection with the sea, but its density was lower than upstream ( $29 \text{ ind m}^{-2}$ ). Tubificidae occurred in August 2001 and from March to July 2002, with a maximum density in May 2002 ( $17139 \text{ ind m}^{-2}$ ).

The values of constancy ( $C_{\text{Upstream}} = C_{\text{Downstream}} = 12.5\%$ ) and fidelity ( $F_{\text{Upstream}} = F_{\text{Downstream}} = 50\%$ ) determined for Enchytraeidae correspond to accessory *taxon* in both stations. Tubificidae was also classified as an accessory *taxon* upstream ( $C_{\text{Upstream}} = 12.5\%$ ), but downstream it was constant ( $C_{\text{Downstream}} = 50\%$ ). In terms of fidelity, Tubificidae was considered accidental or rare *taxon* in the upstream station ( $F_{\text{Upstream}} = 20\%$ ) and an elective *taxon* downstream ( $F_{\text{Downstream}} = 80\%$ ).

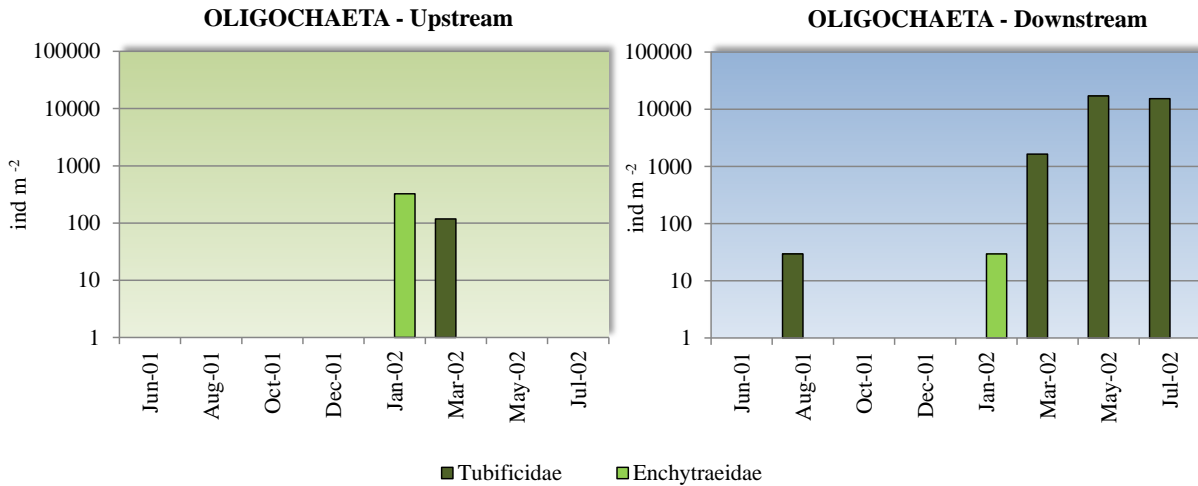


Figure 4.12 - Seasonal variation of Oligochaeta taxa densities in Salgados sampling stations.

Two Crustacea species were identified in Salgados lagoon, the Isopoda *Lekanesphaera hookeri* and the Amphipoda *Corophium multisetosum*, but in the upstream station only *Lekanesphaera hookeri* occurred in July 2002 (354 ind m<sup>-2</sup>) (Figure 4.13). Downstream, the species was absent in October and December 2001, presenting the highest density in July 2002 (40560 ind m<sup>-2</sup>). To what concerns constancy, *Lekanesphaera hookeri* was considered accessory taxon upstream ( $C_{\text{Upstream}} = 12.5\%$ ) and constant taxon downstream ( $C_{\text{Downstream}} = 75\%$ ); regarding fidelity it was classified as accidental or rare upstream ( $F_{\text{Upstream}} = 14.3\%$ ) and elective downstream ( $F_{\text{Downstream}} = 85.7\%$ ).

*Corophium multisetosum* was present just downstream during June and August 2001 and July 2002, when it reached the maximum density (16755 ind m<sup>-2</sup>). It was therefore considered accessory taxon in terms of constancy ( $C_{\text{Downstream}} = 37.5\%$ ) and an exclusive taxon concerning fidelity ( $F_{\text{Downstream}} = 100\%$ ).

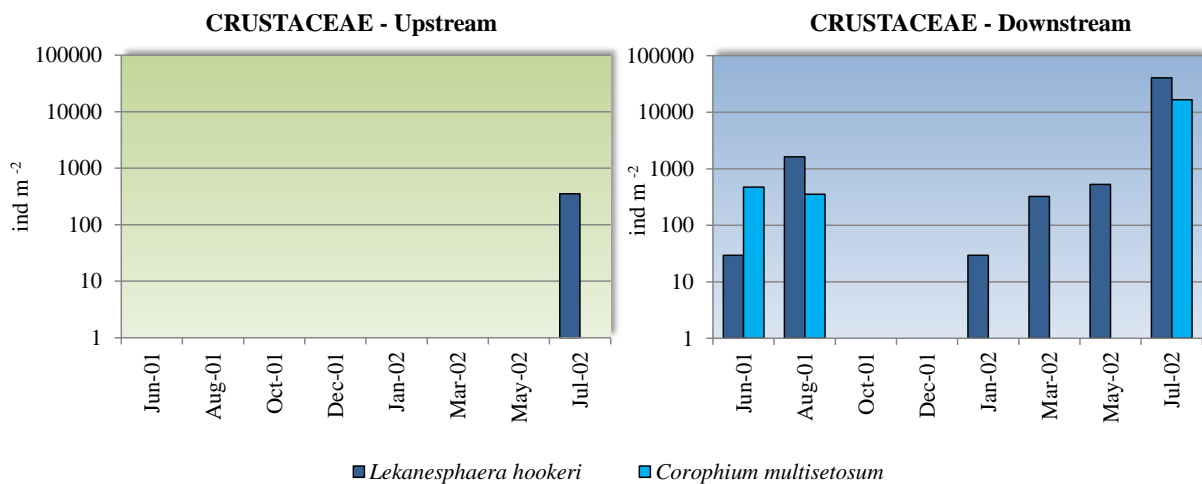


Figure 4.13 - Seasonal variation of Crustacea taxa densities in Salgados sampling stations.

In terms of constancy, both stations had similar percentages of accessory *taxa* (upstream = 79%; downstream = 77%) and constant *taxa* (upstream = 21%; downstream = 23%) (Figure 4.14).

Regarding fidelity, the majority of *taxa* in the two stations were accessory (upstream = 43%; downstream = 54%) and exclusive (upstream = 22%; downstream = 15%). The upstream station presented higher percentages of accidental (or rare) *taxa* and preferential *taxa*, while downstream the percentage of elective *taxa* was greater than in the upstream station.

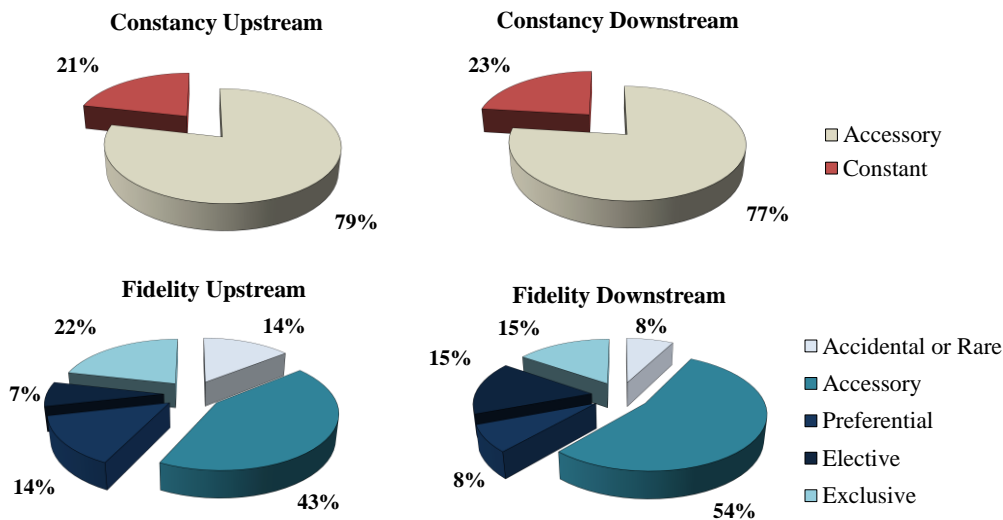


Figure 4.14 – Relative frequency of *taxa* in terms of constancy and fidelity in Salgados sampling stations.

The highest annual means for total Insecta, Insecta Diptera, Insecta Coleoptera, the Insecta Hemiptera *Corixa affinis* and *Parasigara infuscata* and the Oligochaeta Enchytraeidae densities were determined upstream (Table 4.4). Taxonomic richness means were similar in the two stations, but all other benthic macroinvertebrate parameters had higher mean values downstream, namely total benthic macroinvertebrate density, Shannon-Wiener diversity, evenness, the Insecta Hemiptera *Hesperocorixa sahlbergi* and *Notonecta* sp. densities, total Oligochaeta and Oligochaeta Tubificidae densities, Isopoda (*Lekanesphaera hookeri*) and Amphipoda (*Corophium multisetosum*) densities.

Significant statistical differences between stations ( $p < 0.05$ ) were found just for the Insecta Coleoptera *Berosus spinosus* densities (upstream > downstream) and the Isopoda *Lekanesphaera hookeri* densities (upstream < downstream) (Appendix I.K).

Table 4.4 – Annual mean values and standard deviation of benthic macroinvertebrate parameters in Salgados sampling stations.

	<i>Upstream Station</i>	<i>Downstream Station</i>
<b>Benthic communities</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total benthos density (ind m <sup>-2</sup> )	11178 ± 18280	19416 ± 32647
Taxonomic richness	4.38 ± 2.20	4.38 ± 1.30
Shannon-Wiener diversity (bits)	0.90 ± 0.92	1.27 ± 0.43
Evenness	0.38 ± 0.31	0.59 ± 0.21
<b>INSECTA density (ind m<sup>-2</sup>)</b>	<b>11077 ± 18199</b>	<b>7555 ± 13316</b>
<b>Insecta Diptera density (ind m<sup>-2</sup>)</b>	<b>10880 ± 117949</b>	<b>7477 ± 13295</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	10594 ± 18111	7367 ± 13185
Ephydriidae density (ind m <sup>-2</sup> )	147 ± 285	44 ± 74
Tabanidae density (ind m <sup>-2</sup> )	66 ± 176	35 ± 71
Ceratopogonidae density (ind m <sup>-2</sup> )	33 ± 62	24 ± 52
Empididae density (ind m <sup>-2</sup> )	18 ± 52	0 ± 0
Rhagionidae density (ind m <sup>-2</sup> )	4 ± 10	0 ± 0
Brachyceres density (ind m <sup>-2</sup> )	18 ± 52	7 ± 21
<b>Insecta Coleoptera density (ind m<sup>-2</sup>)</b>	<b>72 ± 62</b>	<b>41 ± 115</b>
<i>Berosus spinosus</i> density (ind m <sup>-2</sup> )	72 ± 62	41 ± 115
<b>Insecta Hemiptera density (ind m<sup>-2</sup>)</b>	<b>125 ± 262</b>	<b>37 ± 54</b>
<i>Hesperocorixa sahlbergi</i> density (ind m <sup>-2</sup> )	9 ± 17	11 ± 31
<i>Corixa affinis</i> density (ind m <sup>-2</sup> )	9 ± 17	0 ± 0
<i>Parasigara infusata</i> density (ind m <sup>-2</sup> )	107 ± 268	22 ± 52
<i>Notonecta</i> sp. density (ind m <sup>-2</sup> )	0 ± 0	4 ± 10
<b>OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	<b>56 ± 116</b>	<b>4276 ± 7430</b>
Tubificidae density (ind m <sup>-2</sup> )	15 ± 42	4272 ± 7432
Enchytraeidae density (ind m <sup>-2</sup> )	41 ± 115	4 ± 10
<b>ISOPODA density (ind m<sup>-2</sup>)</b>	<b>44 ± 125</b>	<b>5387 ± 14223</b>
<i>Lekanesphaera hookeri</i> density (ind m <sup>-2</sup> )	44 ± 125	5387 ± 14223
<b>AMPHIPODA density (ind m<sup>-2</sup>)</b>	<b>0 ± 0</b>	<b>2198 ± 5885</b>
<i>Corophium multisetosum</i> density (ind m <sup>-2</sup> )	0 ± 0	2198 ± 5885

In terms of trophic groups, most of the time, the two stations presented distinct compositions, except in July 2002 (Figure 4.15). In the upstream station, deposit feeders and carnivorous/scavengers/omnivorous occurred during all studied months and deposit feeders were dominant, with densities varying from 265 ind m<sup>-2</sup> (67%) in October 2001 to 51593 ind m<sup>-2</sup> (98%) in July 2002. Carnivorous/scavengers/omnivorous minimum and maximum densities were determined in August 2001 (29 ind m<sup>-2</sup>; 3%) and March 2002 (1032 ind m<sup>-2</sup>; 38%). July 2002 was the only month with herbivores (354 ind m<sup>-2</sup>; < 1%), besides deposit feeders and carnivorous/scavengers/omnivorous.

Downstream, deposit feeder was the only trophic group present in all samples, but it was not always the dominant, namely in August 2001. Densities ranged from 59 ind m<sup>-2</sup> (67%) in January to 55723 ind m<sup>-2</sup> (98%) in May 2002, showing the minimum and

maximum densities in different months than upstream. Herbivores were absent in October and December 2001, but in August 2001 they were the most abundant macroinvertebrates downstream (1622 ind m<sup>-2</sup>; 63%). In January 2002, deposit feeders and herbivores were the only trophic groups observed, although herbivores density was minimum (29 ind m<sup>-2</sup>; 33%). The maximum density was reached in July 2002 (40560 ind m<sup>-2</sup>), representing 48% of total macroinvertebrates density in this month. Carnivorous/scavengers/omnivorous were present from August to December 2001 and from March to July 2002. Higher densities were found in October 2001 (442 ind m<sup>-2</sup>; 38%) and May 2002 (501 ind m<sup>-2</sup>; 1%), occurring in different months than upstream and with lower maximum values.

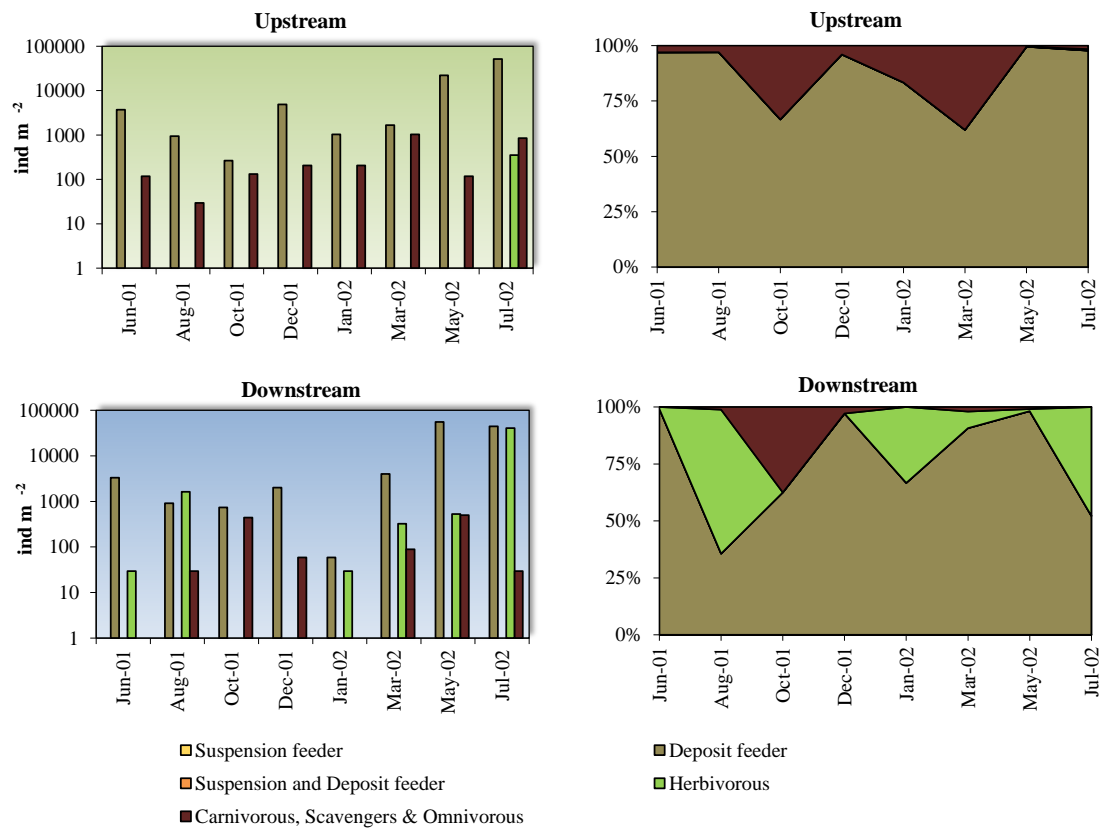


Figure 4.15 - Seasonal variation of trophic groups densities and relative frequency of each group in Salgados sampling stations.

To what concerns ecological groups, macroinvertebrate communities in both stations were mainly composed by *taxa* tolerant to excess organic matter enrichment (Group III – AMBI), except in March 2002, when the upstream station presented higher densities of second-order opportunistic *taxa* (Insecta Diptera, excluding *Chironomus* sp.), which

are indicators of slight to pronounced unbalanced situations (Group IV - AMBI) (Figure 4.16).

Group III *taxa* ranged from 28% (March 2002) to 99% of monthly macroinvertebrate densities in the upstream (annual mean: 79%), whereas in the downstream station it accounted from 61% to 100% (annual mean: 78%).

During all studied period, the presence of *taxa* not assigned in any of the AMBI groups defined by Borja *et al.* (2010) (Insecta Coleoptera and Hemiptera) was registered upstream. This situation also occurred downstream, but just in a few months. In October 2001, these *taxa* accounted 33% and 35% of total macroinvertebrate densities determined in the upstream and downstream stations, respectively.

First-order opportunistic *taxa* (Oligochaeta Tubificidae and Enchytraeidae), associated to pronounced unbalanced situations (Group V – AMBI), occurred in the upstream station just in January and March 2002 representing 26% and 4% of total macroinvertebrate densities in these months. Downstream, these *taxa* were observed during most samplings, being particularly relevant from January to July 2002 as the second ecological group with a greater percentage of macroinvertebrate densities (18% in July 2002; 37% in March 2002).

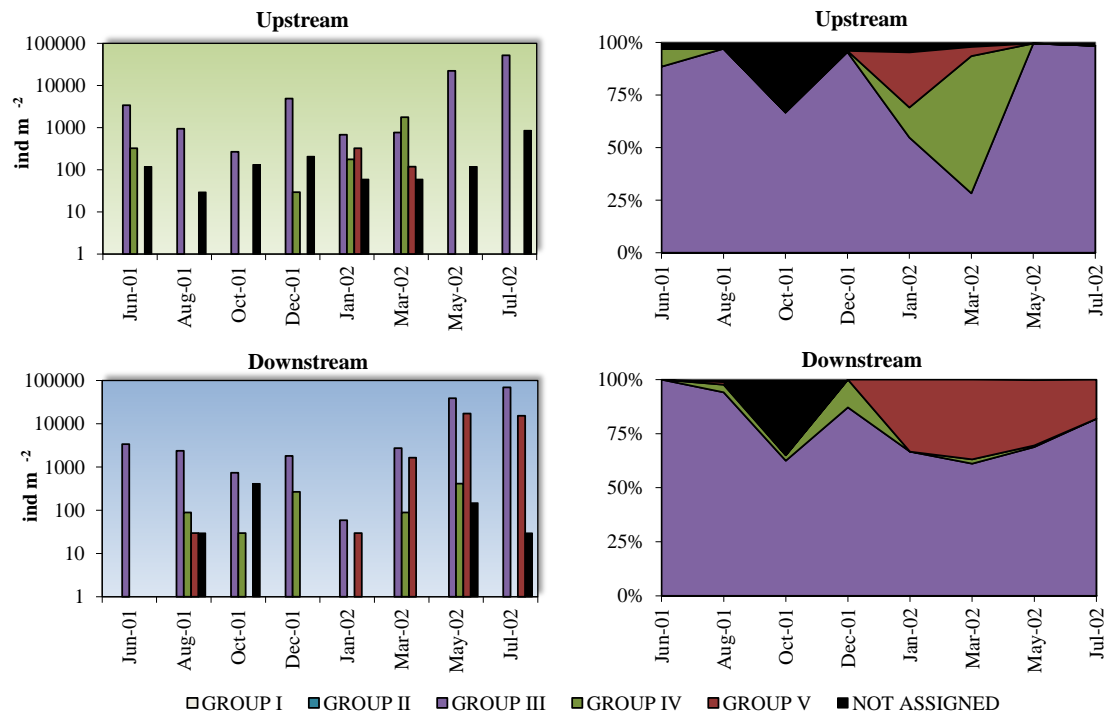


Figure 4.16 - Seasonal variation of AMBI groups densities and relative frequency of each group in Salgados sampling stations.

#### 4.3.2.2. Environmental parameters and benthic macroinvertebrate communities

Some of the benthic macroinvertebrate parameters presented significant linear correlations with environmental parameters (Table 4.5).

Total benthos density was correlated just with water parameters, showing a positive correlation with phaeo-pigments concentration in water and negative correlations with N: P ratio, total solids in suspension, nitrates and total dissolved inorganic nitrogen concentrations. For evenness a positive correlation was determined with cumulative rainfall.

Total Insecta, Insecta Diptera and *Chironomus* sp. densities were negatively correlated with nitrites concentration and N: P ratio. Negative correlations were also determined between the densities of total Insecta, Insecta Diptera and nitrates concentration. Besides these environmental parameters, total Insecta density was negatively associated with total dissolved inorganic nitrogen concentration and positively correlated with phaeo-pigments concentration in water. The densities of Insecta Hemiptera increased with the rise of water level in the lagoon and decreased with the augment of nitrates concentration and *vice-versa*.

The Insecta Coleoptera *Berosus spinosus* was positively correlated with the percentage of phaeo-pigments in the sediment (chlorophyll *a* degradation index), clay and silt contents, presenting negative correlations with sand content and sand: mud ratio.

The densities of the Crustacea Isopoda *Lekanesphaera hookeri*, besides being negatively associated with organic matter content in the sediment, were correlated with the same parameters as *Berosus spinosus*, but the linear relations were inverted.

Total Oligochaeta density showed a negative correlation with the percentage of phaeo-pigments in the sediment (chlorophyll *a* degradation index), just as *Lekanesphaera hookeri*.

The strongest and most significant linear associations were determined between: 1) *Berosus spinosus* density vs. sand: mud ratio, silt content, clay content and chlorophyll *a* degradation index; 2) *Lekanesphaera hookeri* density vs. chlorophyll *a* degradation index.

Table 4.5 - Significant correlations between benthos and environmental parameters (water and sediment) from Salgados lagoon. \*- Correlation significant at the 0.05 level; \*\* - Correlation significant at the 0.01 level.

<i>Benthos</i>	<i>Environmental parameters</i>	<i>Results</i>
<b>Total Benthos Density</b>	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.537; <i>p</i> = 0.032 *
	Total dissolved inorganic nitrogen concentration (DIN)	Rho = -0.512; <i>p</i> = 0.043 *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	R = -0.557; <i>p</i> = 0.025 *
	Total Solids in Suspension (TSS)	R = -0.611; <i>p</i> = 0.012 *
	Phaeo-pigments concentration in water (PhaeW)	R = 0.579; <i>p</i> = 0.019 *
<b>Evenness (J)</b>	Cumulative rainfall (Rain)	Rho = 0.527; <i>p</i> = 0.036 *
<b>Total Insecta density</b>	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.561; <i>p</i> = 0.024 *
	Total dissolved inorganic nitrogen concentration (DIN)	Rho = -0.526; <i>p</i> = 0.036 *
	Nitrites concentration (NO <sub>2</sub> )	R = -0.519; <i>p</i> = 0.039 *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	R = -0.594; <i>p</i> = 0.015 *
	Phaeo-pigments concentration in water (PhaeW)	Rho = 0.508; <i>p</i> = 0.045 *
<b>Insecta Diptera density</b>	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.543; <i>p</i> = 0.030 *
	Nitrites concentration (NO <sub>2</sub> )	R = -0.506; <i>p</i> = 0.046 *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	R = -0.567; <i>p</i> = 0.022 *
<b>Chironomus sp. density</b>	Nitrites concentration (NO <sub>2</sub> )	R = -0.512; <i>p</i> = 0.043 *
	Dissolved inorganic nitrogen and Total phosphorus ratio (N:P)	R = -0.543; <i>p</i> = 0.030 *
<b>Insecta Coleoptera density</b> <i>Berosus spinosus</i>	Phaeo-pigments percentage in sediment (% PhaeS)	Rho = 0.631; <i>p</i> = 0.009 **
	Clay content (%Clay)	Rho = 0.638; <i>p</i> = 0.008 **
	Silt content (%Silt)	Rho = 0.641; <i>p</i> = 0.008 **
	Sand content (%Sand)	Rho = -0.550; <i>p</i> = 0.027 *
	Sand Mud ratio (Sand: Mud)	Rho = -0.687; <i>p</i> = 0.003**
<b>Insecta Hemiptera density</b>	Water level in the lagoon (WLevel)	Rho = 0.559; <i>p</i> = 0.024 *
	Nitrates concentration (NO <sub>3</sub> )	Rho = -0.528; <i>p</i> = 0.036 *
<b>Oligochaeta density</b>	Phaeo-pigments percentage in sediment (% PhaeS)	Rho = -0.603; <i>p</i> = 0.013 *
<b>Isopoda density</b> <i>Lekanesphaera hookeri</i>	Organic matter content in sediment (%OM)	Rho = -0.566; <i>p</i> = 0.022 *
	Phaeo-pigments percentage in sediment (% PhaeS)	Rho = -0.647; <i>p</i> = 0.007 **
	Clay content (%Clay)	Rho = -0.559; <i>p</i> = 0.024 *
	Silt content (%Silt)	Rho = -0.568; <i>p</i> = 0.022 *
	Sand content (%Sand)	Rho = 0.535; <i>p</i> = 0.033 *
	Sand Mud ratio (Sand: Mud)	Rho = 0.570; <i>p</i> = 0.021 *

A first Canonical Correspondence Analysis (CCA) was done, including five of the environmental variables that were more relevant for the characterization of stations and months (cumulative rainfall, water temperature, total dissolved inorganic nitrogen concentration in the water, orthophosphates concentration in the water and phaeo-pigments concentration in the sediment), according to the PCA previously performed (Figure 2.25). However, the relations among these environmental variables and benthic macroinvertebrate groups densities were not statistically significant (Monte Carlo test: *p* = 0.602).

A new CCA was then performed (Figure 4.17) with four environmental variables (water temperature, chlorophyll *a* concentration in the sediment, sand content and organic matter content in the sediment), that presented significant relations with the benthic macroinvertebrate groups densities (Monte Carlo test: *p* = 0.002).

In both CCA's, benthic macroinvertebrate densities were presented as major taxonomic groups instead of *taxa* due to the low frequency occurrence of most *taxa* in the lagoon. The Crustacea Isopoda (*Lekanesphaera hookeri*) and Crustacea Amphipoda (*Corophium multisetosum*) densities were positively related with higher values of water temperature, chlorophyll *a* concentration in the sediment, sand content, and lower values of organic matter content in the sediment. Oligochaeta densities were also higher in sediments with greater sand content and lower organic matter. Insecta Diptera, Hemiptera and Coleoptera preferred sediments with higher organic matter content and less sandy, being most abundant in the upstream station.

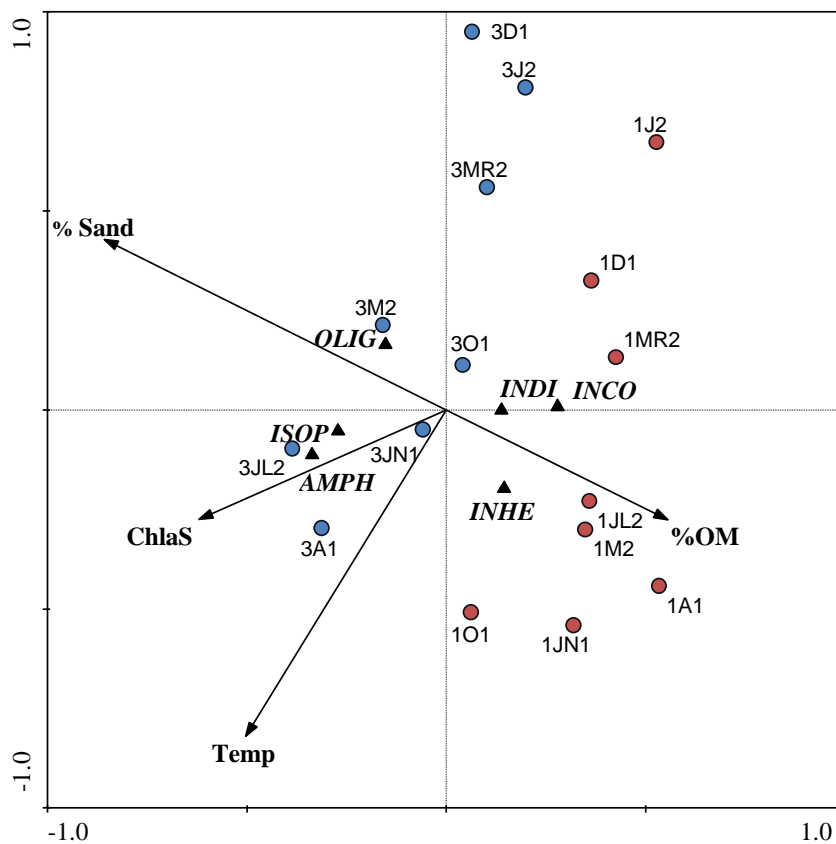


Figure 4.17 - Canonical correspondence analysis performed with the benthic macroinvertebrate *taxa* (total density *per station*) from Salgados lagoon. Cumulative percentage variance explained by axes: Species - I = 47.0% and I + II = 56.1%; and species-environment relation - I = 80.0% and I + II = 95.6%. Monte Carlo test of all canonical axes:  $p = 0.002$ .

*Station codes*: First character corresponds to the sampling station (1 - upstream, 3 - downstream) and subsequent ones to month (JN- June; A- August; O- October; D- December; J- January; M- May; JL- July) and year of survey (1 - 2001, 2 - 2002). *Environmental parameters*: Temp- water temperature; %Sand- Sand content; %OM- Percentage of organic matter content in the sediment; ChlaS- Chlorophyll *a* concentration in the sediment. *Benthic macroinvertebrate groups*: OLIG – Oligochaeta; AMPH – Amphipoda; ISOP- Isopoda; INHE – Insecta Hemiptera; INDI- Insecta Diptera; INCO- Insecta Coleoptera

#### 4.3.2.3. Comparison of benthic macroinvertebrate communities during isolation and connection of the lagoon with the sea

Salgados sampling in January 2002 was made when the lagoon was opened and with direct influence from the sea. In December 2001 the lagoon was isolated and so benthic communities from these two periods were compared. Two previous openings occurred, one in the middle of October (before sampling) and another between December and January samplings.

Both stations showed a decrease of total benthos densities, which was more accentuated upstream. Nevertheless, total density was greater upstream (Table 4.6). This decrease was associated to the reduction in Insecta densities, particularly *Chironomus* sp. Oligochaeta (Enchytraeidae) were absent in December and in January, it was the only taxonomic group to increase in the two stations, presenting a greater variation and density upstream.

In January, taxonomic richness in the upstream station was composed by six *taxa*, most of them Insecta, *Chironomus* sp. (54.72% of total benthos in this station), Ceratopogonidae (9.52%), *Berosus spinosus* (4.76%), Tabanidae (2.42%), Ephydriidae (2.34%) and Oligochaeta Enchytraeidae (26.23%). The Insecta Ceratopogonidae and Tabanidae did not occur in this station during December sampling. For the downstream station, only three *taxa* were determined, the Isopoda *Lekanesphaera hookeri* (34.48% of total benthos in this station), the Insecta *Chironomus* sp. (33.33% of total benthos in this station) and Oligochaeta Enchytraeidae (32.18%). Insecta Brachyceres was observed in December, but the Isopoda *Lekanesphaera hookeri* was not.

Shannon-Wiener diversity and evenness increased in both stations, but the values and variation augment was in opposite directions. Diversity was lower downstream and evenness was higher and *vice-versa* in the upstream station.

When the lagoon was opened to the sea, in the upstream station the trophic groups recorded were the same as in December, deposit feeders (83%) and carnivorous/ scavengers/ omnivorous (17%), nevertheless the density of deposit feeders showed a decrease whereas the density of carnivorous/ scavengers/ omnivorous had no variation. Downstream, the number of trophic groups reduced from three to two. In January, the community was composed by deposit feeders (67%) and herbivores (33%), and none of the suspension feeders or carnivorous/ scavengers/ omnivorous observed in December

were present. The major decrease was determined in deposit feeders' density, being greater upstream.

Table 4.6 – Benthic macroinvertebrate communities in January 2002 and variation between values when the lagoon was isolated (December 2001) and connected to the sea (January 2002) in Salgados sampling stations.

	<i>Upstream station</i>		<i>Downstream station</i>	
	<b>Jan-02</b>	<b>Variation</b>	<b>Jan-02</b>	<b>Variation</b>
<b>Benthic communities</b>				
Total benthos density (ind m <sup>-2</sup> )	1239	<b>-3864</b>	87	<b>-1976</b>
Taxonomic richness	6	<b>3</b>	3	<b>0</b>
Shannon-Wiener diversity (bits)	1.77	<b>1.44</b>	1.58	<b>0.77</b>
Evenness	0.69	<b>0.52</b>	1.00	<b>0.59</b>
<b>Total INSECTA density (ind m<sup>-2</sup>)</b>	<i>914</i>	<b>-4189</b>	<i>29</i>	<b>-2035</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	678	<b>-4190</b>	29	<b>-1770</b>
Ephydriidae density (ind m <sup>-2</sup> )	29	<b>0</b>	0	<b>-206</b>
Tabanidae (ind m <sup>-2</sup> )	30	<b>30</b>	0	<b>0</b>
Ceratopogonidae (ind m <sup>-2</sup> )	118	<b>118</b>	0	<b>0</b>
Empididae density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Rhagionidae density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Brachyceres density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-59</b>
<i>Berosus spinosus</i> density (ind m <sup>-2</sup> )	59	<b>-147</b>	0	<b>0</b>
<i>Hesperocorixa sahlbergi</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Corixa affinis</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Parasigara infuscata</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Notonecta</i> sp. density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<b>Total OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	<i>325</i>	<b>325</b>	<i>28</i>	<b>28</b>
Tubificidae density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Enchytraeidae density (ind m <sup>-2</sup> )	325	<b>325</b>	28	<b>28</b>
<b>Total ISOPODA density (ind m<sup>-2</sup>)</b>	<i>0</i>	<b>0</b>	<i>30</i>	<b>30</b>
<i>Lekanesphaera hookeri</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	30	<b>30</b>
<b>AMPHIPODA density (ind m<sup>-2</sup>)</b>	<i>0</i>	<b>0</b>	<i>0</i>	<b>0</b>
<i>Corophium multisetosum</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<b>TROPHIC GROUPS DENSITIES</b>				
Deposit feeders (ind m <sup>-2</sup> )	1032	<b>-3865</b>	57	<b>-1948</b>
Suspension/ deposit feeders (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Herbivores (ind m <sup>-2</sup> )	0	<b>0</b>	30	<b>30</b>
Carnivorous/ scavengers/ omnivorous (ind m <sup>-2</sup> )	207	<b>0</b>	0	<b>-59</b>
<b>AMBI GROUPS DENSITIES</b>				
Group III (ind m <sup>-2</sup> )	678	<b>-4189</b>	59	<b>-1740</b>
Group IV (ind m <sup>-2</sup> )	177	<b>147</b>	0	<b>-265</b>
Group V (ind m <sup>-2</sup> )	324	<b>324</b>	29	<b>29</b>

In January 2002, the densities of *taxa* tolerant to excess organic matter enrichment (Group III - AMBI) declined in both stations. The downstream station also presented a

decrease in densities of second-order opportunistic *taxa*, which are indicators of slight to pronounced situations (Group IV - AMBI); and no *taxa* from this group was found when the lagoon was in connection with the sea. In this station, there was also a small increase in densities of second-order opportunistic *taxa* associated to pronounced unbalanced situations (Group V –AMBI). Upstream was determined an augment of group IV and group V *taxa* densities.

### **4.3.3. Comparison of the two coastal lagoons**

#### **4.3.3.1. Benthic macroinvertebrate communities**

The total number of *taxa* identified in the Foz de Almargem (10) was lower than and the number determined for Salgados lagoon (16).

Benthic macroinvertebrate community in Foz de Almargem was mostly composed by Mollusca (40.0%) and Polychaeta *taxa* (30.0%), while in Salgados the majority of the *taxa* belonged to Insecta (75.0%) and no Mollusca *taxa* were collected in the lagoon.

Taxonomic richness in the two lagoons followed distinct trends, except in August and December 2001 (Figure 4.18). In Foz de Almargem, the lowest number of *taxa* (5) was observed in January 2002 and the highest value (9) occurred in August and October 2001. The maximum richness registered in Salgados was 10 *taxa*, found in March 2002, and the minimum value of 4 *taxa* was determined in December 2001.

Total macroinvertebrate densities in the lagoons showed a different evolution along time. Monthly mean values determined in Foz de Almargem were higher than the ones obtained in Salgados lagoon, except in May and July 2002. In Foz de Almargem, the highest mean density was 33717 ind m<sup>-2</sup> (January 2002) and the lowest was 5585 ind m<sup>-2</sup> (July 2002). In Salgados lagoon, the lowest mean density was found in January 2002 (664 ind m<sup>-2</sup>), whereas the higher value occurred in July 2002 (68864 ind m<sup>-2</sup>).

Shannon-Wiener diversity index and evenness presented higher mean values in Foz de Almargem during May ( $H' = 1.45$ ;  $E = 0.56$ ) and July 2002 ( $H' = 1.52$ ;  $E = 0.72$ ), corresponding to the greatest values determined in the lagoon during the studied period. The lower values of diversity ( $H' = 0.59$ ) were observed in June 2001 and January 2002. Evenness was minimum in December 2001 ( $E = 0.27$ ). In Salgados lagoon, the months

with lower diversity and evenness were December 2001 ( $H' = 0.51$ ;  $E = 0.29$ ) and May 2002 ( $H' = 0.55$ ;  $E = 0.22$ ), while the highest values were recorded in January ( $H' = 1.68$ ;  $E = 0.84$ ) and March 2002 ( $H' = 2.03$ ;  $E = 0.72$ ). The minimum and the maximum values of diversity and evenness were determined in Salgados lagoon.

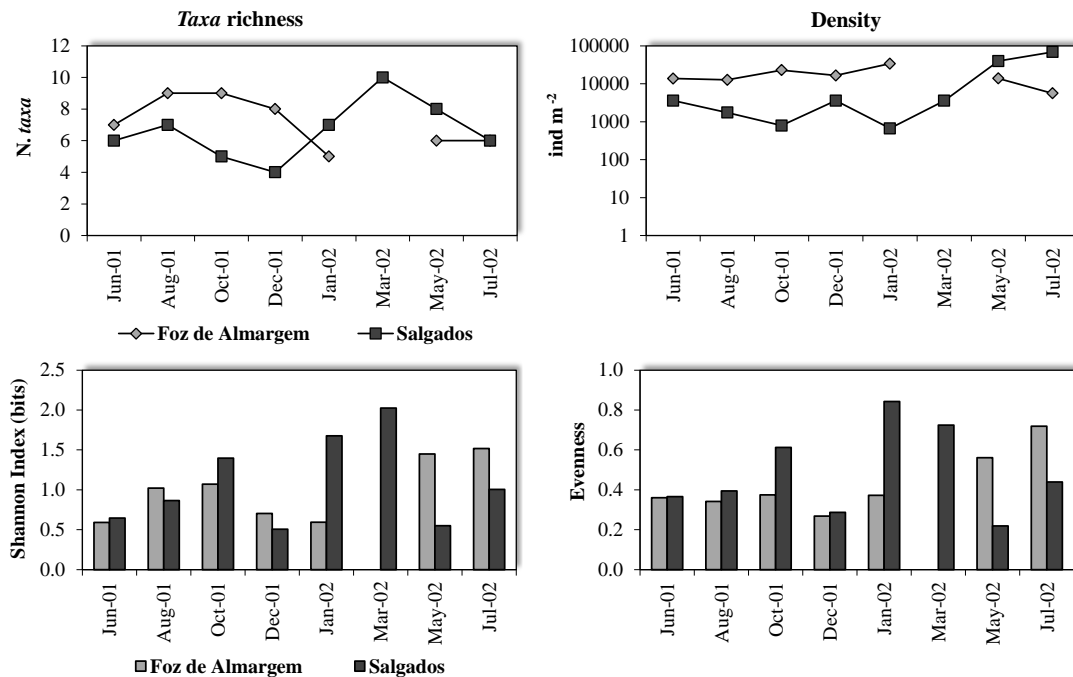


Figure 4.18 - Evolution of benthic macro invertebrate richness, total density, Shannon-Wiener diversity ( $H'$ ) and evenness ( $E$ ) monthly mean values in Foz de Almargem and Salgados lagoons.

The two lagoons were dominated by different groups of benthic macroinvertebrates, with Mollusca being the most abundant group in Foz de Almargem, while Insecta, and Crustacea were the more relevant groups in Salgados lagoon (Figure 4.19).

In Foz de Almargem, Mollusca presented monthly mean densities greater than any other benthic macroinvertebrate group, except in July 2002, when Mollusca decreased to a minimum density of 2271 ind m<sup>-2</sup> (41% of total density) and Crustacea density was slightly higher (2311 ind m<sup>-2</sup>; 41%). July 2002 was also the month with greater percentage of Insecta (18%). From June 2001 to January 2002, Mollusca monthly mean densities were above 10000 ind m<sup>-2</sup>, reaching the maximum value in January 2002 (32075 ind m<sup>-2</sup>; 95%) when the lagoon was connected to the sea. Although Crustacea taxa were not observed in the lagoon during June 2001, this was the second major group with densities ranging from 167 ind m<sup>-2</sup> (1%) in August 2001 to 4376 ind m<sup>-2</sup> (32%) in May 2002.

Insecta and Polychaeta were observed during all studied period and represented the third and fourth most important groups in terms of densities. Both groups presented their maximum densities in October 2001 (Insecta: 1377 ind m<sup>-2</sup>; 6%; Polychaeta: 1042 ind m<sup>-2</sup>; 5%). The lowest Insecta density was determined in June 2001 (59 ind m<sup>-2</sup>; < 1%) and July 2002 was the month with lowest density of Polychaeta (20 ind m<sup>-2</sup>; < 1%). Oligochaeta were observed in Foz de Almargem in May 2002, with a mean density of 69 ind m<sup>-2</sup> and accounting less than 1% of macroinvertebrates density.

In Salgados lagoon, Insecta was the only taxonomic group observed during the whole studied period and most of the time it showed monthly mean densities greater than the other groups, varying from 472 ind m<sup>-2</sup> (71%) in January 2002 to 32345 ind m<sup>-2</sup> (47%) in July 2002. Comparatively to Foz de Almargem, monthly mean values were higher in Salgados, except in October 2001. Crustacea were absent from Salgados lagoon in October and December 2001, but in August 2001 it was the group with higher mean density (988 ind m<sup>-2</sup>; 57%), together with Insecta (737 ind m<sup>-2</sup>; 42%). In July 2002 was determined the highest mean density of Crustacea (28835 ind m<sup>-2</sup>; 42%), which was the second greatest value after Insecta. Crustacea monthly mean values in Salgados lagoon were lower than in Foz de Almargem, except in June and August 2001 and in July 2002. Oligochaeta presence in Salgados lagoon was more regular than in Foz de Almargem, although there were no observations in June, October and December 2001. The highest mean densities were registered in May (8570 ind m<sup>-2</sup>; 22%) and July 2002 (7685 ind m<sup>-2</sup>; 11%).

Only two *taxa* were found in both lagoons, the Isopoda *Lekanesphaera hookeri* and the Insecta Diptera *Chironomus* sp..

*Chironomus* sp. was observed during the whole studied period in both lagoons (Figure 4.20), but monthly mean densities were higher in Salgados lagoon, except in October 2001. Mean densities in Foz de Almargem ranged from 59 ind m<sup>-2</sup> in June 2001 to 1377 ind m<sup>-2</sup> in October 2001, whereas in Salgados lagoon, mean densities varied from 354 ind m<sup>-2</sup> in January 2005 to 31903 ind m<sup>-2</sup> in July 2002.

The presence of *Lekanesphaera hookeri* simultaneously in the two lagoons was registered in August 2001, January, May and July 2002. Salgados lagoon showed higher densities than Foz de Almargem in August 2001 (811 ind m<sup>-2</sup> > 167 ind m<sup>-2</sup>) and July 2002 (20457 ind m<sup>-2</sup> > 2311 ind m<sup>-2</sup>), and lower densities in January (15 ind m<sup>-2</sup> < 1406

ind m<sup>-2</sup>) and May 2002 (265 ind m<sup>-2</sup> < 4376 ind m<sup>-2</sup>). Besides these months, *Lekanesphaera hookeri* was identified in Foz de Almargem in October and December 2001, while in June 2001 the species was found only in Salgados lagoon.

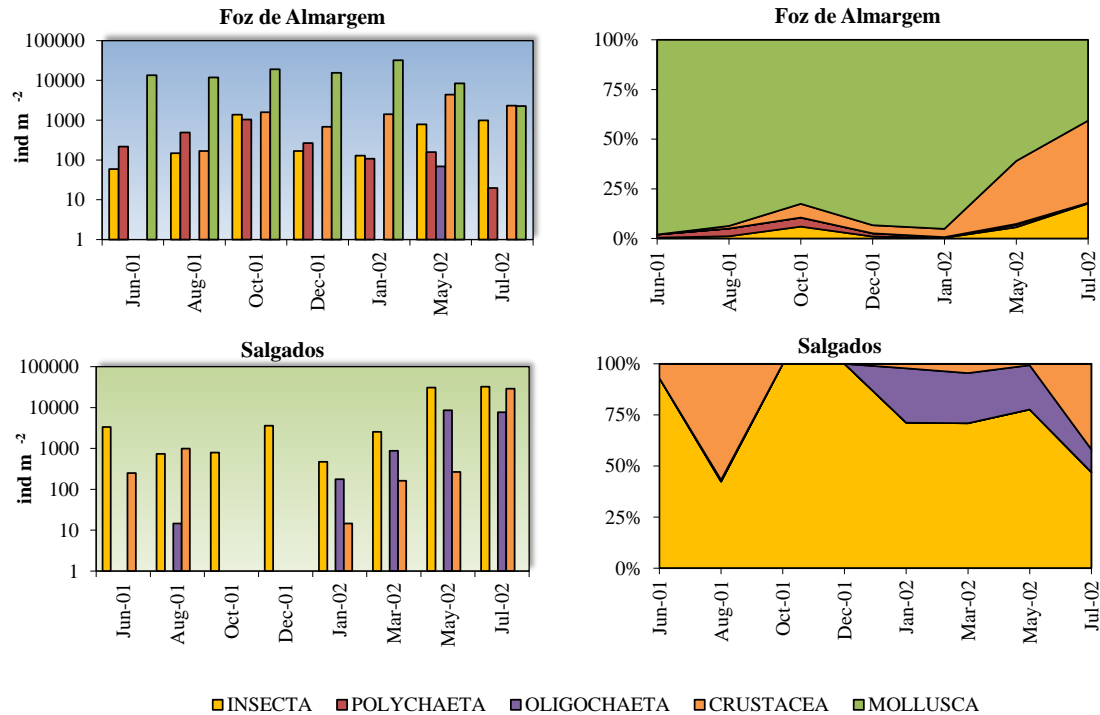


Figure 4.19 - Seasonal variation of the main taxonomic benthic groups' densities and relative frequency of each group in Foz de Almargem and Salgados lagoons.

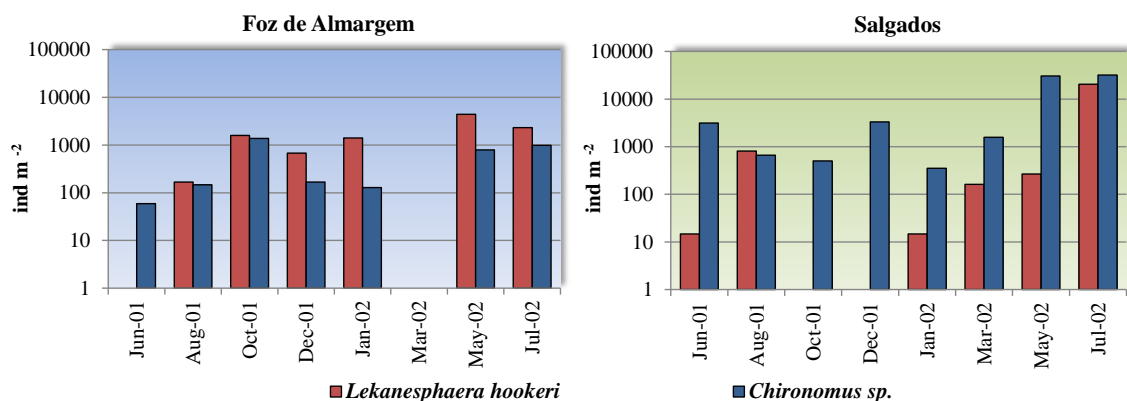


Figure 4.20 - Monthly mean abundances of benthic macroinvertebrate *taxa* that occurred in both lagoons.

In terms of constancy, during the whole studied period, Foz de Almargem presented a greater percentage of constant *taxa* (60%), followed by accessory *taxa* (30%) and

accidental *taxa* (10%). The constant *taxa* category ( $C \geq 50$ ) included *Chironomus* sp., *Hediste diversicolor*, *Lekanesphaera hookeri*, *Hydrobia ulvae*, *Abra segmentum* and *Cerastoderma glaucum*. The accessory *taxa* ( $10 \leq C < 50$ ) in the lagoon were *Capitella capitata*, *Ventrosia ventrosa* and Oligochaeta. An unidentified Polychaeta was the accidental *taxon* ( $C < 10$ ).

In Salgados lagoon, constant *taxa* represented just 13% of the *taxa* identified in the lagoon and the majority of *taxa* were accessory (69%). Accidental *taxa* accounted 19%. *Chironomus* sp. and *Berosus spinosus* were the constant *taxa* in Salgados lagoon; accessory *taxa* included the Insecta Ephydriidae, Tabanidae, Ceratopogonidae, Brachyceres, *Corixa affinis*, *Hesperocorixa sahlbergi*, *Parasigara infuscata*, Oligochaeta Tubificidae and Enchytraeidae, *Lekanesphaera hookeri* and the Amphipoda *Corophium multisetosum*; accidental *taxa* were the Insecta Empididae, Rhagionidae and *Notonecta* sp..

In Foz de Almargem lagoon, the annual mean values of total benthic macroinvertebrates, Polychaeta and Mollusca densities were higher than in Salgados. The remaining parameters presented higher values in Salgados lagoon, with the exception of taxonomic richness which was approximately the same in the two lagoons (Table 4.7).

The T-Student test and Mann-Whitney U test, used to compare the annual mean values of benthic macroinvertebrate parameters from Foz de Almargem and Salgados lagoons, determined that total macroinvertebrate densities presented significant differences ( $0.05 \geq p > 0.01$ ) and that Insecta densities, *Chironomus* sp. densities and *Lekanesphaera hookeri* densities had highly significant differences ( $p \leq 0.01$ ) in the two lagoons (Appendix I.L).

Regarding trophic groups, Foz de Almargem presented a greater diversity of groups (suspension feeders, deposit feeders, suspension/deposit feeders, herbivorous and carnivorous/scavengers/omnivorous) than Salgados lagoon (deposit feeders, herbivorous and carnivorous/scavengers/omnivorous) (Figure 4.21).

Table 4.7 – Annual mean values and standard deviation of benthic macroinvertebrate parameters in Foz de Almagem and Salgados lagoons.

	<i>Foz de Almagem</i>	<i>Salgados</i>
<b>Benthic macroinvertebrate communities</b>	<b>Mean±Std.dev.</b>	<b>Mean±Std.dev.</b>
Total benthos density (ind m <sup>-2</sup> )	16995 ± 8986	15297 ± 25292
Taxonomic richness	7.14 ± 1.57	6.63 ± 1.85
Shannon-Wiener diversity (bits)	0.99 ± 0.39	1.08 ± 0.56
Evenness	0.43 ± 0.16	0.49 ± 0.22
<b>INSECTA density (ind m<sup>-2</sup>)</b>	<b>521 ± 524</b>	<b>9318 ± 13771</b>
<b>Insecta Diptera density (ind m<sup>-2</sup>)</b>	<b>521 ± 524</b>	<b>9180 ± 13678</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	521 ± 524	8980 ± 13732
Ephydriidae density (ind m <sup>-2</sup> )	0 ± 0	96 ± 135
Tabanidae density (ind m <sup>-2</sup> )	0 ± 0	51 ± 96
Ceratopogonidae density (ind m <sup>-2</sup> )	0 ± 0	29 ± 41
Empididae density (ind m <sup>-2</sup> )	0 ± 0	9 ± 26
Rhagionidae density (ind m <sup>-2</sup> )	0 ± 0	2 ± 5
Brachyceres density (ind m <sup>-2</sup> )	0 ± 0	13 ± 27
<b>Insecta Coleoptera density (ind m<sup>-2</sup>)</b>	<b>0 ± 0</b>	<b>56 ± 60</b>
<i>Berosus spinosus</i> density (ind m <sup>-2</sup> )	0 ± 0	56 ± 60
<b>Insecta Hemiptera density (ind m<sup>-2</sup>)</b>	<b>0 ± 0</b>	<b>82 ± 136</b>
<i>Hesperocorixa sahlbergi</i> density (ind m <sup>-2</sup> )	0 ± 0	5 ± 9
<i>Corixa affinis</i> density (ind m <sup>-2</sup> )	0 ± 0	10 ± 23
<i>Parasigara infusate</i> (ind m <sup>-2</sup> )	0 ± 0	65 ± 141
<i>Notonecta</i> sp. (ind m <sup>-2</sup> )	0 ± 0	2 ± 5
<b>POLYCHAETA density (ind m<sup>-2</sup>)</b>	<b>329 ± 350</b>	0 ± 0
<i>Capitella capitata</i> (ind m <sup>-2</sup> )	206 ± 368	0 ± 0
<i>Hediste diversicolor</i> (ind m <sup>-2</sup> )	118 ± 67	0 ± 0
<b>OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	<b>10 ± 26</b>	<b>2165 ± 3699</b>
<b>Tubificidae (ind m<sup>-2</sup>)</b>	0 ± 0	2143 ± 3713
<b>Enchytraeidae (ind m<sup>-2</sup>)</b>	0 ± 0	22 ± 63
<b>CRUSTACEAE density (ind m<sup>-2</sup>)</b>	<b>1503 ± 1508</b>	<b>3814 ± 10115</b>
<b>Isopoda density (ind m<sup>-2</sup>)</b>	<b>1503 ± 1508</b>	<b>2716 ± 7174</b>
<i>Lekanesphaera hookeri</i> (ind m <sup>-2</sup> )	1503 ± 1508	2716 ± 7174
<b>Amphipoda density (ind m<sup>-2</sup>)</b>	<b>0 ± 0</b>	<b>1099 ± 2943</b>
<i>Corophium multisetosum</i> (ind m <sup>-2</sup> )	0 ± 0	1099 ± 2943
<b>MOLLUSCA density (ind m<sup>-2</sup>)</b>	<b>14633 ± 9341</b>	<b>0 ± 0</b>
<b>Gastropoda density (ind m<sup>-2</sup>)</b>	<b>13901 ± 9726</b>	<b>0 ± 0</b>
<i>Ventrosia ventrosa</i> (ind m <sup>-2</sup> )	236 ± 300	0 ± 0
<i>Hydrobia ulvae</i> (ind m <sup>-2</sup> )	13665 ± 9715	0 ± 0
<b>Bivalvia density (inds m<sup>-2</sup>)</b>	<b>732 ± 904</b>	<b>0 ± 0</b>
<i>Abra segmentum</i> (ind m <sup>-2</sup> )	225 ± 382	0 ± 0
<i>Cerastoderma glaucum</i> (ind m <sup>-2</sup> )	507 ± 853	0 ± 0

In Foz de Almagem, during the whole studied period deposit feeders presented higher monthly mean densities than the other trophic groups, ranging from 2763 ind m<sup>-2</sup> (50%) in July 2002 to 32193 ind m<sup>-2</sup> (95%) in January 2002. Suspension feeders represented

the second major trophic group in June (560 ind m<sup>-2</sup>; 4%) and August 2001 (2370 ind m<sup>-2</sup>; 19%); in the following months, herbivores were the macroinvertebrates with greater mean densities after deposit feeders. In May 2002 was determined the highest mean abundance of herbivores (4376 ind m<sup>-2</sup>; 32%), but it was in July 2002 that this group showed a mean density closer to deposit feeders (2311 ind m<sup>-2</sup>; 41%). Suspension/Deposit feeders and carnivorous/scavengers/omnivorous were found in the lagoon during all sampled months; mean densities of suspension/deposit feeders varied from 10 ind m<sup>-2</sup> (< 1%) in January 2002 to 1072 ind m<sup>-2</sup> (8%) in May 2002, whereas carnivorous/scavengers/omnivorous mean densities went from 20 ind m<sup>-2</sup> (< 1%) in July 2002 to 197 ind m<sup>-2</sup> (1%) in December 2001.

In Salgados lagoon, deposit feeders and carnivorous/scavengers/omnivorous were the trophic groups observed in the lagoon during the whole studied period. In June and August 2001 and from January to July 2002, herbivores were also present; no suspension feeders were found during the studied period.

Deposit feeders were the macroinvertebrates with greater mean densities during most of samplings; the highest mean value occurred in July 2002 (47965 ind m<sup>-2</sup>; 70%). Monthly mean densities of deposit feeders in Salgados lagoon were lower than in Foz de Almargem, except in May and July 2002. Herbivores presented the highest mean densities in August 2001 (811 ind m<sup>-2</sup>; 46%) and July 2002 (20457 ind m<sup>-2</sup>; 30%), being the second most relevant trophic group in these months. Carnivorous/scavengers/omnivorous mean densities ranged from 29 ind m<sup>-2</sup> (2%) in August 2001 to 560 ind m<sup>-2</sup> (16%) in March 2002. From October 2001 until March 2002, this was the second trophic group with higher densities, after deposit feeders.

Salgados presented greater annual mean densities of herbivorous (Salgados: 2716 ind m<sup>-2</sup>; Foz de Almargem: 1503 ind m<sup>-2</sup>) and carnivorous/scavengers/omnivorous (Salgados: 241 ind m<sup>-2</sup>; Foz de Almargem: 118 ind m<sup>-2</sup>), whereas the annual mean density of deposit feeders was higher in Foz de Almargem (Salgados: 14643 ind m<sup>-2</sup>; Foz de Almargem: 12344 ind m<sup>-2</sup>).

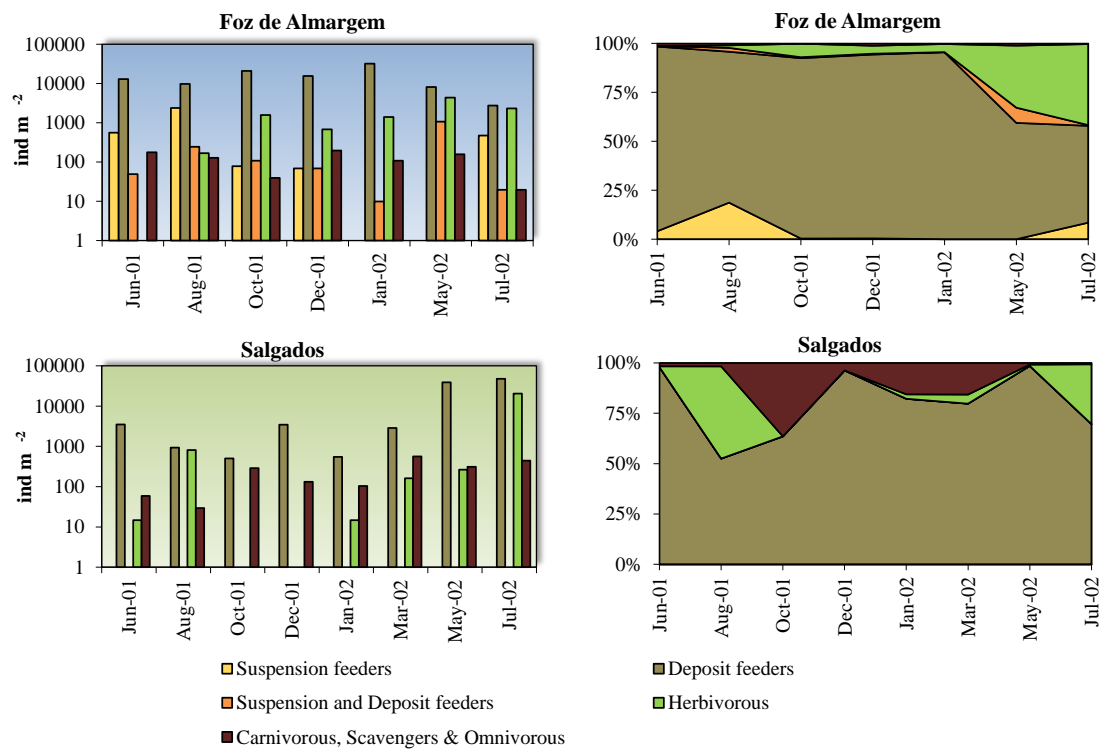


Figure 4.21 - Seasonal variation of trophic groups densities and relative frequency of each group in Foz de Almagem and Salgados lagoons.

In terms of ecological groups, the composition of benthic macroinvertebrate communities in Foz de Almagem and Salgados was distinct, although both lagoons were dominated by *taxa* tolerant to excess organic matter enrichment (Group III – AMBI) (Fig. 4.22). In Foz de Almagem, the densities of group III *taxa* accounted 96% (October 2001) to 100% of the monthly mean densities (annual mean: 99%), whereas in Salgados lagoon these *taxa* represented 49% (March 2002) to 94% (June 2001) (annual mean: 77%).

The remaining *taxa* identified in Foz de Almagem belong to group V, which include first-order opportunistic *taxa* associated to pronounced unbalanced situations. Although these *taxa* occurred during most of the studied period, they had little expression in terms of monthly mean densities and annual mean percentage (1%).

In Salgados lagoon, group V *taxa* were absent in June, October and December 2001, but in the other months comprised 3% to 27% of monthly mean densities (annual mean: 11%). Second-order opportunistic *taxa*, indicators of slight to pronounced unbalanced situations (Group IV - AMBI), were observed in Salgados lagoon during all studied period except in July 2002. These *taxa* represented 1% to 26% of monthly mean densities (annual mean: 7%).

Some of the *taxa* found in Salgados are not assigned in any of the AMBI groups defined by Borja *et al.* (2010).

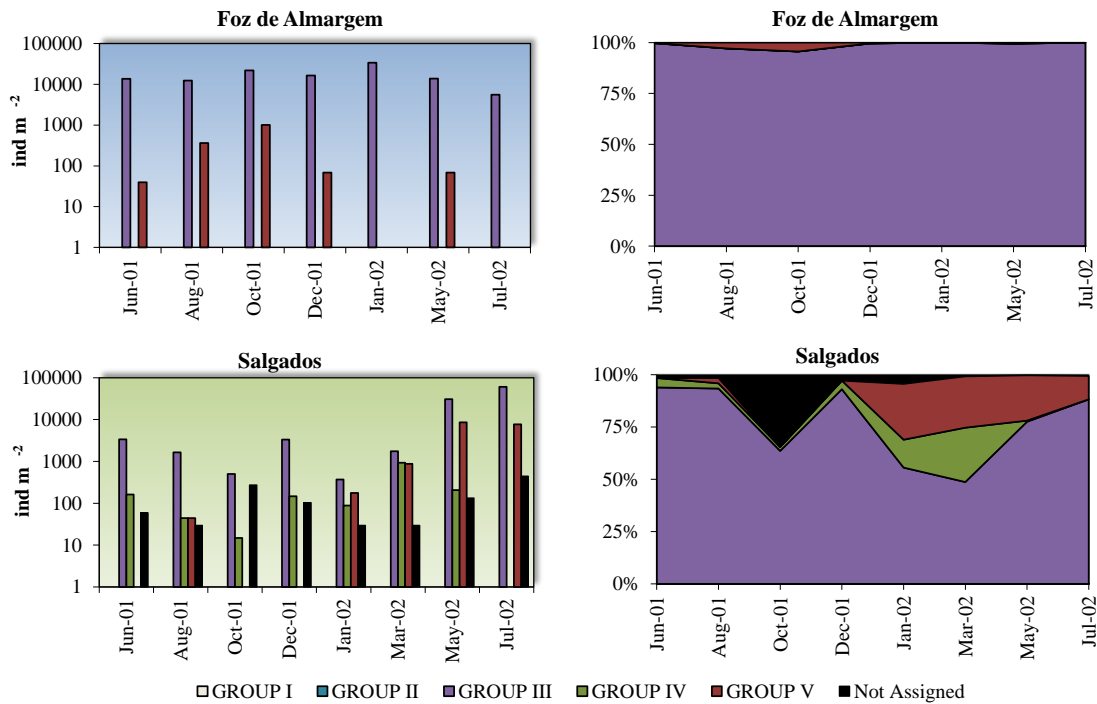


Figure 4.22 - Seasonal variation of AMBI groups densities and relative frequency of each group in Foz de Almagem and Salgados lagoons.

#### 4.3.3.2. Environmental parameters and benthic macroinvertebrate communities

Foz de Almagem presented a greater number of significant correlations between benthic macroinvertebrates and environmental parameters than Salgados lagoon, but for the same benthic macroinvertebrate parameters were determined correlations with different environmental parameters in the two lagoons (Table 4.8).

In Foz de Almagem, benthic macroinvertebrate parameters were mainly correlated with phytopygments in the sediment (chlorophyll *a* and phaeo-pigments), phytopygments in the water column (chlorophyll *a*, phaeo-pigments and phytopygments diversity), some physical and chemical water parameters such as salinity, total solids in suspension, dissolved oxygen concentration, and also the water level in the lagoon.

Benthic macroinvertebrate parameters in Salgados lagoon were mostly correlated with sediment grain size characteristics, organic matter content and chlorophyll *a* degradation in the sediment; some physical and chemical water parameters (nitrogen

compounds and total solids in suspension concentrations) and also with cumulative rainfall.

Table 4.8 - Resume of the significant correlations between benthic macroinvertebrate communities and environmental parameters in Foz de Almargem and Salgados lagoons. (+) Positive correlation; (-) Negative correlation; (\*) Correlation significant at the 0.05 level; (\*\*) Correlation significant at the 0.01 level.

<i>Benthic macroinvertebrate parameters</i>	<i>Environmental parameters</i>	<i>Foz de Almargem</i>	<i>Salgados</i>
Benthos Density	pH	(-) *	
	Nitrates concentration		(-) *
	Total dissolved inorganic nitrogen concentration		(-) *
	Diss. inorg. nitrogen and Total phosphorus ratio		(-) *
	Total Solids in Suspension		(-) *
	Phaeo-pigments concentration in water		(+) *
Shannon-Wiener Diversity Index (H')	Chlorophyll <i>a</i> concentration in sediment	(+) **	
	Phaeo-pigments concentration in sediment	(+) *	
	Dissolved oxygen concentration	(-) *	
	Chlorophyll <i>a</i> concentration in water	(+) *	
	Phaeo-pigments concentration in water	(+) *	
	Pigment diversity index in water	(+) *	
Evenness (J)	Chlorophyll <i>a</i> concentration in sediment	(+) **	
	Phaeo-pigments concentration in sediment	(+) *	
	Chlorophyll <i>a</i> concentration in water	(+) *	
	Salinity	(-) *	
	Total solids in suspension	(-) *	
	Water level in the lagoon	(+) *	
	Cumulative rainfall		(+) *
<i>Chironomus</i> sp. density	Nitrites concentration		(-) *
	Diss. inorg. nitrogen and Total phosphorus ratio		(-) *
	Chlorophyll <i>a</i> concentration in sediment	(+) *	
	Phaeo-pigments concentration in sediment	(+) *	
	Phaeo-pigments concentration in water	(+) *	
	Dissolved oxygen concentration	(-) *	
<i>Lekanesphaera hookeri</i> density	Organic matter content in sediment		(-) *
	Phaeo-pigments percentage in sediment		(-) **
	Clay content		(-) *
	Silt content		(-) *
	Sand content		(+) *
	Sand Mud ratio		(+) *
	Chlorophyll <i>a</i> concentration in sediment	(+) **	
	Phaeo-pigments concentration in sediment	(+) *	
	Salinity	(-) **	
	Water level in the lagoon	(+) **	
	Nitrates concentration	(+) **	
	Nitrites concentration	(+) *	
	Total dissolved inorganic nitrogen concentration	(+) *	
	Diss. inorg. nitrogen and Total phosphorus ratio	(+) *	
Total solids in suspension	(-) *		

The analysis of all *Chironomus* sp. and *Lekanesphaera hookeri* data with environmental parameters from the two lagoons revealed the following linear associations:

- *Chironomus* sp. was positively correlated with chlorophyll *a* concentration in water ( $\rho = 0.674$ ;  $p < 0.001$ ), phaeo-pigments concentration in water ( $\rho = 0.605$ ;  $p < 0.001$ ), total phosphorus concentration ( $\rho = 0.565$ ;  $p < 0.001$ ), orthophosphates concentration ( $\rho = 0.543$ ;  $p = 0.001$ ), chlorophyll *a* concentration in the sediment ( $\rho = 0.486$ ;  $p = 0.002$ ), ammonia concentration ( $\rho = 0.424$ ;  $p = 0.009$ ), phaeo-pigments concentration in the sediment ( $\rho = 0.359$ ;  $p = 0.029$ ); *Chironomus* sp. was negatively correlated with N: P ratio ( $\rho = -0.514$ ;  $p = 0.001$ ) and salinity ( $\rho = -0.439$ ;  $p = 0.007$ ). All correlations were highly significant ( $p \leq 0.01$ ), except for phaeo-pigments concentration in the sediment that was significantly correlated with *Chironomus* sp. ( $0.01 < p \leq 0.05$ ).
- *Lekanesphaera hookeri* was positively correlated with N: P ratio ( $\rho = 0.491$ ;  $p = 0.002$ ) and negatively correlated with total solids in suspension ( $\rho = -0.543$ ;  $p = 0.001$ ), ammonia concentration ( $\rho = -0.447$ ;  $p = 0.006$ ), pH ( $\rho = -0.358$ ;  $p = 0.030$ ) and clay content in the sediment ( $\rho = -0.341$ ;  $p = 0.039$ ). The correlations with pH and clay content in the sediment were significant ( $0.01 < p \leq 0.05$ ), but the strongest and highly significant correlations were the ones with N: P ratio, total solids in suspension and ammonia concentration ( $p \leq 0.01$ ).

The Canonical Correspondence Analysis (CCA) executed with six environmental variables (temperature, total dissolved inorganic nitrogen, total phosphorus, chlorophyll *a* concentration in water, chlorophyll *a* concentration in sediment and clay content), showed that the macroinvertebrate communities in the two lagoons were distinct and that densities of the main taxonomic groups were associated with different environmental variables (Figure 4.23).

The Mollusca Bivalvia (*Abra segmentum*, *Cerastoderma glaucum*) and Gastropoda (*Hydrobia ulvae*, *Ventrosia ventrosa*) occurred only in Foz de Almargem stations and their densities were negatively related to total phosphorus concentration and chlorophyll *a* concentration in water, clay content and chlorophyll *a* concentration in the sediment.

Higher densities of Polychaeta *Capitella capitata* and *Hediste diversicolor* were also associated with lower concentrations of chlorophyll *a* in water and total dissolved inorganic nitrogen, clay content and chlorophyll *a* concentration in the sediment. *Capitella capitata* density was also positively influenced by temperature.

The Amphipoda *Corophium multisetosum* occurred exclusively in Salgados stations and, just as *Chironomus* sp., other Insecta and Oligochaeta, densities were positively related to total phosphorus concentration, clay content and chlorophyll *a* concentration in the sediment, chlorophyll *a* concentration in water and with total dissolved inorganic nitrogen.

The Isopoda *Lekanesphaera hookeri* was placed closer to the centre of the diagram, as it was present in most samples from the two lagoons; however, its density was negatively associated with temperature and positively related with chlorophyll *a* in the sediment and total dissolved inorganic nitrogen concentration.

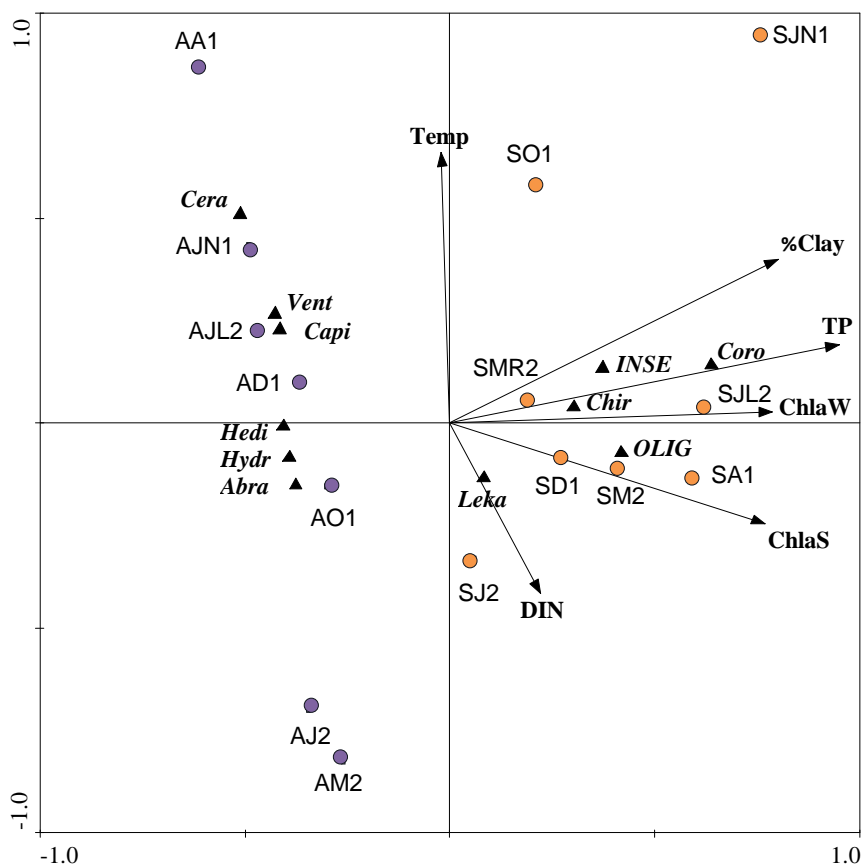


Figure 4.23 - Canonical correspondence analysis performed with the benthic macroinvertebrate groups (mean density) from Foz de Almargem and Salgados lagoon. Cumulative percentage variance explained by axes: Species - I = 48.8 % and I + II = 56.6 %; and Species-environment relation - I = 70.7 % and I + II = 82.0 %. Monte Carlo test of all canonical axes  $p = 0.0018$ .

*Station codes:* First character corresponds to lagoon (S- Salgados, A-Foz de Almargem) and subsequent ones to month and year of survey (1- 2001, 2-2002).

*Environmental variables:* TEMP – water temperature; DIN- Total dissolved inorganic nitrogen concentration; TP- Total phosphorus; ChlaW- Chlorophyll *a* concentration in water; ChlaS- Chlorophyll *a* concentration in sediment; %Clay- Percentage of clay content.

*Benthic macroinvertebrate groups:* Hedi – *Hediste diversicolor*; Capi – *Capitella capitata*; Leka – *Lekanesphaera hookeri*; Coro – *Corophium multisetosum*; Vent – *Ventrosia ventrosa*; Hydr – *Hydrobia ulvae*; Abr – *Abra segmentum*; Cera – *Cerastoderma glaucum*; Chiro – *Chironomus* sp.; INSE – Other Insecta; OLIG – Oligochaeta.

The Monte Carlo permutations test determined significantly high relations ( $p = 0.0018$ ) between the benthic macroinvertebrate groups from the two lagoons and the selected environmental variables.

#### **4.3.3.3. Comparison of benthic communities during isolation and connection of the lagoons with the sea**

Foz de Almargem and Salgados lagoons were isolated from the sea during December 2001 samplings and in January 2002, the two lagoons were in connection with the sea. Thereby, mean data from the benthic macroinvertebrate communities of these two periods were compared.

Total benthos densities presented different tendencies in the two lagoons, increasing in Foz de Almargem and decreasing in Salgados (Table 4.9). The higher density found in Foz de Almargem was due to an augment in the Gastropoda *Hydrobia ulvae* and the Isopoda *Lekanesphaera hookeri*, while in Salgados lagoon, the major cause for the reduction of total benthos density was the diminution of the Insecta *Chironomus* sp.

During the period of connection with the sea, the benthic macroinvertebrate community in Foz de Almargem was composed by five *taxa*, the Gastropoda *Hydrobia ulvae* (95.10% of total benthos in the lagoon), the Isopoda *Lekanesphaera hookeri* (4.17%), the Insecta *Chironomus* sp. (0.38%), the Polychaeta *Hediste diversicolor* (0.32%) and the Bivalvia *Abra segmentum* (0.03%). In December, three other *taxa* had been registered (Polychaeta *Capitella capitata*; Gastropoda *Ventrosia ventrosa*; Bivalvia *Cerastoderma glaucum*), but were not observed in January.

The benthic macroinvertebrate community of Salgados in January presented seven *taxa*, most of them Insecta (*Chironomus* sp. – 53.16%; Ceratopogonidae – 8.89%; *Berosus spinosus* – 4.52%; Ephydriidae – 2.26%; Tabanidae – 2.26%), which accounted 71.08 % of the benthos density in the lagoon; Oligochaeta Enchytraeidae (26.66%) and the Isopoda *Lekanesphaera hookeri* (2.26%). Four of these *taxa* were absent in December (Insecta Ceratopogonidae and Tabanidae; Oligochaeta Enchytraeidae; Isopoda *Lekanesphaera hookeri*) and one other that was present in December was not registered when the lagoon was opened (Insecta Brachyceres).

Table 4.9 – Benthic macroinvertebrate communities in January 2002 and variation between mean values when the lagoons were isolated (December 2001) and connected to the sea (January 2002).

	<i>Foz de Almargem lagoon</i>		<i>Salgados lagoon</i>	
	Jan-02	Variation	Jan-02	Variation
<b>Benthic macroinvertebrate communities</b>				
Total benthos density (ind m <sup>-2</sup> )	33717	<b>17168</b>	664	<b>-2920</b>
Taxonomic richness	5	<b>-3</b>	7	<b>3</b>
Shannon-Wiener diversity (bits)	0.59	<b>-0.11</b>	1.68	<b>1.17</b>
Evenness	0.37	<b>0.10</b>	0.84	<b>0.56</b>
<b>Total INSECTA density (ind m<sup>-2</sup>)</b>	<i>128</i>	<b>-39</b>	<i>472</i>	<b>-3112</b>
<i>Chironomus</i> sp. density (ind m <sup>-2</sup> )	128	<b>-39</b>	353	<b>-2980</b>
Ephydriidae density (ind m <sup>-2</sup> )	0	<b>0</b>	15	<b>-103</b>
Tabanidae (ind m <sup>-2</sup> )	0	<b>0</b>	15	<b>15</b>
Ceratopogonidae (ind m <sup>-2</sup> )	0	<b>0</b>	59	<b>59</b>
Empididae density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Rhagionidae density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Brachyceres density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>-29</b>
<i>Berosus spinosus</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	30	<b>-74</b>
<i>Hesperocorixa sahlbergi</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Corixa affinis</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Parasigara infuscata</i> density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<i>Notonecta</i> sp. density (ind m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<b>Total POLYCHAETA density (ind m<sup>-2</sup>)</b>	<i>108</i>	<b>-157</b>	<i>0</i>	<b>0</b>
<i>Capitella capitata</i> density (ind m <sup>-2</sup> )	0	<b>-69</b>	0	<b>0</b>
<i>Hediste diversicolor</i> density (ind m <sup>-2</sup> )	108	<b>-88</b>	0	<b>0</b>
<b>Total OLIGOCHAETA density (ind m<sup>-2</sup>)</b>	<i>0</i>	<b>0</b>	<i>177</i>	<b>177</b>
Tubificidae density (inds m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
Enchytraeidae density (ind m <sup>-2</sup> )	0	<b>0</b>	177	<b>177</b>
<b>Total ISOPODA density (ind m<sup>-2</sup>)</b>	<i>1406</i>	<b>728</b>	<i>15</i>	<b>15</b>
<i>Lekanesphaera hookeri</i> density (ind m <sup>-2</sup> )	1406	<b>728</b>	15	<b>15</b>
<b>AMPHIPODA density (inds m<sup>-2</sup>)</b>	<i>0</i>	<b>0</b>	<i>0</i>	<b>0</b>
<i>Corophium multisetosum</i> density (inds m <sup>-2</sup> )	0	<b>0</b>	0	<b>0</b>
<b>Total GASTROPODA density (ind m<sup>-2</sup>)</b>	<i>32065</i>	<b>16765</b>	<i>0</i>	<b>0</b>
<i>Ventrosia ventrosa</i> density (ind m <sup>-2</sup> )	0	<b>-747</b>	0	<b>0</b>
<i>Hydrobia ulvae</i> density (ind m <sup>-2</sup> )	32065	<b>17512</b>	0	<b>0</b>
<b>Total BIVALVIA density (ind m<sup>-2</sup>)</b>	<i>10</i>	<b>-129</b>	<i>0</i>	<b>0</b>
<i>Abra segmentum</i> density (ind m <sup>-2</sup> )	10	<b>-61</b>	0	<b>0</b>
<i>Cerastoderma glaucum</i> density (ind m <sup>-2</sup> )	0	<b>-68</b>	0	<b>0</b>
<b>TROPHIC GROUPS DENSITIES</b>				
Suspension feeders (ind m <sup>-2</sup> )	0	<b>-69</b>	0	<b>0</b>
Deposit feeders (ind m <sup>-2</sup> )	32193	<b>16657</b>	546	<b>-2906</b>
Suspension/ deposit feeders (ind m <sup>-2</sup> )	10	<b>-59</b>	0	<b>0</b>
Herbivores (ind m <sup>-2</sup> )	1406	<b>728</b>	15	<b>15</b>
Carnivorous/ scavengers/ omnivorous (ind m <sup>-2</sup> )	108	<b>-88</b>	103	<b>-29</b>
<b>AMBI GROUPS DENSITIES</b>				
Group III (ind m <sup>-2</sup> )	33717	<b>17237</b>	369	<b>-2965</b>
Group IV (ind m <sup>-2</sup> )	0	<b>0</b>	88	<b>-59</b>
Group V (ind m <sup>-2</sup> )	0	<b>-69</b>	177	<b>177</b>
Not assigned	0	<b>0</b>	29	<b>-74</b>

Shannon-Wiener diversity decreased in Foz de Almagem and increased in Salgados, while evenness increased in both lagoons. Salgados lagoon presented higher values for both indices during the period of connection with the sea.

When the lagoons were connected to the sea, there was a reduction in the mean densities of carnivorous/ scavengers/ omnivorous in the two lagoons. In Foz de Almagem, suspension feeders and suspension/deposit feeders also diminished, while deposit feeders and herbivores augmented. Herbivores increased in Salgados lagoon too, but deposit feeders decreased. Trophic groups' densities variation in Foz de Almagem was greater than in Salgados lagoon.

To what concerns ecological groups, Foz de Almagem showed a reduction in group V *taxa* densities (first-order opportunistic *taxa*, indicators of pronounced unbalanced situations) and an augment in the densities of *taxa* tolerant to excess organic matter enrichment (Group III-AMBI), which were the only *taxa* observed in the lagoon, during January 2002. In Salgados lagoon, the only group that presented an increase in densities was group V. All other ecological groups' densities registered a decrease, when the lagoon was opened to the sea. The greatest variation was determined in group III, although it was the group with higher density.

#### 4.3.3.4. Benthic macroinvertebrate community's preference/tolerance to salinity

Benthic macroinvertebrate community in Foz de Almagem lagoon was mainly composed by brackish *taxa* (4 *taxa*; 40%) and eurihaline *taxa* (6 *taxa*; 60%) (Table 4.10).

Table 4.10 - Classification of benthic macroinvertebrate *taxa* based on the salinity preference/tolerance in Foz de Almagem and Salgados lagoons. *Salinity conditions*: OM- Oligohaline and Mesohaline; M- Mesohaline; OMP- Oligohaline, Mesohaline and Polyhaline, MPE- Mesohaline, Polyhaline and Euhaline; OMPE- Oligohaline, Mesohaline, Polyhaline and Euhaline.

<i>Lagoon</i>	<i>Brackish (OM)</i>	<i>Brackish (M)</i>	<i>Brackish (MP)</i>	<i>Brackish (OMP)</i>	<i>Eurihaline Marine (MPE)</i>	<i>Eurihaline Marine (OMPE)</i>
Foz de Almagem			<i>C. capitata</i> <i>S. vermicularis</i> UI.Polychaeta	<i>Chironomus</i> sp.	<i>C. glaucum</i>	<i>H. diversicolor</i> <i>L. hookeri</i> <i>V. ventrosa</i> <i>H. ulvae</i> <i>A. segmentum</i>
Salgados	Ephyrididae Ceratopogonidae Brachyceres <i>B. spinosus</i> <i>C. affinis</i> <i>P. infuscata</i> Tubificidae	Empididae Rhagionidae <i>Notonecta</i> sp. <i>C. multisetosum</i>	Enchytraeidae	<i>Chironomus</i> sp. Tabanidae <i>H. sahlbergi</i> <i>L. hookeri</i>		

In Salgados lagoon all *taxa* were brackish, although most of them (7 *taxa*; 44%) occurred in oligohaline and mesohaline conditions (OM) or just in mesohaline conditions (4 *taxa*; 25%). Just one *taxon* (6%) was present in mesohaline and polyhaline conditions, but four *taxa* (25%) were observed during periods with oligohaline, mesohaline and polyhaline conditions.

During the studied period, eurihaline marine *taxa* was the category with greatest richness and higher densities in Foz de Almagem lagoon (Figure 4.24).

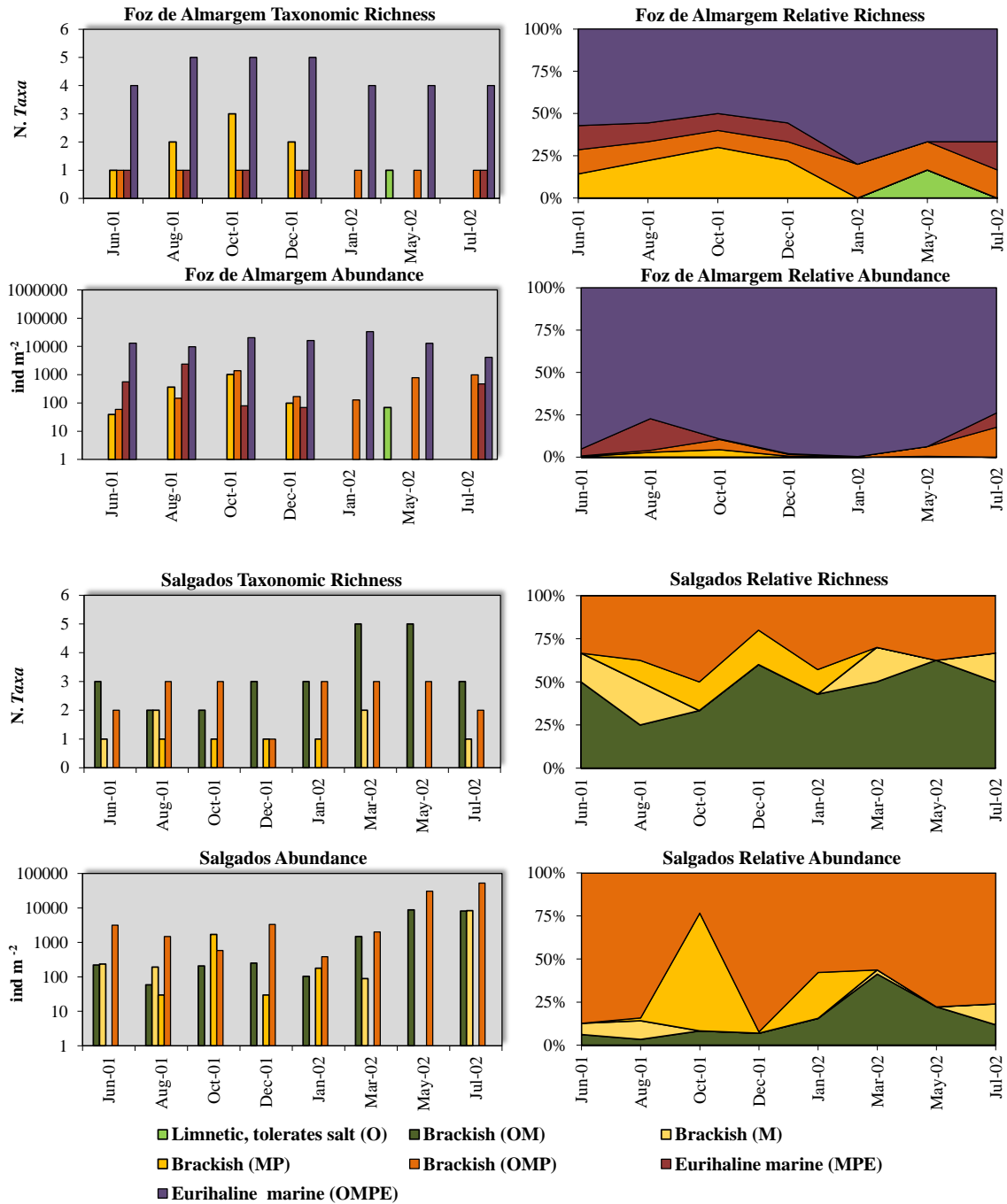


Figure 4.24 – Evolution of taxonomic richness and density of benthic macroinvertebrate *taxa* according to salinity preferences in Foz de Almagem and Salgados lagoons.

In Salgados lagoon, brackish *taxa* (oligohaline-mesohaline) presented a taxonomic richness superior than other *taxa* categories most of the time, except in August and October 2001 when brackish *taxa* (oligohaline-mesohaline-polyhaline) had a greater richness; and in January 2002, month during which the two categories presented the same number of *taxa*.

In terms of density, brackish *taxa* (oligohaline-mesohaline-polyhaline) showed the highest values during most of the studied period, with the exception of October 2001, when brackish *taxa* (mesohaline-polyhaline) density was superior.

## 5. General discussion and conclusions

### 5.1. Hydrological aspects

Foz de Almagem and Salgados lagoons showed some differences in their hydrological regimens, which were mainly related to the sources of freshwater, the date and process of lagoon opening, the morphology and the geographic location of the lagoons.

Freshwater inputs in Foz de Almagem were determined by rainfall and river runoff. When the lagoon was isolated from the sea, water level resulted from the balance between freshwater inputs and water evaporation. In Salgados lagoon, besides the natural freshwater inputs, water level was also influenced by the volume of wastewater discharged, which was particularly relevant during the dry season, keeping water in the lagoon in a medium level (Soares, 2000).

With the start of the raining season, water level increased and several openings occurred in the lagoons. Salgados lagoon was opened five times and in Foz de Almagem there were four openings, but the total number of days Foz de Almagem lagoon was in connection with the sea almost doubled the period of connection in Salgados. The shorter or longer time lagoons maintained the connection with the sea depended on different factors, namely wave energy and direction, rainfall and river runoff, water level when the channel was opened and tides. Longer periods of connection are favored by waves with low energy, which accumulate less sand in the channel; greater rainfall and river runoff, that keep a permanent flow into the sea; high water level in the lagoon when the barrier is opened, which allows the formation of a larger and deeper channel; and tides with low range (neap tides), that have a smaller effect on sediment deposition in the channel (Pinto *et al.*, 2001)

Besides the greater rainfall in Foz de Almagem, two other factors might have contributed to the longer period of connection with the sea and that was the smaller dimension of the lagoon and its location in a coastal area with great erosion. The lower capacity of water retention by the lagoon associated with greater rainfall and river runoff facilitated the connection with the sea. There is also the fact that, Foz de

Almargem is located in one of the areas of Algarve with highest rate of coastal erosion (Teixeira, 2009) and during winter, storms removed great part of the sand from the beach and easily destroyed the sand barrier.

Salgados lagoon has a greater capacity of water retention due to its bigger surface and is located in a coastal area (Armação de Pêra bay) which tends to accumulate sediments from the sea and does not suffer from significant coastal erosion (Teixeira, 2009). Thereby, the formation of the sand barrier and closing of the lagoon would take less time than in Foz de Almargem.

The artificial openings in Foz de Almargem were illegally done by local fishermen with the purpose of catching fish in the channel and in the mouth of the lagoon, while in Salgados, artificial openings were performed by the regional environmental services to promote water renewal and prevent flooding in the golf course nearby. The natural openings in both lagoons happened after artificial interventions, when the sand barriers were thinner and more exposed to the sea.

When the lagoons were in connection with the sea, water renewal was depended not just on freshwater inputs but also on the strength and height of tides. In many occasions, the sea water influence was noticed only in the downstream and intermediate stations, and water was flowing just in the deepest channels of the lagoons, leaving a great extension of sediment emerged. Sea water inflow was greater in Foz de Almargem, nevertheless the shallowest areas of the lagoon were not submerged most of the times.

## **5.2. Water parameters**

The comparison of Foz de Almargem and Salgados monthly mean values of water parameters showed that the two lagoons had distinct characteristics, particularly in orthophosphates concentration, total phosphorus, chlorophyll *a* concentration, ammonia, pigments diversity, N:P ratio and total solids in suspension. Both lagoons presented a seasonal variation that was mainly defined by temperature, dissolved oxygen concentration and pH.

Some physical and chemical water parameters have reference values accordingly to European Directives concerning the water quality for different purposes (e.g. 74/440/EEC; 76/464/EEC; 78/659/EEC; 80/68/EEC; 98/15/EC), namely nitrogen compounds (DIN  $< 15 \text{ mg L}^{-1} \text{ N} \approx 1072 \text{ } \mu\text{M N}$ ; Nitrates  $< 25 \text{ mg L}^{-1} \text{ NO}_3 \approx 403 \text{ } \mu\text{M N}$ ; Ammonia  $< 1 \text{ mg L}^{-1} \text{ N} \approx 71 \text{ } \mu\text{M N}$ ), total phosphorus concentration ( $< 1 \text{ mg L}^{-1} \text{ P} \approx 32 \text{ } \mu\text{M P}$ ), dissolved oxygen concentration ( $> 5 \text{ mg O}_2 \text{ L}^{-1}$ ), pH (5.0-9.0), total solids in suspension ( $< 35 \text{ mg L}^{-1}$ ).

In Foz de Almargem, just the pH (August 2001) and the total solids in suspension (June to December 2001) presented values superior to the recommended intervals. The pH value of 9.45 found in August 2001 might have been caused by an occasional discharge of domestic detergents from houses in the vicinity of the lagoon, and it coincided with an increase of phosphorus concentrations in the lagoon. The high values of total solids in suspension measured in October 2001 were probably due to the transportation of a big amount of particulate matter by the river into the lagoon associated with the first rains of the season.

Besides the occasional discharges of domestic effluents, no other direct sources of organic pollution were detected in Foz de Almargem. The main source of nitrates in Foz de Almargem was the runoff from agriculture lands and eventually the high concentrations of nitrates in groundwater, as the lagoon aquifer (Quarteira aquifer) in 2001 and 2002 registered concentrations approximate to  $50 \text{ mg L}^{-1}$  (DRAOT-ALGARVE, 2001; 2002). In addition to agriculture runoff, in Salgados lagoon the runoff from the golf course nearby might also have contributed as a source of nitrates.

Some of the water parameters in Salgados lagoon registered values outside the intervals recommended by European Directives concerning the water quality.

Dissolved oxygen concentration showed values lower than  $5 \text{ mg L}^{-1}$  at the intermediate station in June 2001 and in May 2002, during the 24 hours monitoring cycle (from 10 p.m. till 8 a.m.). These concentrations were measured at the surface and the minimum value of  $1.97 \text{ mg L}^{-1}$  (6 a.m., May 2002) suggests that near the bottom and in the deepest part of the lagoon, anoxia conditions might have occurred. The great fluctuations observed during the daily cycle are usually related to high abundance of phytoplankton and organic matter, characteristic of eutrophic waters (Bernardo, 1990; Conte de Barros, 1996; Cravo, 2003). The pH also presented a great variation in the intermediate station during the 24 hours monitoring, but values remained inside the

recommended interval (5.0-9.0). These oscillations can be explained by the intense absorption of carbon dioxide during the day in photosynthesis and its release during the night due to autotrophic organisms and bacteria respiration (Cortes *et al.*, 1991; Cravo, 2003). In December 2001 and in March 2002 (intermediate station), pH values went above 9.0, somehow coinciding with dissolved oxygen concentration peaks.

Salgados ammonia concentrations in August 2001 reached 486.11  $\mu\text{M N}$  in the intermediate station and 410.56  $\mu\text{M N}$  downstream, values that are far from the maximum recommended 71  $\mu\text{M N}$ . These concentrations might have been caused by liberation of ammonia from the sediments into the water column, in situation of anoxia near the bottom (Cancela da Fonseca, 1989; Falcão, 1996), but also by discharges from the wastewater treatment plant. Neves (1999) described high concentrations of ammonia during the summer in Espiche river, associated to discharges of effluents that had not been properly treated.

Total phosphorus concentrations in Salgados lagoon were very high all over the year and just in a few occasions (December 2001, January 2002) values were under the recommended value of 32  $\mu\text{M P}$ . Orthophosphates concentrations represented almost all total phosphorus and Neves (1999), also found high concentrations of this compound near the effluent of the wastewater treatment plant, concluding that this was the main source of orthophosphate in the lagoon. Nevertheless, the greatest concentrations determined during the summer months (Intermediate station: June 2001 – 161.29  $\mu\text{M P}$ ; August 2001 – 158.70  $\mu\text{M P}$ ; July 2002 – 100.32  $\mu\text{M P}$ ), might also have been caused by orthophosphate release from the sediment in situations of anoxia (Cancela da Fonseca, 1989; Welch, 1992; Falcão, 1996;), just as it happened with ammonia. Thereby, the internal recycling of orthophosphate from the sediment inside the lagoon also constitutes an important source of phosphorus to the water (Welch, 1992).

In Salgados lagoon, total solids in suspension presented concentrations within the recommended values ( $< 35 \text{ mg L}^{-1}$ ), only from March to July 2002. Most of the time and particularly during the summer, concentrations were very high (maximum 260  $\text{mg L}^{-1}$ ). It might have been related to inefficient removal of solids from wastewater effluents and the discharge of high abundance of microalgae that grow in the maturation tanks of the plant (Cravo, 2003).

In Ria Formosa (South Portugal), a coastal lagoon with permanent tidal influence and water renewal, Cravo (2003) registered high values for some of the water parameters, near the wastewater discharge channels (*e.g.* TSS: 711.1 mg L<sup>-1</sup>; Ammonia: 257.5 µM N; Nitrates: 112.9 µM N; Orthophosphates: 97.6 µM P; N/P ratio: 129.0). In other areas affected by wastewater, Welch (1992) refers to the increase of the above parameters and also of pH, total dissolved inorganic nitrogen and total phosphorus concentrations.

### **5.3. Phytoplankton**

#### **5.3.1. Chlorophyll *a* concentration**

Chlorophyll *a* annual mean concentration in Foz de Almargem (3.0 µg L<sup>-1</sup>) was below the mean values found by Bernardo (1990) in Santo André coastal lagoon in 1984 (27.8 µg L<sup>-1</sup>) and 1985 (5.5 µg L<sup>-1</sup>). Also the monthly mean chlorophyll *a* variation in Foz de Almargem (0.38 – 9.68 µg L<sup>-1</sup>) was lower than the variation determined for Santo André coastal lagoon by Cancela da Fonseca *et al.* (1989) (1.8 – 61.9 µg L<sup>-1</sup>) and by Macedo *et al.* (2001) (0.2 - 52 µg L<sup>-1</sup>).

In Salgados lagoon, chlorophyll *a* annual mean concentration was much higher (158.5 µg L<sup>-1</sup>) and there was a greater oscillation in concentrations during the studied months (19.34 – 368.69 µg L<sup>-1</sup>). In the surrounding areas of wastewater discharge channels, Cravo (2003) determined concentrations of chlorophyll *a* (378.2 µg L<sup>-1</sup>) similar to the maximum mean value found in Salgados lagoon.

#### **5.3.2. Phytoplankton communities**

Phytoplankton mean abundances determined in Foz de Almargem were much lower than the ones described for other coastal lagoons, namely Quinta do Lago in Ria Formosa (Morais *et al.*, 2003); Tancada, Encañizada and Buda coastal lagoons from the Ebro Delta (Comin, 1982). Phytoplankton growth might have been limited by phosphorus concentrations in winter and spring as the N:P ratio was greater than the Redfield ratio 16:1. This is a rare situation, also found in Ria Formosa coastal lagoon, as

nitrogen is usually the limiting nutrient in temperate lagoons (Newton *et al.*, 2003; Newton and Mudge, 2005).

Phytoplankton mean abundances determined in Salgados lagoon were similar or higher than those described for coastal lagoons where phytoplankton blooms occurred associated to eutrophication phenomena, such as in Quinta do Lago - Ria Formosa (Morais *et al.*, 2003), wastewater discharge channels in Ria Formosa (Cravo, 2003) and the lagoon of Venice (Bianchi *et al.*, 2003).

Phytoplankton communities in Foz de Almargem presented seasonal and spatial variations that were related with some of the environmental parameters studied. Dinophyceae and Bacillariophyceae were present in all samples, being associated to the general conditions in the lagoon. Euglenophyceae and Chlorophyceae occurred in stations and months with lower DIN, salinity, total phosphorus and higher TSS; Cryptophyceae densities increased with higher values of salinity, total phosphorus, TSS and low water level; Cyanophyceae greatest densities coincided with higher DIN, salinity, total phosphorus and lower TSS; Pico-nano flagellate algae were positively associated with salinity and total phosphorus and negatively with water level.

Wetzel (1983) considered that Dinophyceae and Bacillariophyceae dominance indicate oligo-mesotrophic and mesotrophic conditions, respectively. Nevertheless, most of the Dinophyceae that occurred in the Foz de Almargem lagoon with higher abundances are potentially toxic, *e.g.* *Prorocentrum minimum*, *Prorocentrum micans*, *Gymnodinium* sp. and *Protoperidinium* sp. (Faust and Gullede, 2002). Pico-nano flagellate algae were relatively important mainly upstream and in the intermediate station, just after the periods with low dominance of Bacillariophyceae and Dinophyceae.

Phytoplankton assemblages in coastal waters tend to be dominated by Bacillariophyceae (diatoms), with secondary contributions from Dinophyceae (dinoflagellates), flagellates and Cyanophyceae (Cloern *et al.*, 1985; Jarry *et al.*, 1990; Nuccio *et al.*, 2003; Badylak and Philips, 2004).

Although other factors as temperature or water stratification can be relevant in determining the succession of dominant phytoplankton species, the inversion in the dominance of the major phytoplankton taxonomic groups is usually related to

anthropogenic eutrophication (Balkis, 2003; Hashimoto and Nakano, 2003). The opportunistic dinoflagellates usually over dominate the diatoms in periods of nutrient enrichment and low water turbulence. Furthermore, small phytoplankton, as flagellate algae, should out compete large phytoplankton when nutrient are scarce, while larger phytoplankton, as diatoms and Dinophyceae, should out compete small phytoplankton when nutrient level increases (Pérez-Ruzafa *et al.*, 2002).

Most of the time, Cyanophyceae dominated the phytoplankton communities in Salgados lagoon, with a few exceptions. Bacillariophyceae became the main taxonomic group in December 2001; Chlorophyceae was the major class upstream in June 2001 and downstream in July 2002; pico-nano flagellate algae accounted a high percentage of the intermediate and downstream phytoplankton in March and May 2002.

In Ria Formosa, phytoplankton communities from the wastewater discharge channels were also dominated by Cyanophyceae (Cravo, 2003).

In Salgados lagoon, the seasonal succession of phytoplankton communities showed some similarities with the pattern of eutrophic and hypertrophic freshwater systems. A characteristic pattern of seasonal succession in freshwater systems from temperate regions is, for example, Bacillariophyceae in association with rapidly growing small flagellates in winter and spring, followed by Chlorophyceae in late spring and early summer, and then by species which cannot easily be eaten by zooplankton, such as Dinophyceae, desmids, large yellow-green algae and diatoms in late summer and autumn (Chorus and Bartram, 1999). In eutrophic and hypertrophic waters, Cyanophyceae often dominate the summer phytoplankton (Järvinen *et al.*, 2013, and references there in); as winter approaches, increasing turbulence and the lack of light leads to their replacement by diatoms (Chorus and Bartram, 1999). According to Wetzel (1983), the dominance of Cyanophyceae, Bacillariophyceae and Chlorophyceae indicates eutrophic and hyper-eutrophic conditions.

### **5.3.3. Potentially harmful phytoplankton**

In both lagoons were identified *taxa* that can be potentially harmful due to the production of biotoxins. Most of the potentially harmful *taxa* presented significant

correlations with orthophosphates, total phosphorus or nitrogen compounds concentrations.

In Foz de Almargem, three of the four potentially harmful *taxa* belong to Dinophyceae (*Gymnodinium* sp., *Prorocentrum minimum*, *Scrippsiella trochoidea*) and the fourth *taxon* is a Cyanophyceae (*Anabaena flos-aqua*). *Gymnodinium* sp. and *Prorocentrum minimum* were the *taxa* with higher mean abundances and a more regular presence in Foz de Almargem lagoon.

In Salgados lagoon, the number of potentially harmful *taxa* (six) was greater than in Foz de Almargem and most of the *taxa* presented higher mean abundances. Just one *taxon* belonged to Dinophyceae (*Gymnodinium* sp.) and the remaining *taxa* were Cyanophyceae (*Anabaena flos-aqua*, *Anabaena spiroides*, *Lyngbya* sp., *Microcystis aeruginosa*, *Planktothrix* sp.).

During March 2002, a bloom of *Prorocentrum minimum* (Dinophyceae) occurred in Foz de Almargem, being responsible for the peak observed in phytoplankton abundance, at the same time that the Shannon-Wiener diversity and the evenness indexes were minimum. Similar situations have been described in Santo André coastal lagoon, where *Prorocentrum minimum* contributed more than 90% to the total number of phytoplankton cells counted during bloom events (Macedo *et al.*, 2001). However, the frequency of blooms caused by this species and the phytoplankton abundance in Santo André was much higher than in Foz de Almargem.

*Prorocentrum minimum* is an armoured marine dinoflagellate, with a cosmopolitan distribution in temperate brackish waters to tropical regions (Hajdu *et al.*, 2005). High densities of this species have been reported in coastal waters, particularly in estuaries, causing a change in water colour to brown and causing shellfish poisoning due to the production of a hepatotoxine, venerupin (Witek and Plínski, 2000; Faust and Gullledge, 2002). Although some clones of *Prorocentrum minimum* can produce toxic blooms, this species is not considered to be persistently toxic (Hallegraeff *et al.*, 1995). Effects on organisms were identified at concentrations as low as  $3 \times 10^6$  cells L<sup>-1</sup> (EPA, 2003) providing a threshold for tracking and assessing *Prorocentrum minimum* blooms. In Foz de Almargem, *Prorocentrum minimum* peak abundance did not reach the threshold value and no evidences of toxicity were found in the lagoon. However, a special

attention should be given to this species and to the potential factors that promote its growth, in order to prevent harmful impacts on the ecosystem.

Potentially harmful *taxa* were observed in Salgados lagoon during most of the sampling periods, forming blooms and accounting to a considerable percentage of total phytoplankton abundance, particularly during the summer months. Wastewater treatment plants may hold Cyanophyceae populations with toxic production, which are responsible for changes in the microbial dynamics of the waste water treatment plant leading to lower efficiencies on organic matter metabolization, and which are also associated with the contamination of sites located downstream (Vasconcelos and Pereira, 2001). Abundances of potentially harmful *taxa* in Salgados were very high during summer months, when the ecological quality of the lagoon deteriorates even more due to the tourism avalanche and difficulties of the sewage treatment plants to cope with the sudden increase of inhabitants.

The increase of nutrient pollution promotes the development and persistence of many harmful algae blooms (Heisler *et al.*, 2008) and the occurrence of these blooms can cause severe changes in water quality and deeply affect the whole ecosystem. Some of the main effects associated to (potentially) harmful phytoplankton blooms comprise the decrease in water transparency, the great fluctuation of oxygen concentration and the release of toxic compounds (Vasconcelos, 2006). During blooms senescence and phytoplankton cells breakdown, toxins can be released into the water causing, not only the death of aquatic organisms, livestock, waterfowl and domestic animals, but also health problems to people who get in contact with this water (Vasconcelos, 2006). In fact, massive fish and bird kills have been reported to occur in Salgados lagoon with relative frequency.

#### **5.3.4. Phytoplankton salinity tolerance**

Concerning phytoplankton salinity tolerance, the proportion of each category differed in the two lagoons, reflecting the greatest or smallest influence of freshwater and seawater inputs.

Most of the phytoplankton *taxa* identified in Foz de Almargem lagoon were characteristic from brackish/marine *habitats*, freshwater/ brackish *habitats* and freshwater/brackish/marine *habitats*. Freshwater *taxa* were the less represented.

The potentially harmful Dinophyceae *P. minimum* and *S. trochoidea* usually occur in brackish and marine *habitats*, whereas *Gymnodinium* may inhabit freshwater, brackish and marine environments (Sournia, 1986). The only potentially harmful Cyanophyceae that was identified in Foz de Almargem (*A. flos-aqua*) has freshwater affinities (Landsberg, 2002; Cronberg and Haecky, 2006).

Phytoplankton communities in Salgados lagoon were mainly composed by freshwater *taxa* and freshwater/brackish *taxa*. Freshwater/brackish/marine *taxa* were less represented and no *taxa* from brackish/marine *habitats* were found in the lagoon.

Potentially harmful *taxa* identified in Salgados lagoon were mostly Cyanophyceae typical from freshwater *habitats* (*M. aeruginosa*, *A. flos-aqua*, *A. spiroides*, *Planktothrix* sp.) or freshwater and brackish *habitats* (*Lyngbya* sp.) (Landsberg, 2002; Cronberg and Haecky, 2006). The dinoflagellate *Gymnodinium* sp. is the only *taxon* that is found in marine *habitats* (Sournia, 1986), but presented lower abundances than other *taxa*..

#### **5.4. Sediment parameters**

During the studied period, the most relevant sediment parameters that explained the monthly variability observed in the two lagoons were phaeo-pigments concentration, chlorophyll *a* concentration, organic matter content, water content and clay content. On the other hand, Margalef's pigment diversity and chlorophyll *a* degradation index seemed to be the main parameters to separate Foz de Almargem from Salgados sediments. Parameters as phaeo-pigments concentration and organic matter content, silt and clay content, Margalef's pigment diversity and chlorophyll *a* degradation index, were positively correlated.

Monthly mean values of organic matter content in both lagoons (Foz de Almargem: 1.8 % - 4.6 %; Salgados: 1.4 % - 6.8 %) were lower than the interval found in Santo

André coastal lagoon (6.5 % - 16.6 %) (Cancela da Fonseca, 1989). The same happened with chlorophyll *a*, with the monthly mean values in the lagoons (Foz de Almargem: 2.16 – 16.80  $\mu\text{g g}^{-1}$ ; Salgados: 3.37 – 28.15  $\mu\text{g g}^{-1}$ ) oscillating between the average values determined in sands (2.18  $\mu\text{g g}^{-1}$ ) and in muds (46.26  $\mu\text{g g}^{-1}$ ) from Santo André lagoon. Phaeo-pigments concentrations in Foz de Almargem (2.07 – 18.46  $\mu\text{g g}^{-1}$ ) and in Salgados (3.66 – 29.59  $\mu\text{g g}^{-1}$ ) varied in a similar range as chlorophyll *a* concentrations, not being considerably higher (three times greater) as it happened in Santo André (Cancela da Fonseca *et al.*, 1987; Cancela da Fonseca, 1989). This author described a strong correlation between Margalef's pigment diversity and chlorophyll *a* degradation percentage, suggesting that higher values of Margalef's pigment diversity indicate that the microphytobenthic communities are less productive and show greater degradation.

Foz de Almargem presented annual mean values of clay content, silt content, chlorophyll *a* and phaeo-pigments concentrations lower than Salgados and higher means of sand content, water content, chlorophyll *a* degradation index and Margalef's pigments diversity. Statistical analyses indicated that, for most sediment parameters differences between lagoons were not significant. Only chlorophyll *a* concentration and Margalef's pigments diversity showed evidences of significant differences in the two lagoons.

The global analysis of water and sediment parameters along the studied period revealed that there was a differentiation between the two lagoons mainly associated to clay content and chlorophyll *a* in the sediment, orthophosphates concentrations in water and organic matter content in the sediment. Generally, these parameters presented higher values in Salgados lagoon than in Foz de Almargem. Water parameters such as Margalef's pigments diversity, N:P ratio, chlorophyll *a* and dissolved oxygen were also relevant to understand that Salgados lagoon samples presented a certain pattern (lower pigments diversity and N:P ratio; higher chlorophyll *a* and dissolved oxygen), which did not change much during the studied period, while in Foz de Almargem lagoon there was a greater temporal variation, according to these parameters. Temperature was not so important to the characterization of the lagoons, but it was positively correlated to organic matter content and phaeo-pigments concentration in the sediment.

The assessment of chlorophyll *a* in the water ( $\text{mg m}^{-2}$ ) and chlorophyll *a* in the sediment ( $\text{mg m}^{-2}$ ) in the two lagoons revealed that in Foz de Almargem the amount of benthic chlorophyll was greater than chlorophyll in the water column, while in Salgados lagoon it was observed the inverse situation. Just in June and October 2001, chlorophyll *a* in Salgados sediment presented higher values than chlorophyll *a* in the water.

In Foz de Almargem, chlorophyll *a* in the sediment accounted 77.3% to 97.3% of total chlorophyll *a* in the lagoon, whereas in Salgados lagoon it represented 8.1% to 80.9% of total chlorophyll *a*. Although the annual mean value in Salgados lagoon ( $44.33 \text{ mg m}^{-2}$ ) was higher than in Foz de Almargem ( $20.65 \text{ mg m}^{-2}$ ), the percentage of chlorophyll *a* in Salgados sediment (30.6%) was lower than in Foz de Almargem sediment (89.3%), relative to total chlorophyll *a* in the lagoons. Cancela da Fonseca (1989) found higher values in Santo André lagoon (sand:  $53.6 \text{ mg m}^{-2}$ ; mud:  $256.6 \text{ mg m}^{-2}$ ), just as Brito *et al.* (2010) for Ria Formosa coastal lagoon ( $269 \text{ mg m}^{-2}$ ;  $\approx 99\%$  of total microalgal chlorophyll of the lagoon). Nevertheless, the percentages of chlorophyll *a* in the sediment determined in Foz de Almargem and during a few months in Salgados confirm the importance of microphytobenthos in the total microalgal chlorophyll of the lagoons, particularly in Foz de Almargem. Microphytobenthos play a key role in the interactions between sediments and the water column, as they uptake nutrients that otherwise would go to the water column and when high proportions occur in relation to pelagic chlorophyll, their influence in the water column by re-suspension is likely to be large (Brito *et al.*, 2010).

## **5.5. Benthic macroinvertebrates**

### **5.5.1. Communities structure**

Benthic macroinvertebrate communities in the two lagoons were distinct, just as their relations with environmental parameters. The number of *taxa* identified in Foz de Almargem (10 *taxa*) was lower than and the number determined for Salgados lagoon (16 *taxa*). Both lagoons presented low richness compared to other Portuguese coastal

lagoons such as Santo André lagoon (42 *taxa*) (Correia *et al.*, 2012) and Óbidos lagoon (125 *taxa*) (Cancela da Fonseca *et al.*, 2006).

Foz de Almargem and Salgados lagoons were dominated by different groups of benthic macroinvertebrates, with Mollusca being the most abundant group in Foz de Almargem, while Insecta and Crustacea were the more relevant groups in Salgados lagoon.

Total macroinvertebrate densities in the lagoons showed a different evolution along time. Monthly mean values determined in Foz de Almargem were higher than the ones obtained in Salgados lagoon, except in May and July 2002. In Foz de Almargem, the highest mean density was 33717 ind m<sup>-2</sup> (January 2002) and the lowest was 5585 ind m<sup>-2</sup> (July 2002). Salgados lagoon presented the lowest mean density in January 2002 (664 ind m<sup>-2</sup>), whereas the higher value occurred in July 2002 (68864 ind m<sup>-2</sup>).

In Santo André coastal lagoon, Cancela da Fonseca (1989) found mean densities that oscillated between 1000 and 3000 ind m<sup>-2</sup> (1978/1979), but in January 1984 there was an increase up to about 70000 ind m<sup>-2</sup>, that coincided with a period of eutrophication.

Mean densities determined in Foz de Almargem were higher than the ones found in Albufeira coastal lagoon (3000 and 10000 ind m<sup>-2</sup>) (Quintino, 1988); in Óbidos coastal lagoon mean values varied between 7000 and 18000 ind m<sup>-2</sup> (Quintino, 1988), and more recently Cancela da Fonseca *et al.*, (2006) found mean values that ranged from 20 to 13000 ind m<sup>-2</sup>.

The range of diversity values found in Foz de Almargem ( $H'_{\min} = 0.59$ ;  $H'_{\max} = 1.52$ ) and Salgados ( $H'_{\min} = 0.51$ ;  $H'_{\max} = 2.03$ ) were similar to those determined by Cancela da Fonseca (1989) in Santo André coastal lagoon (0.5-1.5 bits) and lower than the values described for Albufeira coastal lagoon (2-3.5 bits). In Óbidos coastal lagoon, Quintino (1988) determined diversity values around 3.0 bits, but a more recent study revealed diversity values that went from 0.37 to 3.31 (Cancela da Fonseca, 2006).

In choked lagoons (such as Foz de Almargem, Salgados, Santo André, Albufeira and Óbidos lagoons), the diversity is low and communities are usually dominated by a few, but very abundant, small sized opportunistic species (*r*-selected species, characterized by a short generation time, a high reproductive effort and many small offspring) (Gamito *et al.*, 2005).

In terms of constancy, during the whole studied period, Foz de Almargem presented a greater percentage of constant *taxa* (60%), followed by accessory *taxa* (30%) and

accidental *taxa* (10%). The constant *taxa* category included *Chironomus* sp., *Hediste diversicolor*, *Lekanesphaera hookeri*, *Hydrobia ulvae*, *Abra segmentum* and *Cerastoderma glaucum*. The accessory *taxa* in the lagoon were *Capitella capitata*, *Ventrosia ventrosa* and Oligochaeta. An unidentified Polychaeta was the accidental *taxon*.

Most *taxa* that occurred in Foz de Almargem were also identified in other Portuguese lagoons (e.g. *Abra segmentum*, *Cerastoderma glaucum*, *Lekanesphaera hookeri*, *Hediste diversicolor*, *Chironomus* sp.) (Correia *et al.*, 2012 and references therein). In Ria Formosa coastal lagoon, one of the sites studied by Gamito (1994) presented a benthic macroinvertebrate community similar to that found in Foz de Almargem, which was dominated by *H. ulvae*, *V. ventrosa*, *C. glaucum*, *A. segmentum*, *C. capitata* and *C. salinarius*. According to the author, this group of species can tolerate high salinity as well as large variations in salinity, suspended matter and pH. Temperature and large variations of dissolved oxygen also influence these species, although to a smaller extent. The site was considered to have characteristics of a stressful environment, caused by very restricted water renewal.

In Salgados lagoon, Insecta was the only taxonomic group observed during the whole studied period and most of the time it showed monthly mean densities greater than the other groups. Constant *taxa* represented just 13% of the *taxa* identified in Salgados lagoon and the majority of *taxa* were accessory (69%). Accidental *taxa* accounted 19%. *Chironomus* sp. and *Berosus spinosus* were the constant *taxa* in Salgados lagoon; accessory *taxa* included the Insecta Ephydriidae, Tabanidae, Ceratopogonidae, Brachyceres, *Corixa affinis*, *Hesperocorixa sahlbergi*, *Parasigara infuscata*, Oligochaeta Tubificidae and Enchytraeidae, *Lekanesphaera hookeri* and the Amphipoda *Corophium multisetosum*; accidental *taxa* were the Insecta Empididae, Rhagionidae and *Notonecta* sp..

In Santo André coastal lagoon, during the decade of 1980, the benthic community was dominated by lagoonal and continental *taxa*, presenting a great number and diversity of insect species. This period of time was characterized by eutrophic conditions (e.g. low dissolved oxygen concentrations) and low salinity values, associated to insufficient water renewal and interchanges with the sea (Cancela da Fonseca, 1989; Bernardo, 1990; Correia *et al.*, 2012).

In Foz de Almargem and Salgados lagoons, differences in densities of benthic macroinvertebrate *taxa* were associated to some environmental parameters, such as temperature, total dissolved inorganic nitrogen, total phosphorus, chlorophyll *a* concentration in water, chlorophyll *a* concentration in sediment and clay content. For example the Isopoda *Lekanesphaera hookeri* was present in most samples from the two lagoons and its density was negatively associated with temperature and positively related with chlorophyll *a* in the sediment and total dissolved inorganic nitrogen concentration.

### **5.5.2. Benthic trophic groups**

Regarding trophic groups, Foz de Almargem presented a greater diversity of groups (suspension feeders, deposit feeders, suspension/deposit feeders, herbivorous and carnivorous/scavengers/omnivorous) than Salgados lagoon (deposit feeders, herbivorous and carnivorous/scavengers/omnivorous). Both lagoons were dominated by deposit feeders.

Benthic macroinvertebrate assemblages in choked lagoons are mainly composed by deposit-feeders and some herbivorous crustaceans (*e.g. Sphaeroma, Corophium*) grazer gastropods (*e.g. Hydrobia*), carnivorous polychaetes (*e.g. Hediste*) and deposit-feeders insect larvae (*e.g. Chironomids*) (Gamito *et al.*, 2005).

Boaventura *et al.* (1999) refer to deposit feeders as the most abundant group in Santo André coastal lagoon. Other studies point out that this trophic group dominates benthic communities in coastal ecosystems, mentioning the relation between deposit feeders abundance and fine sediments, rich in organic matter (Barnes and Villiers, 2000; Levinton and Kelaher, 2004).

Some authors (*e.g. Pearson and Rosenberg, 1978; Weston, 1990; Warwick and Clarke, 1994*) found that the effects of anoxia, due to the increase of organic matter load, cause strong changes on the benthic community by limiting the growth of benthic filter feeders and thus, providing a shift in species composition from bivalves to polychaetes and oligochaetes. In Salgados lagoon, the absence of filter feeders can be related to hypoxia and anoxia events, associated to eutrophication.

### 5.5.3. Benthic ecological groups

In terms of ecological groups, the composition of benthic macroinvertebrate communities in Foz de Almargem and Salgados was distinct, although both lagoons were dominated by *taxa* tolerant to excess organic matter enrichment (Group III – AMBI).

In Foz de Almargem, the densities of group III *taxa* accounted 96% to 100% of the monthly mean densities (annual mean: 99%), whereas in Salgados lagoon these *taxa* represented 49% to 94% (annual mean: 77%).

The remaining *taxa* identified in Foz de Almargem belong to group V, which include first-order opportunistic *taxa* associated with pronounced unbalanced situations. Although these *taxa* occurred during most of the studied period, they had little expression in terms of monthly mean densities and annual mean percentage (1%).

In Salgados lagoon, group V *taxa* were not found in some sampling periods, but in the other months comprised 3% to 27% of monthly mean densities (annual mean: 11%). Second-order opportunistic *taxa*, indicators of slight to pronounced unbalanced situations (Group IV - AMBI), were observed in Salgados lagoon during all studied period except in July 2002. These *taxa* represented 1% to 26% of monthly mean densities (annual mean: 7%). Nevertheless, some of the *taxa* found in Salgados are not assigned in any of the AMBI groups defined by Borja *et al.* (2010).

### 5.5.4. Benthic macroinvertebrate salinity tolerance

The comparison of benthic macroinvertebrate salinity tolerances from the two lagoons showed that Foz de Almargem had a greater marine influence, while Salgados community was mostly influenced by freshwater inputs.

Benthic macroinvertebrate community in Foz de Almargem lagoon was mainly composed by eurihaline marine *taxa* and brackish *taxa*. In Salgados lagoon all *taxa* were brackish, although most of them occurred in oligohaline and mesohaline conditions or just in mesohaline conditions.

In terms of density, brackish *taxa* (oligohaline-mesohaline-polyhaline) showed the highest values during most of the studied period, with the exception of October 2001, when brackish *taxa* (mesohaline-polyhaline) density was superior.

In choked lagoons, which are open to the sea just in a few occasions and during short periods of time as it happens in Foz de Almargem and Salgados lagoons, the migration of marine organisms is intermittent and dependant on the openings of the lagoons to the sea. The presence of marine organisms inside the lagoons can be temporary, depending on the lagoon conditions, and may lead to a situation where only few species survive throughout the year (Pérez-Ruzafa *et al.*, 1987, 1991).

## **5.6. Lagoons inlets open *versus* closed**

The comparison of data when the lagoons were closed (December 2001) and connected to the sea (January 2002) showed that, in both lagoons, there was a decrease in total solids in suspension, dissolved oxygen, chlorophyll *a* concentrations and an increase in temperature, salinity, nitrates, nitrites, total dissolved inorganic nitrogen, N:P ratio and pigments diversity. Orthophosphates, total phosphorus and pH increased in Foz de Almargem and decreased in Salgados lagoon, while ammonia and phaeo-pigments concentrations followed the opposite tendency.

Concerning sediment parameters, it was registered an increase in chlorophyll *a* degradation index in the two lagoons. This parameter, just as silt content, sand content, chlorophyll *a* and phaeo-pigments concentrations were the ones with major variations.

Regarding phytoplankton parameters, both lagoons presented a decrease in Shannon-Wiener diversity, evenness, Bacillariophyceae abundances, Euglenophyceae abundances and an increase in Cyanophyceae abundances.

To what concerns benthic macroinvertebrate communities, total densities presented different tendencies in the two lagoons, increasing in Foz de Almargem and decreasing in Salgados. The higher density found in Foz de Almargem was due to an augment in the Gastropoda *Hydrobia ulvae* and the Isopoda *Lekanesphaera hookeri*, while in Salgados lagoon, the major cause for the reduction of total benthos density was the diminution of the Insecta *Chironomus* sp..

The structure and dynamics of benthic communities are mainly influenced by the communication with the sea and the hydrological and meteorological conditions (Cancela da Fonseca, 1989). The marine renewal and the maintenance of brackish characteristics are essential to the secondary production of the system (Cancela da Fonseca, 1989; Bernardo, 1990) and to prevent eutrophication of coastal lagoons and a decrease in water quality.

When the lagoons were in connection with the sea, some of the major effects described by other authors (Barnes, 1980; Cancela da Fonseca, 1989; Bernardo, 1990; Pereira Coutinho *et al.*, 2012) were observed in Foz de Almargem and Salgados lagoons, that was the increase in salinity and the discharge of biological, organic and inorganic material accumulated in the lagoons (*e.g.* total solids in suspension and chlorophyll *a* in the water, clay and silt from the sediment). Still, the lagoons had an increase in some nutrients concentrations, namely dissolved inorganic nitrogen compounds. These compounds were mainly nitrates and nitrites, which could be associated to the river runoff due to the high rainfall registered in January 2002. In Salgados lagoon, the increase and high value of ammonia concentration could have resulted from the release of this compound from the sediment in situation of oxygen depletion near the bottom, as it was registered by Bernardo (1990) in Santo André lagoon.

During the period of connection with the sea, the mean values of all trophic state and water quality indexes showed a decrease in Salgados lagoon, while in Foz de Almargem there was an increase. TSI (CHL) was the only index in Foz de Almargem lagoon that maintained the same classification as in December 2001, oligomesotrophic system. According to TSI (TP) and TRIX, the trophic state changed from oligotrophic to eutrophic and there was deterioration in water quality from good to mediocre.

In Salgados lagoon, TSI (CHL) classification changed from hypereutrophic to eutrophic-hypereutrophic, but according to TSI (TP) and TRIX the trophic state and water quality was the same in the two periods, hypereutrophic and poor water quality respectively.

## 5.7. Trophic state and ecological quality

The evaluation of the trophic state and water quality through monthly mean values and annual mean values of TSI (CHL), TSI (TP) and TRIX, determined that Foz de Almargem presented a lower trophic state and better water quality than Salgados lagoon.

According to the 90<sup>th</sup> percentile values of chlorophyll *a* and the criteria defined by Pereira Coutinho *et al.* (2012) for semi-enclosed lagoons, during the period lagoons were closed, water quality was high in Foz de Almargem and bad in Salgados lagoon. Data from both lagoons were insufficient for the determination of water quality when the lagoons were opened.

Several indicators, indexes and models have been developed to assess the trophic state and water quality in freshwater, estuarine, coastal and marine systems. Table 5.1 resumes the classification of Foz de Almargem and Salgados lagoons accordingly to some of these indicators.

Salgados lagoon presented serious problems of eutrophication and water quality, as all indicators and indexes from freshwaters to coastal waters classified the lagoon with the highest trophic state and the worst water quality.

The trophic state classification of Foz de Almargem based on chlorophyll *a* criteria was lower than the classifications based on nutrients (nitrogen and phosphorus); water quality was also better when chlorophyll *a* was used as indicator. For instance, when TSI is determined with two or more variables (chlorophyll *a*, total phosphorus and Secchi disk), differences in TSI values might occur, as it happened in Lake Skadar (Rakocevic-Nedovic and Holler, 2005). In this case, chlorophyll *a* is a better predictor of algal biomass than TSI (TP) or TSI (SD) (Carlson and Simpson, 1996).

According to chlorophyll *a* criteria, Foz de Almargem would be an oligomesotrophic system, if TSI (CHL) for lakes and reservoirs is considered (Carlson, 1977; Carlson and Simpson, 1996); a system with low eutrophication, based on estuaries ranges (Bricker *et al.*, 2003); a mesotrophic system, considering the limits defined for freshwaters (Likens, 1975; Wetzel, 1983) and still waters (Vollenweider and Kerekes, 1982).

The trophic categories defined by nitrogen concentrations classified Foz de Almargem as an oligomesotrophic system, according to freshwater criteria (Likens, 1975; Wetzel, 1983) and a system with medium eutrophication, based on the limits established for estuaries (Bricker *et al.*, 2003).

Phosphorus concentration criteria for still waters (Vollenweider and Kerekes, 1982), lakes and reservoirs (Carlson, 1977; Carlson and Simpson, 1996) increased Foz de Almargem trophic state to eutrophic. Nevertheless, phosphorus concentration in the lagoon corresponded to medium eutrophication in estuaries, just as the classification based on nitrogen concentration (Bricker *et al.*, 2003).

None of the previous indicators and indexes seems to be appropriate for the evaluation of the trophic state in Foz de Almargem lagoon, as they diverge in their classification due to the reference intervals defined for each type of system. Freshwater systems in general tend to present lower concentrations of phosphorus, being one of the major limiting elements for primary production in aquatic systems (Wetzel, 1983). In coastal lagoons, phosphorus compounds are generally more abundant than in freshwater systems, thereby the trophic state classification based on this nutrient might be overestimated.

Cloern (2001) found fundamental differences in the system-level responses to nutrient enrichment in lakes compared to estuarine-coastal ecosystems, suggesting that the old models and assumptions on the response of the system to nutrient inputs should be reviewed.

The assessment of eutrophication and water quality classification in coastal lagoons is not an easy task due to the great variability of spatial and temporal conditions. Although most of the year the Foz de Almargem and Salgados lagoons were isolated from the sea presenting some characteristics similar to still waters, lakes and reservoirs, during the short periods of connection with the sea they functioned just like small estuaries. However, the tidal range was smaller than in real estuaries, reducing considerably the flooded area of the lagoons and increasing the area of the sediment exposed to air.

Coastal lagoons exhibit a wide range of salinities, depending on the hydrological balance and on local climatic conditions (Kjerfve, 1994), and may vary from nearly freshwater to hyperhaline systems. Apart from climate, the hydrological features are moulded to a certain extent by the morphology of the lagoon and by the dimension of

the canals through which exchange of water with the sea occurs. The hydrological characteristics (such as salinity) are also influenced by the balance of precipitation, freshwater input, evaporation, tidal range and tidal flushing of the lagoon (Bird, 1994).

Table 5.1 – Trophic state and water quality in Foz de Almargem and Salgados lagoons, based on different authors and approaches: (1) Likens (1975), Wetzel (1983); (2) Vollenweider and Kerekes (1982); (3) Carlson (1977), Carlson and Simpson (1996); (4) USNEEA (Bricker *et al.*, 2003); (5) EEA (1999); (6) Pereira Coutinho *et al.* (2012); (7) Brito *et al.* (2012); (8) Vollenweider *et al.* (1998), Penna *et al.* (2004)

<i>Ecosystems</i>	<i>Indicators/Indexes</i>	<i>Foz de Almargem lagoon</i>	<i>Salgados lagoon</i>
<sup>(1)</sup> Freshwater	Dissolved Inorganic Nitrogen annual average	Oligomesotrophic (250-600 $\mu\text{g L}^{-1}$ N $\approx$ 18-43 $\mu\text{MN}$ )	Hypereutrophic (500-15000 $\mu\text{g L}^{-1}$ N $\approx$ 35-1071 $\mu\text{MN}$ )
	Chlorophyll <i>a</i> annual average	Mesotrophic (2-15 $\mu\text{g L}^{-1}$ )	Eutrophic (10-500 $\mu\text{g L}^{-1}$ )
<sup>(2)</sup> Still Waters	Chlorophyll <i>a</i> annual average	Mesotrophic (2.5-8 $\mu\text{g L}^{-1}$ )	Hypereutrophic ( $>$ 25 $\mu\text{g L}^{-1}$ )
	Maximum Chlorophyll <i>a</i>	Mesotrophic (8-25 $\mu\text{g L}^{-1}$ )	Hypereutrophic ( $>$ 75 $\mu\text{g L}^{-1}$ )
	Total Phosphorus annual average	Eutrophic (35-100 $\mu\text{g L}^{-1}$ P $\approx$ 1.13-3.23 $\mu\text{MP}$ )	Hypereutrophic ( $>$ 100 $\mu\text{g L}^{-1}$ P $\approx$ 3.23 $\mu\text{MP}$ )
<sup>(3)</sup> Lakes and Reservoirs	TSI (CHL)	Oligomesotrophic (30-40)	Hypereutrophic ( $>$ 70)
	TSI (TP)	Eutrophic (50-60)	Hypereutrophic ( $>$ 70)
<sup>(4)</sup> Estuaries	Dissolved Inorganic Nitrogen	Medium Eutrophication (0.1-1 $\text{mg L}^{-1}$ $\approx$ 7-71 $\mu\text{MN}$ )	High Eutrophication ( $\geq$ 1 $\text{mg L}^{-1}$ $\approx$ 71 $\mu\text{MN}$ )
	Total Phosphorus	Medium Eutrophication (0.01-0.1 $\text{mg L}^{-1}$ $\approx$ 0.32-3.22 $\mu\text{MP}$ )	High Eutrophication ( $>$ 0.1 $\text{mg L}^{-1}$ $\approx$ 3.22 $\mu\text{MP}$ )
	Chlorophyll <i>a</i> annual average	Low Eutrophication ( $\leq$ 5 $\mu\text{g L}^{-1}$ )	Hypereutrophic ( $>$ 60 $\mu\text{g L}^{-1}$ )
<sup>(5)</sup> Transitional, Coastal and Marine Waters	Phosphate annual average	Poor water quality (0.7 – 1.1 $\mu\text{mol L}^{-1}$ )	Bad water quality ( $>$ 16 $\mu\text{mol L}^{-1}$ )
	Nitrate + Nitrite annual average	Bad water quality ( $>$ 16 $\mu\text{mol L}^{-1}$ )	Bad water quality ( $>$ 16 $\mu\text{mol L}^{-1}$ )
<sup>(6)</sup> Semi-enclosed coastal lagoons	90 <sup>th</sup> percentile Chlorophyll <i>a</i> (closed period)	High water quality ( $<$ 30 $\mu\text{g L}^{-1}$ )	Bad water quality ( $>$ 101.3 $\mu\text{g L}^{-1}$ )
<sup>(7)</sup> Open coastal lagoons	90 <sup>th</sup> percentile Chlorophyll <i>a</i> (phytoplankton growing period)	Good/High water quality ( $<$ 8 $\mu\text{g L}^{-1}$ ; 8-12 $\mu\text{g L}^{-1}$ )	Bad water quality ( $>$ 27 $\mu\text{g L}^{-1}$ )
<sup>(8)</sup> Coastal Waters	TRIX	Mediocre water quality, moderate to highly productive, high trophic level (5-6)	Poor water quality, highly productive, greatest trophic level ( $>$ 8)

Some authors (*e.g.* Salas *et al.*, 2008; Brito *et al.*, 2010) have used the criteria defined by EEA (1999) for transitional, coastal and marine waters, to assess water quality in coastal lagoons that are permanently in connection with the sea, as the Mar Menor lagoon in Spain and Ria Formosa lagoon. In Foz de Almargem, these criteria classified water quality as poor based on phosphate annual average and if nitrate + nitrate annual average was considered, water quality would be bad. This classification system might not be suitable for coastal lagoons like Foz de Almargem and Salgados, once they are semi-enclosed lagoons with lower water renewal and greater freshwater influence than open coastal lagoons such as the Mar Menor and Ria Formosa lagoons.

The trophic status index TRIX proposed by Vollenweider *et al.* (1998) and the classification of water quality based on its values defined by Penna *et al.* (2004) was originally developed for Italian coastal waters and has been applied in the Lagoon of Venice and many European coastal areas in the Adriatic, Tyrrhenian, Baltic, Black and Northern seas (Pettine *et al.*, 2007). Foz de Almargem was the first semi-enclosed coastal lagoon where TRIX was applied as a water quality index and the annual mean value pointed to a mediocre water quality, characteristic of moderate to highly productive waters, with a great trophic level (Coelho *et al.*, 2007). As the lagoon-sea exchanges are restricted and the water parameters inside a lagoon differ from the adjacent coastal waters, specific classification criteria should be created between TRIX and water quality in the coastal lagoons.

In 2001, the European Environmental Agency suggested that the ranges of TRIX should be defined for different regions or areas in order to increase the index sensibility and that it would be important to decide which data should be used for the TRIX calculation (annual averages, seasonal averages) in order to make the index less sensitive to natural meteorological forced variations. Nevertheless, EEA considered that the general approach of the TRIX had a high potential and after further development it could be a practicable and comparable method for monitoring and assessing the trophic state, determining eutrophication trends of European marine and coastal waters (EEA, 2001). Salas *et al.* (2008) tested the performance and robustness of TRIX for its efficiency in describing the ecological status of two coastal systems of Iberian Peninsula, the Mondego estuary (Portugal) and the Mar Menor lagoon (Spain). Their results demonstrated that TRIX was not a trustworthy tool to classify eutrophication status in estuarine waters and the authors agreed that, if the index would be applied over wide

areas, the classification criteria should be adapted to the specific environments.

The criteria proposed by Brito *et al.* (2012) based on the chlorophyll *a* concentration during the phytoplankton growing period, would be a good approach for water quality classification in Foz de Almargem and Salgados lagoons, as the reference values were defined for coastal lagoons in the same geographic region (South Algarve). However, these criteria were determined for open coastal lagoons (Ria Formosa and Ria de Alvor), which present a greater influence from sea water and are permanently affected by tides. Thereby, the most suitable classification system for Foz de Almargem and Salgados water quality seems to be the one recently proposed by Pereira Coutinho *et al.* (2012) for semi-enclosed lagoons, as it was developed in coastal lagoons with characteristics similar to those found in Foz de Almargem and Salgados. The full integration method was not applied in the studied lagoons, as it requires several data from the closed period and open period to determine a final ecological quality ratio (EQR), but a preliminary classification of high water quality was determined in Foz de Almargem and a bad water quality in Salgados lagoon, for the period lagoons were closed. Besides phytoplankton biomass evaluation (90<sup>th</sup> percentile of chlorophyll *a*) during the closed and open period, this methodology also evaluates water quality based on phytoplankton blooms in each period.

Pereira Coutinho *et al.* (2012) were able to find a phytoplankton response (namely in terms of chlorophyll *a*) to nutrient enrichment in the western semi-enclosed coastal lagoons. In Foz de Almargem, chlorophyll *a* was related to nitrogen compounds concentrations, but in Salgados lagoon no direct relation was found.

It would be important to define reference conditions for nutrients in semi-enclosed lagoons for the periods lagoons are closed and open, just as it was done with chlorophyll *a* by Pereira Coutinho *et al.* (2012), and a similar methodology could be used to determine the ecological quality ratio and evaluate water quality in this particular type of coastal lagoons.

As coastal lagoons are strongly influenced by freshwater inputs, sea water interexchange and the water level suffers great fluctuations from one year to another, further studies should also include inter-annual comparisons in order to define the

evolution of the lagoons trophic state and water quality tendencies, as it is required by the Water Framework Directive.

The complexity of hydrological, physical and chemical factors interacting in a coastal lagoon should be analysed jointly with biological components, particularly phytoplankton and benthic communities. Phytoplankton communities, closely depending on nutrient distribution, can be considered an indirect index of water trophism (Bianchi *et al.*, 2003). Benthic communities, just as phytoplankton communities, are one of the biological elements considered by the Water Framework Directive to assess ecological quality status, since they have long been used to assess quality of aquatic ecosystems (Correia *et al.*, 2002 and references therein). However, their response to the highly variable and unpredictable conditions of coastal lagoons, has been indicated as a major problem since it is difficult to separate between the effects of natural and anthropogenic stress (Elliot and Quintino, 2007).

## 6. REFERENCES

- Badylak, S., Philips, E.J., 2004. Spatial and temporal patterns of phytoplankton composition in subtropical coastal lagoon, the Indian River Lagoon, Florida, USA. *Journal of Plankton Research* 26 (10), 1229-1247.
- Balkis, N., 2003. Seasonal variations in the phytoplankton and nutrient dynamics in the neritic water of Büyükçekmece Bay, Sea of Marmara. *Journal of Plankton Research Part VII* 25, 703-727.
- Barnes, R.S.K., 1980. *Coastal lagoons*. Cambridge University Press, Cambridge, 106 pp.
- Barnes, R.S.K., Villiers, C.J., 2000. Animal abundance and food availability in coastal lagoons and intertidal marine sediments. *Journal of the Marine Biological Assessment of the United Kingdom* 80, 193-202.
- Bernardo, J. M., 1990. *Dinâmica de uma lagoa costeira eutrófica (Lagoa de Santo André)*. Ph. D. Thesis, Faculdade de Ciências da Universidade de Lisboa, 322 pp.
- Berthois, L. 1956. *Technique de l'analyse granulométrique*. Paris, École Pratique des Hautes Études, Lab. Géom. Mém. 6. 76 pp.
- Bettencourt A. M., Bricker, S. B., Ferreira, J. G., Franco, A., Marques, J. C., Melo, J. J., Nobre, A., Ramos, L., Reis, C. S., Salas, F., Silva, M. C., Simas, T., Wolff, W. J., 2004. *Typology and Reference Conditions for Portuguese Transitional and Coastal Waters. Development of Guidelines for the Application of the European Union Water Framework Directive*. INAG/IMAR, 98 p. <http://www.ecowin.org/ticor/>
- Bianchi, F., Acri, F., Bernardi Aubry, F., Berton, A., Boldrin, A., Camatti, E., Cassin, D., Comaschi, A., 2003. Can plankton communities be considered as bio-indicators of water quality in the Lagoon of Venice? *Marine Pollution Bulletin* 46, 964-971.
- Bird, E. C. F., 1994. Physical setting and geomorphology of coastal lagoons. In Kjerfve, B. (Ed.) *Coastal Lagoon Processes*. Elsevier Oceanography Series 60, 9-39.
- Borja, A., Franco, F., Pérez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within European estuarine and coastal environments. *Marine Pollution Bulletin* 40, 1100-1114.
- Bricker, S. B., Ferreira, J. G., Simas, T., 2003. An integrated methodology for assessment of estuarine trophic status. *Ecological Modelling* 169, 39-60.
- Brito, A., Newton, A., Tett, P., Fernandes, T., 2010. Sediment-water interactions in a coastal shallow lagoon, Ria Formosa (Portugal): implications within the water framework directive. *Journal of Environmental Monitoring* 12, 318-328.
- Brito, A.C., Quental T., Coutinho T.P., Branco M.A.C., Falcão M., Newton A., Icely J., Moita T., 2012. Phytoplankton dynamics in Southern Portuguese coastal lagoons during a discontinuous period of 40 years: An overview. *Estuarine, Coastal and Shelf Science* 110, 147-156.

- Cancela da Fonseca, L., 1989. *Estudo da influência da “abertura ao mar” sobre um sistema lagunar costeiro: a Lagoa de Santo André*. Ph. D. Thesis, Faculdade de Ciências da Universidade de Lisboa, 327 pp.
- Cancela da Fonseca, L., Costa, A. M., Bernardo, J.M., Fonseca, R., 1987. Lagoa de Santo André (SW Portugal): Phytopigments as sediment tracers. *Limnetica*, 3(2), 299-306.
- Cancela da Fonseca, L., Pereira, P., Gaspar, M., Moura, A., Carvalho, S., Leitão, F., Drago, T., Falcão, M., 2006. Macrozoobentos e ambiente sedimentar da Lagoa de Óbidos (Costa Oeste de Portugal): um estudo de referência. In ICN/CEZH (Eds.) *Actas do 1º Seminário sobre Sistemas Lagunares Costeiros*, Vila Nova de Milfontes.
- Carlson, R. E., 1977. A trophic state index for lakes. *Limnology and Oceanography* 22, 361- 369.
- Carlson, R. E., Simpson J., 1996. *A Coordinator`s Guide to Volunteer Lake Monitoring Methods*. North American Lake Management Society, Madison, 96 pp.
- C.E.C., 2000. Council Directive of 23 October 2000, establishing a framework for community action in the field of water policy (2000/60/EC). *Official Journal of European Community*, L327 of 2000/12/22, 1-72.
- CEMAGREF-IARE, 1994. *Recherche d'indicateurs de niveaux trophiques dans les lagunes méditerranéennes: Analyse bibliographique*. Agence de l'Eau RMC, Lyon. France, 115 pp.
- Chorus, I., Bartram, J. (eds), 1999. *Toxic Cyanobacteria in Water: A Guide to their Public Health Consequences, Monitoring and Management*. Published on behalf of WHO by E & FN Spon /Chapman & Hall, London, 416 pp.
- Chrétiennot-Dinet, M-J., 1990. *Atlas du Phytoplancton Marin-volume III: Chlorarachniophycées, Chlorophycées, Chrysophycées, Euglénophycées, Eustigmaphycées, Prasinophycées, Prymnésiophycées, Rhodophycées et Tribophycées*. Centre National de la Recherche Scientifique, Paris, 261 pp.
- Cloern, J.E., 2001. Our evolving conceptual model of the coastal eutrophication problem. *Marine Ecology Progress Series* 201, 223-253.
- Cloern, J.E., Cole, B.E., Raymond, L.J.W., Alpine, A.E., 1985. Temporal dynamics of estuarine phytoplankton: a case study of San Francisco Bay. *Hydrobiologia* 129, 153-176.
- Coelho, S., Gamito, S., Pérez-Ruzafa, A., 2007. Trophic state of Foz de almargem coastal lagoon (Algarve, South Portugal) based on the water quality and the phytoplankton community. *Estuarine, Coastal and Shelf Science* 71: 218-231.
- Colombo, G., 1977. Lagoons. In: Barnes, R.S.K. (Ed.), *The Coastline*, John Wiley and Sons, Chichester, pp: 63-82.
- Comin, F.A., 1982. Seasonal changes of phytoplankton in three coastal lagoons of the Ebro delta in relation to environmental factors. *Oceanologica Acta Part V 4 Supplement*, 259-267.
- Conte de Barros, M., 1996. Eutrofização: Um problema de Desenvolvimento Sustentável- Controlo de Qualidade. *Indústria da Água* 20, Julho-Setembro.

- Correia, M.J., Costa, J.L., Chainho, P., Félix, P.M., Chaves, M.L., Medeiros, J.P., Silva, G., Azeda, C., Tavares, P., Costa, A., Costa, A.M., Bernardo, J., Cabral, H.N., Costa, M.J., Cancela da Fonseca, L., 2012. Inter-annual variations of macrobenthic communities over three decades in a land-locked coastal lagoon (Santo André, SW Portugal). *Estuarine, Coastal and Shelf Science* 110, 168-175.
- Cortes, R.M., Carvalho, L.H., Carvalho, M.J., 1991. Caracterização físico-química das águas dulciaquícolas, implicações biológicas. Série didáctica Ciências Aplicadas 15, Universidade de Trás-os-Montes e alto Douro, Vila Real, 131 pp.
- Cox, E. J., 1996. *Identification of Fresh Diatoms from Live Material*. Chapman & Hall, Oxford, 158 pp.
- Cravo, A. 2003. *Avaliação do Efeito das Descargas de Águas Residuais Urbanas na Ria Formosa (DRAOT/UALG)*. Final report, Universidade do Algarve, Faro, 131 pp.
- Cronberg, G., Haecky, P., 2006. *Manual on aquatic cyanobacteria: a photo guide and a synopsis of their toxicology*. International Society for the Study of Harmful Algae and the United Nations Educational, Scientific and Cultural Organization, Denmark, 106 pp.
- Dogde, J. D. 1982. *Marine Dinoflagellates of the British Isles*. University of London, London, 295 pp.
- DRAOT-ALGARVE, 2001. Recursos Hídricos na Região do Algarve: Situação em Novembro de 2001. Ministério do Ambiente e do Ordenamento do Território, 96 pp.
- DRAOT-ALGARVE, 2002. Recursos Hídricos na Região do Algarve: Situação em Maio de 2002. Ministério do Ambiente e do Ordenamento do Território, 97 pp.
- Elliot, M., Quintino, V., 2007. The estuarine quality paradox. Environmental homeostasis and the difficulty of detecting anthropogenic stress in naturally stressed areas. *Marine Pollution Bulletin* 54, 640-645.
- EEA, European Environmental Agency, 1999. *Nutrients in European Ecosystems. Environmental Assessment Report N° 4*. Office for official publications of the European Communities, 155 pp.
- EEA, European Environmental Agency, 2001. *Eutrophication in Europe's coastal waters*. Topic Report N° 7, Copenhagen, 86 pp.
- EPA, US Environmental Protection Agency, 2003. *Ambient water quality criteria for dissolved oxygen, water clarity and chlorophyll a for the Chesapeake Bay and its tidal tributaries*. EPA, Office of Water/ Office of Science and Technology, Washington DC, 341 pp.
- Falcão, M., 1996. *Dinâmica dos nutrientes na Ria Formosa: efeitos da interacção da laguna com as suas interfaces na reciclagem do azoto, fósforo e sílica*. Ph.D. Thesis, Universidade do Algarve, 223 pp.
- Faust, M.A., Gullledge, R.A., 2002. Botany: Identifying Harmful Marine Dinoflagellates. *Contributions from the United States National Herbarium* 42, 1-144.

- Fauvel, P., 1975. *Faune de France: 5- Polychètes errantes*. Reprit Libraire de la Faculté des Sciences, Paris, 488 pp.
- Fernandes, J., 2001. *Caracterização dos elementos naturais da Praia Grande e da Praia da Marinha e propostas de valorização*. Universidade do Algarve & Direcção Regional do Ambiente do Algarve, 190 pp.
- Ferreira, J. G., Vale, C., Soares, C.V., Salas, F., Stacey, P.E., Bricker, S. B., Silva, M.C., Marques, J.C., 2007. Monitoring of coastal and transitional waters under the EU Water Framework Directive. *Environmental Monitoring and Assessment* 135, 195-216.
- Flemming, B.W., 2000. A Revised Textural Classification of Gravel-free Muddy Sediments on the Basis of Ternary Diagrams. *Continental Shelf Research* 20, 1125-1137.
- Gamito, S., 1994. *The benthic ecology of some Ria Formosa lagoons, with reference to the potential for production of the gilthead seabream (Sparus aurata L.)*. PhD Thesis, Universidade do Algarve, 255 pp.
- Gamito, S., 2006. Benthic ecology of semi-natural coastal lagoons, in Ria Formosa (Southern Portugal), exposed to different water renewal regimes. *Hydrobiologia* 555, 75-87.
- Gamito, S., 2008. Three main stressors acting on the Ria Formosa lagoonal system (Southern Portugal): Physical stress, organic matter pollution and land-ocean gradient. *Estuarine, Coastal and Shelf Science* 77, 710-720.
- Gamito, S., Gilabert, J., Marcos Diego, C., Pérez-Ruzafa, A., 2005. Effects of changing environmental conditions on lagoon ecology. In Gönenç, I. E. and Wofflin J. (eds.), *Coastal Lagoons: Ecosystem processes and modeling for sustainable use and development*, CRC Press, Boca Raton, pp: 193–229.
- Glibert, P.M., Seitzinger, S., Heil, C.A., Burkholder, J.M., Parrow, M.W., Codispoti, L.A., Kelly, V., 2005. The role of eutrophication in the global proliferation of harmful algal blooms: new perspectives and new approaches. *Oceanography* 18, 198-209.
- Grall, J., Glémarec, M., 1997. Using biotic indices to estimate macrobenthic community perturbations in the Bay of Brest. *Estuarine, Coastal and Shelf Science* 44 (suppl. A), 43-53.
- Greenberg, A.E., Clesceri, L.S., Eaton, A.D. (eds), 1992. *Standard Methods for the examination of water and wastewater*. 18<sup>th</sup> Edition. American Public Health Association, Washington DC.
- Hajdu, S., Pertola, S., Kuosa, H., 2005. *Prorocentrum minimum* (Dinophyceae) in the Baltic Sea: morphology, occurrence – a review. *Harmful Algae* 4, 471-480.
- Hallegraeff, G.M., Anderson, D.M., Cembella, A.D., 1995. *Manual of Harmful Marine Microalgae*. UNESCO Manuals and Guides N° 33, Paris, 546 pp.
- Hartnett, M., Nash, S., 2004. Modelling nutrient and chlorophyll *a* dynamics in a Irish brackish waterbody. *Environmental Modelling and Software* 19, 47-56.
- Hashimoto, T., Nakano, S., 2003. Effect of nutrient limitation on abundance and growth of phytoplankton in a Japanese pearl farm. *Marine Ecology Progress Series* 258, 43–50.

- Hayward, P.J., Ryland, J.S., 1995. *Handbook of the marine fauna of North-West Europe*. Oxford University Press, New York, 800 pp.
- Heisler, J, Glibert, P.M., Burkholder, J.M., Anderson, D.M., Cochlan, W., Dennison, W.C., Dortch, Q., Gobler, C.J., Heil, C.A., E. Humphries, E., Lewitus, A., Magnien, R., Marshall, H.G., Sellner, K., Stockwell, D.A., Stoecker, D.K., Suddleson, M., 2008. Eutrophication and harmful algae blooms: A scientific consensus. *Harmful Algae* 8, 3-13.
- Holme N.A., McIntyre, A.D., 1984. *Methods for the study of Marine Benthos*. Blackwell Scientific Publications, London. 387 pp
- <http://geo.snirh.pt/AtlasAgua/>
- Järvinen, M., Drakare, S., Free, G., Lyche-Solheim, A., Phillips, G., Skjelbred, B., Mischke, U., Ott, I., Poikane, S., Søndergaard, M., Pasztaleniec, A., Van Wichelen, J., Portielje, R., 2013. Phytoplankton indicator taxa for reference conditions in northern and central European lowland lakes. *Hydrobiologia* 704: 97-113.
- Jarry, V., Fiala, M., Frisoni, G.F., Jacques, G., Neveux, J., Panouse, M., 1990. The spatial distribution of phytoplankton in a Mediterranean lagoon (Étang de Thau). *Oceanologica Acta* 13(4), 503-512.
- John, D. M., Whitton, B.A., Brook, A.J. (Eds), 2002. *The Freshwater Algal Flora of the British Isles: An Identification Guide to Freshwater and Terrestrial Algae*. The Natural History Museum and The British Phycological Society, Cambridge University Press, Cambridge, 702 pp.
- Kjerfve, B. and Magill, K.E., 1989. Geographic and hydrographic characteristics of shallow coastal lagoons, *Marine Geology* 88, 187-199.
- Kjerfve, B., 1994. Coastal Lagoons. In Kjerfve, B. (Ed.) *Coastal Lagoon Processes*. Elsevier Oceanography Series 60, 243-286.
- Krebs, C.J., 1994. *Ecology: The experimental Analysis of Distribution and Abundance*. 4th edition. Harper Collins College Publishers, New York, 801 pp.
- Landsberg, J.H., 2002. The Effects of Harmful Algal Blooms on Aquatic Organisms. *Reviews in Fisheries Science* 10:2, 113–390.
- Levinton, J., Kelaher, B., 2004. Opposing organizing forces of deposit-feeding marine communities. *Journal of Experimental Marine Biology and Ecology* 300, 65-82.
- Likens, G.E., 1975. Primary production of inland aquatic ecosystems. In: Lieth, H., Whittaker R.H. (Eds.) *Primary Productivity of the Biosphere*. Springer-Verlag, New York Inc, pp. 185-202.
- Lorenzen, C.J., 1967. Determination of chlorophyll and phaeo-pigments: spectrophotometric equations. *Limnology and Oceanography* Part II 12, 343-346.
- Macedo, M.F., Duarte, P., Mendes, P., Ferreira, J.G., 2001. Annual variation of environmental variables, phytoplankton species composition and photosynthetic parameters in a coastal lagoon. *Journal of Plankton Research* Part VII 23, 719-732.

- Margalef, R., 1960. Valeur indicatrice de la composition des pigments du phytoplancton sur la productivité, composition taxonomique et propriétés dynamiques des populations. *Rapport et Procès-verbaux des Réunion Commission Internationale pour l'Exploration Scientifique de la Mer Méditerranée* 15, 277-281.
- Margalef, R., 1983. *Limnologia*. Ed Omega, Barcelona, 1010 pp.
- Maroco, J. (2010). *Análise Estatística - Com Utilização do SPSS*. 3ª Edição. Edições Sílabo, Lisboa, 822 pp.
- Ministro, J.S., 2002. A importância da avifauna na gestão sustentável da Lagoa dos Salgados. Graduate thesis, Universidade do Algarve, 78 pp.
- Morais, P., Chícharo, M.A., Barbosa, A., 2003. Phytoplankton dynamics in a coastal saline lake (SE-Portugal). *Acta Oecologica* 24, S87-S96.
- Muxika, I., Borja, A., Bald, J.L., 2007. Using historical data, expert judgment and multivariate analysis in assessing reference conditions and benthic ecological status, according to the European Water Framework Directive. *Marine Pollution* 55: 6–29.
- Naylor, E., 1972. *British Marine Isopods – Keys and notes for the identification of the species*. Academic Press, 86 pp.
- Neves, M., 1999. *Qualidade da água na Lagoa dos Salgados*. Graduate thesis in Environmental Engineering. Universidade do Algarve, Faro, 65 pp.
- Newell, G. E., Newell, R. C., 1977. *Marine Plankton – a practical guide*. 5th Edition, Hutchinson & Co. (Publishers) Ltd, London, 244 pp.
- Newton, A, Mudge, S. M., 2005. Lagoon-sea exchanges, nutrient dynamics and water quality management in Ria Formosa (Portugal). *Estuarine, Coastal and Shelf Science* 62, 405-414.
- Newton, A., Icely, J.D., Falcão, M., Nobre, A., Nunes, J.P., Ferreira, J.G., Vale, C., 2003. Evaluation of eutrophication in the Ria Formosa coastal lagoon, Portugal. *Continental Shelf Research* 23, 1945-1961.
- Nuccio, C., Melillo, C., Massi, L., Innamorati, M., 2003. Phytoplankton abundance, community structure and diversity in the eutrophicated Orbetello lagoon (Tuscany) from 1995 to 2001. *Oceanologica Acta* 26, 15-25.
- Parsons, T., Maita, Y., Lalli, C.M., 1984. *A manual of chemical and biological methods for seawater analysis*. Pergamon Press, New York, 173 pp.
- Pearson, T.H., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *Oceanography and Marine Biology Annual Review* 16, 229-311.
- Penna, N., Capellacci, S., Ricci, F., 2004. The influence of the Po River discharge on phytoplankton bloom dynamics along the coastline of Pesaro (Italy) in the Adriatic Sea. *Marine Pollution Bulletin* 48, 321-326.
- Pereira Coutinho, M.T., Brito A.C., Pereira P., Gonçalves A.S., Moita M.T., 2012. A phytoplankton tool for water quality assessment in semi-closed coastal lagoons: Open vs closed regimes. *Estuarine, Coastal and Shelf Science* 110, 134-146.

- Pérez-Ruzafa, A., Marcos, C., Pérez Ruzafa, I.M., Ros, J.D., 1987. Evolución de las características ambientales y de los poblamientos del Mar Menor (Murcia, SE de España). *Anales de Biología* 12, 53-65.
- Pérez-Ruzafa, A., Marcos, C., Ros, J., 1991. Environmental and biological changes related to recent human activities in the Mar Menor. *Marine Pollution Bulletin* 23, 747-751.
- Pérez-Ruzafa, A., Gilabert, J., Gutiérrez, J.M., Fernández, A.I., Marcos, C., Sabah, S., 2002. Evidence of a planktonic food web response to changes in nutrient input dynamics in the Mar Menor coastal lagoon, Spain. *Hydrobiologia*, 475/476, 359-369.
- Pearson, T.H., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *Oceanography and Marine Biology Annual Review*, 229–311.
- Pettine, M., Casentini, B., Fazi, S., Giovanardi, F., Pagnotta, R., 2007. A revisitation of TRIX for trophic status assessment in the light of the European Water Framework Directive: Application to Italian coastal waters. *Marine pollution Bulletin* 54, 1413-1426.
- Pielou, E.C., 1984. *The interpretation of ecological data: a primer on classification and ordination*. John Wiley & Sons, N.Y. 263 pp.
- Pielou, E.C., 1966. The measurement of diversity in different types of biological collections. *Journal of Theoretical Biology* 13, 131-144.
- Pinto, C., Gaspar P. and Teixeira, S.B., 2001. Influência marinha na qualidade da água de uma lagoa costeira eutrófica (Lagoa dos Salgados, Algarve – Portugal). In Alvalade, H., Gonçalves S. and Dias R. (Eds) *II Jornadas Ibéricas de Jovens Geólogos*, Universidade de Évora.
- Plante-Cuny, M.R., 1974. Evaluation par spectrophotométrie des teneurs en chlorophylle *a* fonctionnelle et en phéopigments des substrats meubles marins. Doc. Sci. Mission O.R.S.T.O.M. Nosy-Bé, 45, 1-76
- Plante-Cuny, M.R., 1978. Pigments photosynthétiques et production primaire des fonds meubles néritiques d'une région tropicale (Nosy-Bé, Madagascar). *Journal de Recherche Oceanographique*, 3: 1-14
- Poppe, G.T., Goto, Y., 1993. *European Seashells-Volume II. Scaphopoda, Bivalvia, Cephalopoda*. Conch Books, Germany, 217 pp.
- Quintino, V., 1988. Structure et cinétique comparées des communautés de macrofauna benthique de deux systèmes lagunaires de la côte Ouest du Portugal: Óbidos et Albufeira. PhD Thesis, Université de Paris, 333 pp.
- Rakocevic-Nedovic, J., Holler, H., 2005. Phytoplankton community and chlorophyll *a* as trophic state indices of Lake Skadar (Montenegro, Balkan). *Environmental Science and Pollution Research Part III* 12, 146-152.
- Reiss, H., Kröncke, I., 2005. Seasonal variability of benthic indices: An approach to test the applicability of different indices for ecosystem quality assessment. *Marine Pollution Bulletin* 50, 1490-1499.
- Rield, R., 1983. *Fauna y Flora del Mar Mediterraneo*. Omega, Barcelona, 856 pp.

- Salas, F., Marcos, C., Neto, J.M., Patricio, J., Pérez-Ruzafa, A., Marques, J.C., 2006. User-friendly guide for using benthic ecological indicators in coastal and marine quality assessment. *Ocean & Coastal Management* 49, 308-331.
- Salas, F., Teixeira, H., Marcos, C., Marques, J.C., Pérez-Ruzafa, A., 2008. Applicability of the trophic index TRIX in two transitional ecosystems : the Mar Menor lagoon ( Spain ) and the Mondego estuary ( Portugal ). *ICES Journal of Marine Science* 65, 1442-1448.
- Shannon, C.E., Weaver, W., 1963. *The Mathematical Theory of Communication*. University of Illinois Press, Urbana, Illinois, USA, 125 pp.
- Soares, M.M., 2000. *Balanço hidrológico da Lagoa dos Salgados*. Graduate thesis, Universidade do Algarve, 47 pp.
- Sournia, A., 1984. Classification et nomenclature de divers dinoflagellés marins (Dinophyceae). *Phycology* 23 (3), 345-355.
- Sournia, A. 1986. *Atlas du Phytoplankton Marin: Volume I Introduction, Cyanophycées, Dictyochophycées, Dinophycées et Raphidophycées*. Centre National de la Recherche Scientifique, Paris, 219 pp.
- Sykes, J. B., 1981. *An illustrated guide to the diatoms of British coastal plankton*. Field Studies 5, 425-468.
- Tachet, H., Richoux, P., Bournaud, M., Usseglio-Polatera, P., 2002. *Invertébrés D'Eau Douce, Systématique, Biologie, Écologie*. CNRS Eds, Paris, 587 pp.
- Taylor, D.I., Nixon, S.W., Granger, S.L., Buckley, B.A., 1999. Responses of coastal lagoon plant communities to levels of nutrient enrichment: a mesocosm study. *Estuaries* 22, 1041-1056.
- Teixeira, S.B., 2009. *Demarcação do leito e da margem das águas do mar no litoral sul do Algarve*. Administração da Região Hidrográfica do Algarve, Faro, 207 pp.
- Teixeira, H., Salas, F., Borja, A., Neto, J.M., Marques, J.C., 2008. A benthic perspective in assessing the ecological status of estuaries: the case of the Mondego estuary (Portugal). *Ecological Indicators* 8: 404–416.
- Teixeira, H., Neto, J.M., Patrício, J., Veríssimo, H., Pinto, R., Salas, F., Marques, J.C., 2009. Quality assessment of benthic macroinvertebrates under the scope of WFD using BAT, the Benthic Assessment Tool. *Marine Pollution Bulletin* 58: 1477–1486.
- Tomas, C. R., Hasle, G. R. (Eds), 1997. *Identifying Marine Phytoplankton*. Academic Press, London, 858 pp.
- Tomàs-Vives, P. (ed.), 1996. *Monitoring Mediterranean Wetlands: A Methodological Guide*. MedWet Publication, Wetlands International, Slimbridge, UK and ICN, Lisbon, 150 pp.
- Trigueros, J. M., Ansotegui, A., Orive, E., 2000. Remarks on morphology and ecology of recurrent dinoflagellates species in the estuary of Urdaibai (Northern Spain). *Botanica Marina*, 43, 93-103.
- Utermöhl, H., 1958. Zur vervollkommnung der quantitativen Phytoplankton-Methodik. Mitt int. Verein.Theor. Angew. *Limnology and Oceanography* 9, 1-38.

- Vasconcelos, V.M., 2006. Eutrophication, cyanobacteria and cyanotoxins: when ecosystems cry for help. *Limnetica* 25 (1-2), 425-432.
- Vasconcelos, V.M., Pereira, E., 2001. Cyanobacteria diversity and toxicity in a Waste Water Treatment Plant (Portugal). *Water Research* 35, 1354-1357.
- Venrick, E.L., 1978. How many cells to count? In: Sournia, A. (Eds.) *Phytoplankton Manual: Monographs on Oceanographic Methodology*. UNESCO, U.K., pp 167-180.
- Viaroli, P., Bartoli, M., Giordani, G, Naldi, M., Orfanidis, S., Zaldivar, J.M., 2008. Community shifts, alternative stable states, biogeochemical controls and feedbacks in eutrophic coastal lagoons: a brief review. *Aquatic Conservation: Marine and Freshwater Ecosystems* 18, S105-S117.
- Vollenweider R.A., Kerekes J. (Eds.), 1982. *Eutrophication of Waters: Monitoring, Assessment and Control*. Report of the OECD cooperative programme on eutrophication. Organisation for the Economic Development and Co-operation. Paris, 154 pp.
- Vollenweider, R.A., Giovanardi, F., Montanari, G., Rinaldi, A., 1998. Characterization of the trophic conditions of marine coastal waters with special reference to the NW Adriatic Sea: Proposal for a trophic scale, turbidity and generalized water quality index. *Environmetrics* 9, 329-357.
- Warwick, R.M., Platt, H.M., Clarke, K.R., Agard, J., Gobin, J., 1990. Analysis of macrobenthic and meiobenthic community structure in relation to pollution and disturbance in Hamilton Harbour, Bermuda. *Journal of Experimental Marine Biology and Ecology* 138, 119-142.
- Wehr, J.D., Descy, J.P., 1998. Use of phytoplankton in large river management. *Journal of Phycology* 34, 741-749.
- Welch, E.B., 1992. *Ecological effects of wastewater: Applied limnology and pollutant effects*. 2<sup>nd</sup> ed. Chapman & Hall, London, 423 pp.
- Weston, D.P., 1990. Qualitative examination of macrobenthic community changes along an organic enrichment gradient. *Marine Ecology Progress Series* 61, 233-244.
- Wetzel, R., 1983. *Limnologia*. 2<sup>nd</sup> ed. Fundação Calouste Gulbenkian, Lisboa, 919 pp.
- Witek, B., Plínski, M., 2000. The first recorded bloom of *Prorocentrum minimum* (Pavillard) Schiller in the coastal zone of the Gulf of Gdańsk. *Oceanologica Part I* 42, 29-36.
- Wolf, B., Kiel, E., Hagge, A., Krieg, H-J., Feld, C.K., 2009. Using salinity preferences of benthic macroinvertebrates to classify running waters in brackish marshes in Germany. *Ecological Indicators* 9, 837-847.

## APPENDICES

Appendix I.A - Results of One-Way ANOVA. Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for water parameters (Ln (x+1)) in Foz de Almagem coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Temperature	ANOVA Test: $F = 0.024; p = 0.976$ LSD Fisher Test: MD (Upstream-Downstream) = 0.023; $p = 0.833$ MD (Upstream-Intermediate) = 0.016; $p = 0.882$ MD (Intermediate-Downstream) = 0.007; $p = 0.109$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Salinity	ANOVA Test: $F = 0.936; p = 0.408$ LSD Fisher Test: MD (Intermediate-Upstream) = 0.336; $p = 0.235$ MD (Downstream-Upstream) = 0.314; $p = 0.266$ MD (Intermediate-Downstream) = 0.022; $p = 0.938$	<u>Non Significant Difference:</u> Intermediate > Upstream Downstream > Upstream Intermediate > Downstream
pH	Kruskal-Wallis Test: $\chi^2 = 1.998; d.f. = 2; p = 0.368$ LSD Fisher Test: MRD (Downstream-Upstream) = 4.875; $p = 0.182$ MRD (Downstream-Intermediate) = 3.375; $p = 0.350$ MRD (Intermediate-Upstream) = 1.500; $p = 0.675$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
Total Solids in Suspension	ANOVA Test: $F = 0.189; p = 0.830$ LSD Fisher Test: MD (Downstream-Upstream) = 0.290; $p = 0.549$ MD (Intermediate-Upstream) = 0.178; $p = 0.713$ MD (Downstream-Intermediate) = 0.112; $p = 0.816$	<u>Non Significant Difference:</u> Downstream > Upstream Intermediate > Upstream Downstream > Intermediate
Dissolved Oxygen concentration	ANOVA Test: $F = 2.982; p = 0.072$ LSD Fisher Test: MD (Downstream-Upstream) = 0.172; $p = 0.036^*$ MD (Downstream-Intermediate) = 0.152; $p = 0.062$ MD (Intermediate-Upstream) = 0.021; $p = 0.791$	<b>* Significant Difference:</b> <b>Downstream &gt; Upstream</b> <u>Non Significant Difference:</u> Downstream > Intermediate Intermediate > Upstream
Nitrates concentration	ANOVA Test: $F = 0.900; p = 0.422$ LSD Fisher Test: MD (Upstream-Downstream) = 0.994; $p = 0.236$ MD (Upstream-Intermediate) = 0.890; $p = 0.287$ MD (Intermediate-Downstream) = 0.104; $p = 0.900$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Nitrites concentration	ANOVA Test: $F = 1.370; p = 0.276$ LSD Fisher Test: MD (Upstream-Intermediate) = 0.225; $p = 0.160$ MD (Upstream-Downstream) = 0.218; $p = 0.173$ MD (Downstream-Intermediate) = 0.007; $p = 0.966$	<u>Non Significant Difference:</u> Upstream > Intermediate Upstream > Downstream Downstream > Intermediate
Ammonia concentration	ANOVA Test: $F = 3.097; p = 0.066$ LSD Fisher Test: MD (Upstream-Intermediate) = 0.740; $p = 0.027^*$ MD (Upstream-Downstream) = 0.565; $p = 0.083$ MD (Downstream-Intermediate) = 0.174; $p = 0.580$	<b>* Significant Difference:</b> <b>Upstream &gt; Intermediate</b> <u>Non Significant Difference:</u> Upstream > Downstream Downstream-Intermediate
Total dissolved inorganic nitrogen concentration	ANOVA Test: $F = 2.221; p = 0.133$ LSD Fisher Test: MD (Upstream-Downstream) = 1.085; $p = 0.080$ MD (Upstream-Intermediate) = 1.066; $p = 0.085$ MD (Intermediate-Downstream) = 0.019; $p = 0.975$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Orthophosphates concentration	ANOVA Test: $F = 0.063; p = 0.940$ LSD Fisher Test: MD (Intermediate-Upstream) = 0.056; $p = 0.744$ MD (Intermediate-Downstream) = 0.047; $p = 0.787$ MD (Downstream-Upstream) = 0.010; $p = 0.955$	<u>Non Significant Difference:</u> Intermediate > Upstream Intermediate > Downstream Downstream > Upstream
Total Phosphorus concentration	ANOVA Test: $F = 0.031; p = 0.969$ LSD Fisher Test: MD (Intermediate-Upstream) = 0.040; $p = 0.828$ MD (Downstream-Upstream) = 0.039; $p = 0.834$ MD (Intermediate-Downstream) = 0.001; $p = 0.994$	<u>Non Significant Difference:</u> Intermediate > Upstream Downstream > Upstream Intermediate > Downstream
N: P ratio	ANOVA Test: $F = 1.624; p = 0.221$ LSD Fisher Test: MD (Upstream-Intermediate) = 1.028; $p = 0.125$ MD (Upstream-Downstream) = 0.975; $p = 0.144$ MD (Downstream-Intermediate) = 0.053; $p = 0.935$	<u>Non Significant Difference:</u> Upstream > Intermediate Upstream > Downstream Downstream > Intermediate
Chlorophyll <i>a</i> concentration	ANOVA Test: $F = 0.104; p = 0.902$ LSD Fisher Test: MD (Upstream-Downstream) = 0.172; $p = 0.660$ MD (Upstream-Intermediate) = 0.116; $p = 0.767$ MD (Intermediate-Downstream) = 0.056; $p = 0.885$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Phaeo-pigments concentration	Kruskal-Wallis Test: $\chi^2 = 1.940; d.f. = 2; p = 0.379$ LSD Fisher Test: MRD (Upstream-Intermediate) = 4.750; $p = 0.194$ MRD (Upstream-Downstream) = 3.500; $p = 0.334$ MRD (Downstream-Intermediate) = 1.250; $p = 0.728$	<u>Non Significant Difference:</u> Upstream > Intermediate Upstream > Downstream Downstream > Intermediate
Pigments diversity index	ANOVA Test: $F = 0.085; p = 0.919$ LSD Fisher Test: MD (Downstream-Intermediate) = 0.017; $p = 0.699$ MD (Downstream-Upstream) = 0.013; $p = 0.762$ MD (Upstream-Intermediate) = 0.004; $p = 0.933$	<u>Non Significant Difference:</u> Downstream > Intermediate Downstream > Upstream Upstream > Intermediate

Appendix I.B - Results of One-Way ANOVA. Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for sediment parameters (Ln x+1) in Foz de Almagem coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Clay content	Kruskal-Wallis Test: $\chi^2 = 13.936$ ; d.f.= 2; $p = 0.001$ LSD Fisher Test: MRD (Intermediate-Downstream) = 11.714; $p = 0.000^*$ MRD (Upstream-Downstream) = 9.286; $p = 0.000^*$ MRD (Intermediate-Upstream) = 2.429; $p = 0.223$	* <b>Significant Difference:</b> <b>Intermediate &gt; Downstream</b> Upstream > Downstream <u>Non Significant Difference:</u> Intermediate > Upstream
Silt content	Kruskal-Wallis Test: $\chi^2 = 15.677$ ; d.f.= 2; $p = 0.000$ LSD Fisher Test: MRD (Intermediate-Downstream) = 13.000; $p = 0.000^*$ MRD (Upstream-Downstream) = 8.000; $p = 0.000^*$ MRD (Intermediate-Upstream) = 5.000; $p = 0.006^*$	* <b>Significant Difference:</b> <b>Intermediate &gt; Downstream</b> <b>Upstream &gt; Downstream</b> <b>Intermediate &gt; Upstream</b>
Sand content	Kruskal-Wallis Test: $\chi^2 = 115.677$ ; d.f.= 2; $p = 0.000$ LSD Fisher Test: MRD (Downstream-Intermediate) = 13.000; $p = 0.000^*$ MRD (Downstream-Upstream) = 8.000; $p = 0.000^*$ MRD (Upstream-Intermediate) = 5.000; $p = 0.006^*$	* <b>Significant Difference:</b> <b>Downstream &gt; Intermediate</b> <b>Downstream &gt; Upstream</b> <b>Upstream &gt; Intermediate</b>
Water content	ANOVA Test: $F = 5.315$ ; $p = 0.015$ LSD Fisher Test: MD (Intermediate-Downstream) = 0.357; $p = 0.004^*$ MD (Upstream-Downstream) = 0.197; $p = 0.089$ MD (Intermediate-Upstream) = 0.160; $p = 0.162$	* <b>Significant Difference:</b> <b>Intermediate &gt; Downstream</b> <u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Upstream
Organic matter content	Kruskal-Wallis Test: $\chi^2 = 15.904$ ; d.f.= 2; $p = 0.000$ LSD Fisher Test: MRD (Intermediate-Downstream) = 13.143; $p = 0.000^*$ MRD (Upstream-Downstream) = 7.857; $p = 0.000^*$ MRD (Intermediate-Upstream) = 5.286; $p = 0.000^*$	* <b>Significant Difference:</b> <b>Intermediate &gt; Downstream</b> <b>Upstream &gt; Downstream</b> <b>Intermediate &gt; Upstream</b>
Chlorophyll a concentration	ANOVA Test: $F = 4.429$ ; $p = 0.027$ LSD Fisher Test: MD (Upstream-Downstream) = 8.996; $p = 0.009^*$ MD (Upstream-Intermediate) = 5.942; $p = 0.069$ MD (Intermediate-Downstream) = 3.053; $p = 0.334$	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b> <u>Non Significant Difference:</u> Upstream > Intermediate Intermediate > Downstream
Phaeo-pigments concentration	ANOVA Test: $F = 3.900$ ; $p = 0.039$ LSD Fisher Test: MD (Upstream-Downstream) = 9.880; $p = 0.015^*$ MD (Intermediate-Downstream) = 7.495; $p = 0.057$ MD (Upstream-Intermediate) = 2.384; $p = 0.527$	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b> <u>Non Significant Difference:</u> Intermediate > Downstream Upstream > Intermediate
Chlorophyll a degradation index	ANOVA Test: $F = 4.702$ ; $p = 0.023$ LSD Fisher Test: MD (Intermediate-Downstream) = 21.288; $p = 0.007^*$ MD (Upstream-Downstream) = 12.798; $p = 0.084$ MD (Intermediate-Upstream) = 8.491; $p = 0.240$	* <b>Significant Difference:</b> <b>Intermediate &gt; Downstream</b> <u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Upstream
Pigment diversity index	Kruskal-Wallis Test: $\chi^2 = 11.273$ ; d.f.= 2; $p = 0.004$ LSD Fisher Test: MRD (Upstream-Downstream) = 9.571; $p = 0.001^*$ MRD (Intermediate -Downstream) = 9.714; $p = 0.001^*$ MRD (Intermediate-Upstream) = 0.143; $p = 0.951$	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b> <b>Intermediate &gt; Downstream</b> <u>Non Significant Difference:</u> Intermediate > Upstream

Appendix I.C - Results of One-Way ANOVA. Kruskal-Wallis test and multiple comparisons LSD Fisher test among sites for water parameters (Ln (x+1)) in Salgados coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Temperature	ANOVA Test: $F = 0.002$ ; $p = 0.998$ LSD Fisher Test: MD (Intermediate-Downstream) = 0.005; $p = 0.956$ MD (Upstream-Downstream) = 0.004; $p = 0.967$ MD (Intermediate-Upstream) = 0.001; $p = 0.989$	<u>Non Significant Difference:</u> Intermediate > Downstream Upstream > Downstream Intermediate > Upstream
Salinity	ANOVA Test: $F = 0.816$ ; $p = 0.456$ LSD Fisher Test: MD (Downstream-Upstream) = 0.318; $p = 0.231$ MD (Downstream-Intermediate) = 0.233; $p = 0.377$ MD (Intermediate-Upstream) = 0.086; $p = 0.743$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
pH	ANOVA Test: $F = 1.937$ ; $p = 0.169$ LSD Fisher Test: MD (Upstream-Intermediate) = 0.058; $p = 0.072$ MD (Downstream-Intermediate) = 0.043; $p = 0.175$ MD (Upstream-Downstream) = 0.015; $p = 0.626$	<u>Non Significant Difference:</u> Upstream > Intermediate Downstream > Intermediate Upstream > Downstream
Total Solids in Suspension	ANOVA Test: $F = 2.021$ ; $p = 0.157$ LSD Fisher Test: MD (Upstream-Downstream) = 0.830; $p = 0.069$ MD (Intermediate-Downstream) = 0.646; $p = 0.151$ MD (Upstream-Intermediate) = 0.184; $p = 0.675$	<u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Downstream Upstream > Intermediate
Dissolved Oxygen concentration	ANOVA Test: $F = 0.586$ ; $p = 0.565$ LSD Fisher Test: MD (Upstream-Intermediate) = 0.223; $p = 0.299$ MD (Upstream-Downstream) = 0.146; $p = 0.492$ MD (Downstream-Intermediate) = 0.076; $p = 0.719$	<u>Non Significant Difference:</u> Upstream > Intermediate Upstream > Downstream Downstream > Intermediate
Nitrates concentration	Kruskal-Wallis Test: $\chi^2 = 1.580$ ; d.f.= 2; $p = 0.454$ LSD Fisher Test: MRD (Downstream-Upstream) = 4.438; $p = 0.227$ MRD (Downstream-Intermediate) = 2.313; $p = 0.524$ MRD (Intermediate-Upstream) = 2.125; $p = 0.558$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
Nitrites concentration	Kruskal-Wallis Test: $\chi^2 = 1.037$ ; d.f.= 2; $p = 0.596$ LSD Fisher Test: MRD (Downstream-Upstream) = 3.313; $p = 0.370$ MRD (Downstream-Intermediate) = 2.875; $p = 0.435$ MRD (Intermediate-Upstream) = 0.438; $p = 0.905$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
Ammonia concentration	ANOVA Test: $F = 0.456$ ; $p = 0.640$ LSD Fisher Test: MD (Upstream-Downstream) = 0.740; $p = 0.353$ MD (Upstream-Intermediate) = 0.430; $p = 0.587$ MD (Intermediate-Downstream) = 0.310; $p = 0.694$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Total dissolved inorganic nitrogen concentration	ANOVA Test: $F = 0.003$ ; $p = 0.997$ LSD Fisher Test: MD (Upstream-Intermediate) = 0.053; $p = 0.942$ MD (Upstream-Downstream) = 0.031; $p = 0.965$ MD (Downstream-Intermediate) = 0.021; $p = 0.976$	<u>Non Significant Difference:</u> Upstream > Intermediate Upstream > Downstream Downstream > Intermediate
Orthophosphates concentration	ANOVA Test: $F = 1.292$ ; $p = 0.296$ LSD Fisher Test: MD (Intermediate-Upstream) = 0.456; $p = 0.125$ MD (Intermediate-Downstream) = 0.275; $p = 0.348$ MD (Downstream-Upstream) = 0.183; $p = 0.531$	<u>Non Significant Difference:</u> Intermediate > Upstream Intermediate > Downstream Downstream > Upstream
Total Phosphorus concentration	ANOVA Test: $F = 0.965$ ; $p = 0.397$ LSD Fisher Test: MD (Intermediate-Upstream) = 0.390; $p = 0.199$ MD (Intermediate-Downstream) = 0.299; $p = 0.321$ MD (Downstream-Upstream) = 0.091; $p = 0.759$	<u>Non Significant Difference:</u> Intermediate > Upstream Intermediate > Downstream Downstream > Upstream
N: P ratio	Kruskal-Wallis Test: $\chi^2 = 0.420$ ; d.f.= 2; $p = 0.811$ LSD Fisher Test: MRD (Upstream-Intermediate) = 2.250; $p = 0.546$ MRD (Downstream-Intermediate) = 1.500; $p = 0.687$ MRD (Upstream-Downstream) = 0.750; $p = 0.840$	<u>Non Significant Difference:</u> Upstream > Intermediate Downstream > Intermediate Upstream > Downstream
Chlorophyll <i>a</i> concentration	ANOVA Test: $F = 1.312$ ; $p = 0.290$ LSD Fisher Test: MD (Upstream-Downstream) = 0.988; $p = 0.120$ MD (Upstream-Intermediate) = 0.515; $p = 0.408$ MD (Intermediate-Downstream) = 0.473; $p = 0.447$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Phaeo-pigments concentration	Kruskal-Wallis Test: $\chi^2 = 0.635$ ; d.f.= 2; $p = 0.728$ LSD Fisher Test: MRD (Intermediate-Downstream) = 2.500; $p = 0.501$ MRD (Upstream-Downstream) = 2.375; $p = 0.522$ MRD (Intermediate-Upstream) = 0.125; $p = 0.973$	<u>Non Significant Difference:</u> Intermediate > Downstream Upstream > Downstream Intermediate > Upstream
Pigments diversity index	Kruskal-Wallis Test: $\chi^2 = 3.788$ ; d.f.= 2; $p = 0.150$ LSD Fisher Test: MRD (Upstream-Downstream) = 6.750; $p = 0.059$ MRD (Intermediate-Downstream) = 4.500; $p = 0.197$ MRD (Upstream-Intermediate) = 2.250; $p = 0.513$	<u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Downstream Upstream > Intermediate

Appendix I.D - Results of Mann-Whitney U test and Student T test for sediment parameters (Ln (x+1)) comparison between sites from Salgados lagoon. \*- Significant at the 0.05 level; \*\*- Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Clay content (% Clay)	Mann-Whitney U Test: $U = 0.000$ ; $Z = -3.363$ ; $p = 0.001$ ** Mean Rank Upstream = 12.50 Mean Rank Downstream = 4.50	** <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
Silt content (%Silt)	Mann-Whitney U Test: $U = 0.000$ ; $Z = -3.361$ ; $p = 0.001$ ** Mean Rank Upstream = 12.50 Mean Rank Downstream = 4.50	** <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
Sand content (%Sand)	Mann-Whitney U Test: $U = 0.000$ ; $Z = -3.361$ ; $p = 0.001$ ** Mean Rank Upstream = 4.50 Mean Rank Downstream = 12.50	** <b>Significant Difference:</b> <b>Downstream &gt; Upstream</b>
Water content (%WSed)	Mann-Whitney U Test: $U = 29.000$ ; $Z = -0.315$ ; $p = 0.753$ Mean Rank Upstream = 8.13 Mean Rank Downstream = 8.81	<u>Non Significant Difference:</u> Downstream > Upstream
Organic matter content (%OM)	Mann-Whitney U Test: $U = 0.000$ ; $Z = -3.361$ ; $p = 0.001$ ** Mean Rank Upstream = 12.50 Mean Rank Downstream = 4.50	** <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
Chlorophyll <i>a</i> concentration (ChlaS)	Student-T Test: $T = 1.529$ ; $df = 14$ ; $p = 0.149$ Mean Difference = 0.708	<u>Non Significant Difference:</u> Upstream > Downstream
Phaeo-pigments concentration (PhaeS)	Student-T Test: $T = 3.245$ ; $df = 14$ ; $p = 0.006$ ** Mean Difference = 1.276	** <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
Chlorophyll <i>a</i> degradation index (%PhaeS)	Mann-Whitney U Test: $U = 11.000$ ; $Z = -2.205$ ; $p = 0.027$ * Mean Rank Upstream = 11.13 Mean Rank Downstream = 5.88	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
Pigment diversity index (PigDS)	Student-T Test: $T = 2.156$ ; $df = 14$ ; $p = 0.049$ * Mean Difference = 0.092	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>

Appendix I.E - Results from Mann-Whitney U test and Student T test for water parameters (Ln (x+1)) comparison between lagoons. \*- Significant at the 0.05 level; \*\*- Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Temperature	Mann-Whitney U Test: $U = 253.0$ ; $Z = -0.722$ ; $p = 0.470$ Mean Rank Foz de Almagem = 25.96 Mean Rank Salgados = 23.04	<u>Non Significant Difference:</u> Foz de Almagem > Salgados
Salinity	Student-T Test: $T = 2.784$ ; $df = 46$ ; $p = 0.008$ ** Mean Difference = 0.432	** <u>Significant Difference:</u> Foz de Almagem > Salgados
pH	Student-T Test: $T = -2.060$ ; $df = 46$ ; $p = 0.045$ * Mean Difference = -0.032	* <u>Significant Difference:</u> Foz de Almagem < Salgados
Total Solids in Suspension	Student-T Test: $T = -2.919$ ; $df = 46$ ; $p = 0.005$ ** Mean Difference = -0.768	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Dissolved Oxygen concentration	Student-T Test: $T = -0.524$ ; $df = 46$ ; $p = 0.604$ Mean Difference = -0.047	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Nitrates concentration	Mann-Whitney U Test: $U = 269.0$ ; $Z = -0.392$ ; $p = 0.695$ Mean Rank Foz de Almagem = 23.71 Mean Rank Salgados = 25.29	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Nitrites concentration	Mann-Whitney U Test: $U = 154.0$ ; $Z = -2.764$ ; $p = 0.006$ ** Mean Rank Foz de Almagem = 18.92 Mean Rank Salgados = 30.08	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Ammonia concentration	Student-T Test: $T = -4.652$ ; $df = 46$ ; $p = 0.000$ ** Mean Difference = -1.579	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Total dissolved inorganic nitrogen concentration	Mann-Whitney U Test: $U = 185.0$ ; $Z = -2.124$ ; $p = 0.034$ * Mean Rank Foz de Almagem = 20.21 Mean Rank Salgados = 28.79	* <u>Significant Difference:</u> Foz de Almagem < Salgados
Orthophosphates concentration	Student-T Test: $T = -24.063$ ; $df = 46$ ; $p = 0.000$ ** Mean Difference = -3.268	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Total Phosphorus concentration	Student-T Test: $T = -24.460$ ; $df = 46$ ; $p = 0.000$ ** Mean Difference = -3.264	** <u>Significant Difference:</u> Foz de Almagem < Salgados
N: P ratio	Mann-Whitney U Test: $U = 29.0$ ; $Z = -5.341$ ; $p = 0.000$ ** Mean Rank Foz de Almagem = 35.29 Mean Rank Salgados = 13.71	** <u>Significant Difference:</u> Foz de Almagem > Salgados
Chlorophyll <i>a</i> concentration	Mann-Whitney U Test: $U = 1.0$ ; $Z = -5.918$ ; $p = 0.000$ ** Mean Rank Foz de Almagem = 12.54 Mean Rank Salgados = 36.46	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Phaeo-pigments concentration	Mann-Whitney U Test: $U = 104.0$ ; $Z = -3.795$ ; $p = 0.000$ ** Mean Rank Foz de Almagem = 16.83 Mean Rank Salgados = 32.17	** <u>Significant Difference:</u> Foz de Almagem < Salgados
Pigments diversity index	Mann-Whitney U Test: $U = 137.0$ ; $Z = -3.114$ ; $p = 0.002$ ** Mean Rank Foz de Almagem = 30.79 Mean Rank Salgados = 18.21	** <u>Significant Difference:</u> Foz de Almagem > Salgados

Appendix I.F - Results of Mann-Whitney U test and Student T test for sediment parameters (Ln (x+1)) comparison between lagoons. \*- Significant at the 0.05 level; \*\*- Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Clay content (% Clay)	Mann-Whitney U Test: $U = 137.0$ ; $Z = -0.950$ ; $p = 0.342$ Mean Rank Foz de Almagem = 17.52 Mean Rank Salgados = 20.94	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Silt content (%Silt)	Mann-Whitney U Test: $U = 164.0$ ; $Z = 0.123$ ; $p = 0.902$ Mean Rank Foz de Almagem = 18.81 Mean Rank Salgados = 19.25	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Sand content (%Sand)	Mann-Whitney U Test: $U = 149.0$ ; $Z = -0.583$ ; $p = 0.560$ Mean Rank Foz de Almagem = 19.90 Mean Rank Salgados = 17.81	<u>Non Significant Difference:</u> Foz de Almagem > Salgados
Water content (%WSed)	Mann-Whitney U Test: $U = 145.0$ ; $Z = -0.705$ ; $p = 0.481$ Mean Rank Foz de Almagem = 20.10 Mean Rank Salgados = 17.56	<u>Non Significant Difference:</u> Foz de Almagem > Salgados
Organic matter content (%OM)	Mann-Whitney U Test: $U = 153.0$ ; $Z = -0.460$ ; $p = 0.646$ Mean Rank Foz de Almagem = 18.29 Mean Rank Salgados = 19.94	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Chlorophyll <i>a</i> concentration (ChlaS)	Student-T Test: $T = -2.272$ ; $df = 35$ ; $p = 0.029$ * Mean Difference = -0.670	<b>*Significant Difference:</b> <b>Foz de Almagem &lt; Salgados</b>
Phaeo-pigments concentration (PhaeS)	Student-T Test: $T = -1.103$ ; $df = 35$ ; $p = 0.278$ Mean Difference = -0.360	<u>Non Significant Difference:</u> Foz de Almagem < Salgados
Chlorophyll <i>a</i> degradation index (%PhaeS)	Mann-Whitney U Test: $U = 119.0$ ; $Z = -1.502$ ; $p = 0.133$ Mean Rank Foz de Almagem = 21.33 Mean Rank Salgados = 15.94	<u>Non Significant Difference:</u> Foz de Almagem > Salgados
Pigment diversity index (PigDS)	Student-T Test: $T = 2.270$ ; $df = 35$ ; $p = 0.030$ * Mean Difference = 0.088	<b>* Significant Difference:</b> <b>Foz de Almagem &gt; Salgados</b>

Appendix I.G - Results of One-Way ANOVA and Kruskal-Wallis test among sites for phytoplankton parameters (Ln (x+1)) in Foz de Almagem coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Total phytoplankton abundance	ANOVA Test: $F = 0.042$ ; $p = 0.959$	No Significant Difference
Taxonomic richness	ANOVA Test: $F = 0.108$ ; $p = 0.898$	No Significant Difference
Shannon-Wiener diversity	Kruskal-Wallis Test: $\chi^2 = 1.085$ ; $d.f. = 2$ ; $p = 0.581$	No Significant Difference
Evenness	Kruskal-Wallis Test: $\chi^2 = 1.040$ ; $d.f. = 2$ ; $p = 0.595$	No Significant Difference
Bacillariophyceae abundance	ANOVA Test: $F = 0.217$ ; $p = 0.807$	No Significant Difference
Cryptophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 0.135$ ; $d.f. = 2$ ; $p = 0.935$	No Significant Difference
Dinophyceae abundance	ANOVA Test: $F = 0.006$ ; $p = 0.994$	No Significant Difference
Euglenophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 0.283$ ; $d.f. = 2$ ; $p = 0.868$	No Significant Difference
Pico-nano flagellate algae abundance	Kruskal-Wallis Test: $\chi^2 = 0.422$ ; $d.f. = 2$ ; $p = 0.810$	No Significant Difference

Appendix I.H - Results of One-Way ANOVA and Kruskal-Wallis test among sites for phytoplankton parameters (Ln (x+1)) in Salgados coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Total phytoplankton abundance	ANOVA Test: $F = 0.004$ ; $p = 0.996$	No Significant Difference
Taxonomic richness	ANOVA Test: $F = 0.710$ ; $p = 0.503$	No Significant Difference
Shannon-Wiener diversity	ANOVA Test: $F = 0.080$ ; $p = 0.924$	No Significant Difference
Evenness	ANOVA Test: $F = 0.032$ ; $p = 0.968$	No Significant Difference
Chlorophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 0.005$ ; $d.f. = 2$ ; $p = 0.997$	No Significant Difference
Bacillariophyceae abundance	ANOVA Test: $F = 0.252$ ; $p = 0.779$	No Significant Difference
Cryptophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 0.291$ ; $d.f. = 2$ ; $p = 0.865$	No Significant Difference
Dinophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 2.399$ ; $d.f. = 2$ ; $p = 0.301$	No Significant Difference
Euglenophyceae abundance	Kruskal-Wallis Test: $\chi^2 = 0.302$ ; $d.f. = 2$ ; $p = 0.860$	No Significant Difference
Cyanophyceae abundance	ANOVA Test: $F = 0.002$ ; $p = 0.998$	No Significant Difference
Pico-nano flagellate algae abundance	Kruskal-Wallis Test: $\chi^2 = 0.000$ ; $d.f. = 2$ ; $p = 1.000$	No Significant Difference

Appendix I.I - Results of Mann-Whitney U test and Student T test for phytoplankton parameters (Ln (x+1)) comparison between lagoons. \*- Significant at the 0.05 level; \*\*- Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Total phytoplankton abundance	Student-T Test: $T = -7.533$ ; $df = 46$ ; $p = 0.000$ ** Mean Difference = -3.66	<b>**Significant Difference:</b> Foz de Almargem < Salgados
Taxonomic richness	Student-T Test: $T = 1.390$ ; $df = 46$ ; $p = 0.173$ Mean Difference = 0.17	Non Significant Difference: Foz de Almargem > Salgados
Shannon-Wiener diversity	Student-T Test: $T = 1.257$ ; $df = 46$ ; $p = 0.215$ Mean Difference = 0.12	Non Significant Difference: Foz de Almargem > Salgados
Evenness	Mann-Whitney U Test: $U = 263.5$ ; $Z = -0.505$ ; $p = 0.613$ Mean Rank Foz de Almargem = 25.52 Mean Rank Salgados = 23.48	Non Significant Difference: Foz de Almargem > Salgados
Chlorophyceae abundance	Mann-Whitney U Test: $U = 31.5$ ; $Z = -5.554$ ; $p = 0.000$ ** Mean Rank Foz de Almargem = 13.81 Mean Rank Salgados = 35.19	<b>**Significant Difference:</b> Foz de Almargem < Salgados
Bacillariophyceae abundance	Mann-Whitney U Test: $U = 128.0$ ; $Z = -3.302$ ; $p = 0.001$ ** Mean Rank Foz de Almargem = 17.83 Mean Rank Salgados = 31.17	<b>**Significant Difference:</b> Foz de Almargem < Salgados
Cryptophyceae abundance	Mann-Whitney U Test: $U = 179.0$ ; $Z = -2.285$ ; $p = 0.022$ * Mean Rank Foz de Almargem = 19.96 Mean Rank Salgados = 29.04	<b>*Significant Difference:</b> Foz de Almargem < Salgados
Dinophyceae abundance	Mann-Whitney U Test: $U = 139.0$ ; $Z = -3.121$ ; $p = 0.002$ ** Mean Rank Foz de Almargem = 30.71 Mean Rank Salgados = 18.29	<b>**Significant Difference:</b> Foz de Almargem > Salgados
Euglenophyceae abundance	Mann-Whitney U Test: $U = 111.0$ ; $Z = -3.717$ ; $p = 0.000$ ** Mean Rank Foz de Almargem = 17.13 Mean Rank Salgados = 3.88	<b>**Significant Difference:</b> Foz de Almargem < Salgados
Cyanophyceae abundance	Mann-Whitney U Test: $U = 3.0$ ; $Z = -6.139$ ; $p = 0.000$ ** Mean Rank Foz de Almargem = 12.63 Mean Rank Salgados = 36.38	<b>**Significant Difference:</b> Foz de Almargem < Salgados
Pico-nano flagellate algae abundance	Mann-Whitney U Test: $U = 234.0$ ; $Z = -1.152$ ; $p = 0.249$ Mean Rank Foz de Almargem = 22.25 Mean Rank Salgados = 26.75	Non Significant Difference: Foz de Almargem > Salgados

Appendix I.J - Results of One-Way ANOVA and Kruskal-Wallis and multiple comparisons LSD Fisher test among sites for benthic macroinvertebrate parameters (Ln (x+1)) in Foz de Almagem coastal lagoon.

	<b>Results</b>	<b>Conclusions</b>
Total benthos density	Kruskal-Wallis Test: $\chi^2 = 0.237$ ; d.f.= 2; $p = 0.792$ LSD Fisher Test: MRD (Downstream-Intermediate) = 0.430; $p = 0.508$ MRD (Downstream-Upstream) = 0.288; $p = 0.657$ MRD (Upstream-Intermediate) = 0.142; $p = 0.825$	<u>Non Significant Difference:</u> Downstream > Intermediate Downstream > Upstream Upstream > Intermediate
Taxonomic richness	ANOVA Test: $F = 0.396$ ; $p = 0.679$ LSD Fisher Test: MD (Upstream-Downstream) = 0.157; $p = 0.397$ MD (Upstream-Intermediate) = 0.110; $p = 0.551$ MD (Intermediate- Downstream) = 0.047; $p = 0.798$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
Shannon-Wiener diversity	ANOVA Test: $F = 4.061$ ; $p = 0.035$ LSD Fisher Test: MD (Upstream-Downstream) = 0.395; $p = 0.011^*$ MD (Intermediate-Downstream) = 0.223; $p = 0.126$ MD (Upstream-Intermediate) = 0.172; $p = 0.232$	* <u>Significant Difference:</u> <b>Upstream &gt; Downstream</b> <u>Non Significant Difference:</u> Intermediate > Downstream Upstream > Intermediate
Evenness	ANOVA Test: $F = 3.277$ ; $p = 0.061$ LSD Fisher Test: MD (Upstream-Downstream) = 0.208; $p = 0.022^*$ MD (Intermediate-Downstream) = 0.142; $p = 0.105$ MD (Upstream-Intermediate) = 0.066; $p = 0.436$	* <u>Significant Difference:</u> <b>Upstream &gt; Downstream</b> <u>Non Significant Difference:</u> Intermediate > Downstream Upstream > Intermediate
INSECTA density <i>Chironomus</i> sp.	Kruskal-Wallis Test: $\chi^2 = 5.258$ ; d.f.= 2; $p = 0.072$ LSD Fisher Test: MRD (Intermediate-Downstream) = 7.500; $p = 0.021^*$ MRD (Intermediate-Upstream) = 4.286; $p = 0.166$ MRD (Upstream-Downstream) = 3.214; $p = 0.293$	* <u>Significant Difference:</u> <b>Intermediate &gt; Downstream</b> <u>Non Significant Difference:</u> Intermediate > Upstream Upstream > Downstream
POLYCHAETA density	Kruskal-Wallis Test: $\chi^2 = 1.381$ ; d.f.= 2; $p = 0.501$ LSD Fisher Test: MRD (Upstream-Downstream) = 3.857; $p = 0.263$ MRD (Upstream-Intermediate) = 1.929; $p = 0.571$ MRD (Intermediate-Downstream) = 1.725; $p = 0.602$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
<i>Hediste diversicolor</i>	Kruskal-Wallis Test: $\chi^2 = 1.205$ ; d.f.= 2; $p = 0.547$ LSD Fisher Test: MRD (Upstream-Downstream) = 3.571; $p = 0.301$ MRD (Intermediate-Downstream) = 2.214; $p = 0.518$ MRD (Upstream-Intermediate) = 1.357; $p = 0.691$	<u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Downstream Upstream > Intermediate
ISOPODA density <i>Lekanesphaera hookeri</i>	Kruskal-Wallis Test: $\chi^2 = 0.571$ ; d.f.= 2; $p = 0.751$ LSD Fisher Test: MRD (Upstream-Downstream) = 2.500; $p = 0.477$ MRD (Upstream-Intermediate) = 1.357; $p = 0.698$ MRD (Intermediate-Downstream) = 1.143; $p = 0.744$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
GASTROPODA density	Kruskal-Wallis Test: $\chi^2 = 0.868$ ; d.f.= 2; $p = 0.648$ LSD Fisher Test: MRD (Downstream-Upstream) = 3.000; $p = 0.392$ MRD (Downstream-Intermediate) = 2.143; $p = 0.539$ MRD (Intermediate-Upstream) = 0.857; $p = 0.805$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
<i>Hydrobia ulvae</i>	Kruskal-Wallis Test: $\chi^2 = 0.868$ ; d.f.= 2; $p = 0.648$ LSD Fisher Test: MRD (Downstream-Upstream) = 3.000; $p = 0.392$ MRD (Downstream-Intermediate) = 2.143; $p = 0.539$ MRD (Intermediate-Upstream) = 0.857; $p = 0.805$	<u>Non Significant Difference:</u> Downstream > Upstream Downstream > Intermediate Intermediate > Upstream
BIVALVIA density	Kruskal-Wallis Test: $\chi^2 = 1.116$ ; d.f.= 2; $p = 0.572$ LSD Fisher Test: MRD (Upstream-Downstream) = 3.143; $p = 0.363$ MRD (Upstream-Intermediate) = 2.857; $p = 0.408$ MRD (Intermediate-Downstream) = 0.286; $p = 0.933$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream
<i>Abra segmentum</i>	Kruskal-Wallis Test: $\chi^2 = 1.616$ ; d.f.= 2; $p = 0.446$ LSD Fisher Test: MRD (Upstream-Downstream) = 3.643; $p = 0.265$ MRD (Intermediate-Downstream) = 3.214; $p = 0.323$ MRD (Upstream-Intermediate) = 0.429; $p = 0.894$	<u>Non Significant Difference:</u> Upstream > Downstream Intermediate > Downstream Upstream > Intermediate
<i>Cerastoderma glaucum</i>	Kruskal-Wallis Test: $\chi^2 = 840$ ; d.f.= 2; $p = 0.657$ LSD Fisher Test: MRD (Upstream-Downstream) = 2.857; $p = 0.388$ MRD (Upstream-Intermediate) = 1.643; $p = 0.617$ MRD (Intermediate-Downstream) = 1.214; $p = 0.711$	<u>Non Significant Difference:</u> Upstream > Downstream Upstream > Intermediate Intermediate > Downstream

Appendix I.K - Results of Mann-Whitney U test and Student T test for benthic macroinvertebrate parameters (Ln (x+1)) in Salgados coastal lagoon. . \*- Significant at the 0.05 level; \*\* - Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Total benthos density	Student-T Test: $T = -0.222$ ; $df = 14$ ; $p = 0.827$ Mean Difference = -0.21	<u>Non Significant Difference:</u> Upstream < Downstream
Taxonomic richness	Student-T Test: $T = -0.497$ ; $df = 14$ ; $p = 0.627$ Mean Difference = -0.08	<u>Non Significant Difference:</u> Upstream < Downstream
Shannon-Wiener diversity	Student-T Test: $T = -1.506$ ; $df = 14$ ; $p = 0.154$ Mean Difference = -0.26	<u>Non Significant Difference:</u> Upstream < Downstream
Evenness	Student-T Test: $T = -1.769$ ; $df = 14$ ; $p = 0.099$ Mean Difference = -0.16	<u>Non Significant Difference:</u> Upstream < Downstream
Total INSECTA density	Student-T Test: $T = 0.670$ ; $df = 14$ ; $p = 0.514$ Mean Difference = 0.64	<u>Non Significant Difference:</u> Upstream > Downstream
INSECTA Diptera density	Student-T Test: $T = 0.638$ ; $df = 14$ ; $p = 0.534$ Mean Difference = 0.62	<u>Non Significant Difference:</u> Upstream > Downstream
<i>Chironomus</i> sp.	Student-T Test: $T = 0.486$ ; $df = 14$ ; $p = 0.635$ Mean Difference = 0.49	<u>Non Significant Difference:</u> Upstream > Downstream
INSECTA Coleoptera density <i>Berosus spinosus</i>	Mann-Whitney U Test: $U = 11.5$ ; $Z = -2.304$ ; $p = 0.021$ * Mean Rank Upstream = 11.06 Mean Rank Downstream = 5.94	* <b>Significant Difference:</b> <b>Upstream &gt; Downstream</b>
INSECTA Hemiptera density	Mann-Whitney U Test: $U = 28.5$ ; $Z = -0.385$ ; $p = 0.701$ Mean Rank Upstream = 8.94 Mean Rank Downstream = 8.06	<u>Non Significant Difference:</u> Upstream > Downstream
OLIGOCHAETA density	Mann-Whitney U Test: $U = 19.0$ ; $Z = -1.506$ ; $p = 0.132$ Mean Rank Upstream = 6.88 Mean Rank Downstream = 10.13	<u>Non Significant Difference:</u> Upstream < Downstream
ISOPODA density <i>Lekanesphaera hookeri</i>	Mann-Whitney U Test: $U = 12.0$ ; $Z = -2.317$ ; $p = 0.021$ * Mean Rank Upstream = 6.00 Mean Rank Downstream = 11.00	* <b>Significant Difference:</b> <b>Upstream &lt; Downstream</b>

Appendix I.L - Results of Mann-Whitney U test and Student T test for benthic macroinvertebrate parameters (Ln (x+1)) comparison between lagoons. \*- Significant at the 0.05 level; \*\* - Significant at the 0.01 level.

	<b>Results</b>	<b>Conclusions</b>
Total benthos density	Mann-Whitney U Test: $U = 102.0$ ; $Z = -2.023$ ; $p = 0.043$ * Mean Rank Foz de Almargem = 22.14 Mean Rank Salgados = 14.88	* <b>Significant Difference:</b> <b>Foz de Almargem &gt; Salgados</b>
Taxonomic richness	Student-T Test: $T = 1.431$ ; $df = 35$ ; $p = 0.161$ Mean Difference = 0.150	<u>Non Significant Difference:</u> Foz de Almargem > Salgados
Shannon-Wiener diversity	Student-T Test: $T = -0.253$ ; $df = 35$ ; $p = 0.802$ Mean Difference = -0.028	<u>Non Significant Difference:</u> Foz de Almargem < Salgados
Evenness	Student-T Test: $T = -0.600$ ; $df = 35$ ; $p = 0.552$ Mean Difference = -0.036	<u>Non Significant Difference:</u> Foz de Almargem < Salgados
Total INSECTA density	Mann-Whitney U Test: $U = 40.0$ ; $Z = -3.934$ ; $p = 0.000$ ** Mean Rank Foz de Almargem = 12.90 Mean Rank Salgados = 27.00	** <b>Significant Difference:</b> <b>Foz de Almargem &lt; Salgados</b>
<i>Chironomus</i> sp.	Mann-Whitney U Test: $U = 53.0$ ; $Z = -3.535$ ; $p = 0.000$ ** Mean Rank Foz de Almargem = 13.52 Mean Rank Salgados = 26.19	** <b>Significant Difference:</b> <b>Foz de Almargem &lt; Salgados</b>
ISOPODA density <i>Lekanesphaera hookeri</i>	Mann-Whitney U Test: $U = 79.5$ ; $Z = -2.762$ ; $p = 0.006$ ** Mean Rank Foz de Almargem = 23.21 Mean Rank Salgados = 13.47	** <b>Significant Difference:</b> <b>Foz de Almargem &gt; Salgados</b>

Appendix II.A – List of phytoplankton *taxa* identified in Foz de Almargem coastal lagoon.

---

**CHLOROPHYCEAE**

*Cosmarinum* sp.

*Staurastrum* sp.

**BACILLARIOPHYCEAE**

*Asterionellopsis formosa*

*Aulacoseira granulate* (Ehrenberg) Simonsen 1979

*Cocconeis* sp.

*Cyclotella* spp.

*Cymbella* sp.

*Diploneis* sp.

*Fragilaria* sp. 1

*Fragilaria* sp. 2

*Gomphonema* sp.

*Grammatophora* sp.

*Gyrosigma* sp.

*Leptocylindrus danicus* Cleve, 1889

*Navicula* spp.

*Nitzschia acicularis* (Kützing) W. Smith, 1853

*Nitzschia* sp.

*Stauroneis* sp.

Unidentified diatoms

**CRYPTOPHYCEAE**

*Cryptomonas* sp.

*Rhodomonas* sp.

**DINOPHYCEAE**

*Gymnodinium* sp.

*Prorocentrum micans* Ehrenberg, 1833

*Prorocentrum minimum* (Pavillard) Schiller, 1933

*Protoperidinium* sp.

*Scrippsiella trochoidea* (Stein) Balech ex Loeblich III, 1965

**EUGLENOPHYCEAE**

*Eutreptiella* sp.

**CYANOPHYCEAE**

*Anabaena flos-aquae* (Lyngbye) Brebisson ex Bornet & Flauhault, 1886

**PICO-NANO FLAGELLATE ALGAE**

Unidentified pico-nano flagellate algae

---

Appendix II.B – List of phytoplankton *taxa* identified in Salgados coastal lagoon.

---

**CHLOROPHYCEAE**

*Ankistrodesmus acicularis* (Braun) Korshikov, 1953

*Ankistrodesmus falcatus* (Corda) Ralfs, 1848

*Chlamydomonas* sp.

*Coelastrum microporum* Nägeli, 1855

*Crucigenia quadrata* Morren, 1830

*Geminella interrupta* Turpin

*Oocystis lacustris* Chodat, 1897

*Scenedesmus acuminatus* (Lagerheim) Chodat, 1902

*Scenedesmus acutus* Meyen, 1829

*Scenedesmus opoliensis* P. Richter, 1897

*Selenastrum gracile* Reinsch, 1866

*Kirchneriella lunaris* (Kirchner) K. Möbius, 1897

*Kirchneriella obesa* (G.S. West) Schmidle, 1893

*Tetraedron muticum* (S. Braun) Hansgirg

**BACILLARIOPHYCEAE**

*Achnanthes* sp.

*Amphora* sp.

*Cyclotella* spp.

*Cymbella* sp.

*Gomphonema* sp.

*Navicula* spp.

*Nitzschia* sp.

**CRYPTOPHYCEAE**

*Cryptomonas* sp.

*Rhodomonas* sp.

**DINOPHYCEAE**

*Gymnodinium* sp.

**EUGLENOPHYCEAE**

*Euglena acus* Ehrenberg, 1830

*Euglena caudate* Hübner, 1886

*Euglena oblonga* F. Schmitz, 1884

*Phacus acuminatus* Stokes, 1885

*Trachelomonas* sp.

**CYANOPHYCEAE**

*Anabaena flos-aquae* (Lyngbye) Brebisson ex Bornet & Flauhault, 1886

*Anabaena spiroides* Klebahn, 1895

*Chroococcus limneticus* Lemmermann, 1898

*Chroococcus* sp.

*Lyngbya* sp.

*Merismopedia punctate* Meyen, 1839

*Microcystis aeruginosa* (Kützing) Kützing, 1846

*Microcystis* sp.

*Planktothrix* sp.

Unidentified Cyanophyceae

**PICO-NANO FLAGELLATE ALGAE**

Unidentified pico-nano flagellate algae

---

Appendix II.C – List of benthic macroinvertebrate *taxa* identified in Foz de Almagem coastal lagoon.

---

<b>ANNELIDA</b>
<b>Polychaeta</b>
<i>Capitella capitata</i> (Fabricius, 1780)
<i>Hediste diversicolor</i> (O.F.Müller, 1776)
Unidentified polychaeta
<b>Oligochaeta</b>
Unidentified oligochaeta
<b>CRUSTACEA</b>
<b>Isopoda</b>
<i>Lekanesphaera hookeri</i> (Leach, 1814)
<b>MOLLUSCA</b>
<b>Gastropoda</b>
<i>Ventrosia ventrosa</i> (Montagu, 1803)
<i>Hydrobia ulvae</i> (Pennant, 1777)
<b>Bivalvia</b>
<i>Abra segmentum</i> (Récluz, 1843)
<i>Cerastoderma glaucum</i> (Bruguière, 1789)
<b>INSECTA</b>
<b>Diptera</b>
<i>Chironomus</i> sp.

---

Appendix II.D – List of benthic macroinvertebrate *taxa* identified in Salgados coastal lagoon.

---

<b>ANNELIDA</b>
<b>Oligochaeta</b>
Tubificidae
Enchytraeidae
<b>CRUSTACEA</b>
<b>Isopoda</b>
<i>Lekanesphaera hookeri</i> (Leach, 1814)
<b>Amphipoda</b>
<i>Corophium multisetosum</i> (Stock, 1952)
<b>INSECTA</b>
<b>Diptera</b>
<i>Chironomus</i> sp.
Ephydriidae
Tabanidae
Ceratopogonidae
Empididae
Rhagionidae
Brachyceres
<b>Coleoptera</b>
<i>Berosus spinosus</i> (Steven, 1808)
<b>Hemiptera</b>
<i>Corixa affinis</i> (Leach, 1817)
<i>Hesperocorixa sahlbergi</i> (Fieber, 1848)
<i>Parasigara infuscata</i> (Rey, 1890)
<i>Notonecta</i> sp.

---