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Scientific and technical background for intercalibration of Danish coastal waters

NERI Tehnical Report, No. 563

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2005

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Datasheet

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Abstract:	This report contributes to the development of tools that can be applied to assess the five classes of ecological status of the Water Framework Directive based on the biological quality elements phytoplankton, macroalgae and macrobenthos. The work on phytoplankton biomass (Chl a) based on monitoring data and historical Secchi depth measurements provides a first step in establishing reference conditions for phytoplankton in Danish waters. The identification of appropriate macroalgal indicators provides the results that total cover and cumulated cover of coastal macroalgal communities are suitable indicators of water quality. The results also indicate that a strategy with focus on algae from deeper, light-limited waters and exclusion of algae from shallow exposed waters renderes algal indicators sensitive to changes in water quality. The macrobenthos project evaluates some of the earlier proposed indices on Danish data along the environmental gradient of oxygen deficiency. Results from this evaluation indicate that both diversity-based indices and the sensitivity based index AMBI can be used to evaluate quality status of benthic communities in Danish waters. The result applies to bottoms in the salinity regime > 18 psu.
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Preface

This report is part of a series of projects initiated by the Danish Environmental Protection Agency (EPA) – Water Unit dealing with the implementation of the Water Frame Work Directive. The Danish Environmental Protection Agency (EPA) – Water Unit has also funded the project though part of the work within Chapter 3 has been completed by financial support from EU (contract no. SSPI-CT-2003-502158 'REBECCA'). Jens Kjerulf Petersen (NERI) and Jesper H. Andersen (DHI Water & Environment) have been project managers.

Several reports from the Danish EPA projects have already been published - eg. Andersen et al. (2005) and Dahl et al. (2005). These two reports both cover work on eelgrass, which is therefore not presented in this report.

Apart from the data analysis and writing up this report the project has included participation in the work of the North East Atlantic Geographical Intercalibration Group (NEA GIG). The objective within the group is an attempt to intercalibrate biological quality elements to meet as a part of the requirements of the Water Framework Directive (WFD).

Furthermore, a 1D model describing the dynamics of water column stratification of the Danish fjords has been developed within this project. The model will be used in the future work as a tool to understand the natural variations in phytoplankton and macrobenthic fauna indicators.

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Summary

Based on analyses of the biological quality elements; phytoplankton, macroalgae and macrobenthos this report seeks to establish a scientific foundation to the development of tools that can be applied to assess the five classes of ecological status of the Water Frame Work Directive (WFD) in coastal waters.

The first part of this report has its focus on establishing preliminary reference conditions for phytoplankton biomass, expressed as chlorophyll *a*, using secchi depth data from the early 1900s and Secchi depth and Chl *a* obtained from recent monitoring data. The recent data were used to establish Secchi depth–Chl *a* relationships and correlations between Secchi depth and Chl *a* (90th percentiles of Chl *a* concentrations).

For all investigated areas a significant correlation was found between the Secchi depth and the Chl *a* concentrations and calculated 90th percentiles of historical Chl *a* concentrations were lower than recent ones. The outcome of these analyses was a first step in establishing reference conditions for phytoplankton in Danish waters.

The focus on macroalgae was the identification of useful indicators. The study was based on hypotheses that cover of the algal community in deeper water increases and composition of the algal community changes towards reduced cover of opportunistic species when water quality improves and that macroalgal cover on unstable substratum is lower than on firm substratum. Algal variables were analysed with reference to substratum and physiochemical variables along with descriptive analyses of algal communities. Coupling algal variables to water quality demonstrated significant relationships between several algal variables and water quality. In conclusion total cover and cumulated cover of coastal macroalgal communities was found to be suitable indicators of water quality if appropriate reference levels for these indicators are defined. Furthermore, there were indications that focus on algae from deeper, light-limited waters renders algal indicators sensitive to changes in water quality.

A somewhat different approach in the study on macrobenthos was to evaluate earlier proposed indices on Danish data along the environmental gradient of oxygen deficiency to arrive at useful indices applicable on Danish conditions. Macrobenthic data from the period 1999-2003 from 11 areas monitored in the Danish National Monitoring and Assessment Programme were analysed. The areas ranged from those hit by severe seasonal hypoxia nearly each year to those who never experience hypoxia. The work was the first test of the AMBI index on Danish monitoring data and both diversity measures and AMBI where significantly correlated with rank numbers based on hypoxic conditions. Overall, the results indicated that both diversity-based indices and the sensitivity based index AMBI can be used to evaluate quality status of benthic communities in Danish waters with respect to bottoms in the salinity regime > 18 psu.

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Resumé

På basis af de biologiske kvalitetselementer phytoplankton, makroalger og makrofauna giver denne rapport et bidrag til udviklingen af værktøjer til at fastsætte Vandrammedirektivets fem miljømålsklasser.

Rapportens første del fokuserer på at fastsætte foreløbige værdier for referencetilstanden for phytoplanktonbiomasser, udtrykt som klorofyl, ved at bruge målinger af Secchi-dybde fra starten af 1900-tallet og Secchi-dybde og klorofyl fra nutidige overvågningsdata. De nutidige data blev brugt til at bestemme relationer mellem Secchi-dybde og klorofyl (90. percentiler af klorofylkoncentrationer). I alle undersøgte områder var der en signifikant sammenhæng mellem Secchi-dybde og klorofylkoncentrationer, og de beregnede 90. percentiler af de historiske data var lavere end nutidige data. Resulaterne af disse analyser bidrager til et første skridt i retning af at etablere referencetilstande for phytoplankton i danske farvande.

Anden del har fokus på makroalgekvalitetselementet og en identifikation af anvendelige indikatorer. Undersøgelsen var baseret på hypoteserne: at makroalgesamfunds dækning på dybere vand stiger, og den relative dækning af opportunistiske arter (eutrofieringsbetingede alger) falder, når vandkvaliteten bliver bedre, og at makroalgedækningen er lavere på ustabilt end på stabilt substrat. Algevariable blev analyseret i forhold til substrat og fysiokemiske variable sammen med deskriptive analyser af makroalgesamfund. I sammenhængen mellem algevariable og vandkvalitet var der signifikante relationer mellem flere algevariable og vandkvalitet. Det blev konkluderet, at total og kummuleret dækning af kystnære makroalgesamfund var bedst egnede indikatorer på vandkvalitet - under forudsætning af, at passende værdier for referencetilstand bliver defineret. Der var endvidere tegn på, at et fokus på makroalger fra dybere mere lysbegrænsede dybder giver indikatorer, der er følsomme over for ændringer i vandkvalitet.

I tredje del blev tidligere foreslåede index for danske makrobenthosdata evalueret langs en iltsvindsgradient med det mål at fremkomme med index, som er anvendelige under danske forhold. Makrobenthosdata fra 1999-2003 fra 11 områder i det danske nationale overvågningsprogram blev analyseret. De 11 områder repræsenterer områder, der rammes af alvorlig iltsvind næsten årligt til områder, der aldrig rammes af iltsvind. Analysen var den første undersøgelse af AMBI-indexet på danske overvågningdata, og både diversitetsmål og AMBI var signifikant korreleret til en rangorden baseret på iltsvindsforhold. De overordnede resultater antyder, at både diversitetsbaserede index og det følsomhedsbaserede AMBI-index kan anvendes til at evaluere kvalitetstilstanden af de bentiske faunasamfund i danske farvande i områder med en salinitet over 18.

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1 Introduction

The Water Framework Directive (WFD) aims to achieve at least a good ecological status in all European rivers, lakes and coastal waters and demands that the ecological status is quantified based primarily on biological indicators, i.e. phytoplankton and benthic flora and fauna. As a consequence the WFD requires reference conditions, response curves and acceptable deviations for these biological quality elements that are part of the classification of ecological status in order to develop classification tools.

Before this can be achieved it is necessary to establish a tool, which, for the single quality elements, describes the correlation between environmental impact/anthropogenic pressures and effect. A first estimate of these tools should be established by the end of 2004. In this way the tools can be applied at the required EU intercalibration in 2005-2006, where Denmark participates in two Geographical Intercalibration Groups (GIG) which cover the North Sea and the Baltic respectively.

A part of the necessary foundation for the basis investigation is an evaluation of which water bodies are being at risk of failing to meet the good ecological status in 2015. Consequently it is necessary to establish type areas and values for the boarder between good and moderate ecological status for the type areas a.o. based on the correlation between environmental impact and effect in the coastal areas.

1.1 The objectives and focus of the project

The aim of this project was to establish a scientific foundation which can contribute to the development of tools that can be applied to assess the five classes of ecological status of the WFD in coastal waters based on the biological quality elements phytoplankton, macroalgae and macrobenthos. Hereby the aim also was to assure a first needed background material regarding coastal waters in relation to the preparation of instructions and manuals on classification of ecological quality as an example needed by the Danish counties/river basin districts. The aim includes an assessment of values for the boundaries between ecological status classes with main emphasis on the boundaries between good and moderate ecological status in relation to the implementation of WFD and the EU intercalibration in 2005-2006 of boundaries between high and good ecological status and good and moderate ecological status.

More specifically the present work on phytoplankton, macroalgae and macrobenthos, respectively, aimed at:

- Establishing preliminary reference conditions for phytoplankton biomass with the use of historical, approximately 100 years old, Secchi depth measurements and relationships between Secchi depth and chlorophyll *a* obtained from recent monitoring data.

- Identifying appropriate macroalgal indicators to be used in the assessment of water quality according to the WFD and more specifically test the following hypotheses: 1) The cover of the algal community in deeper water increases when water quality improves. 2) The composition of the algal community changes towards reduced cover of opportunistic species as water quality improves. 3) Macroalgal cover on unstable substratum is lower than on firm substratum. 4) Diver effects are an important source of variation.

- Testing some quality metrics for macrozoobenthos on Danish conditions. This will be done by evaluating some of the earlier proposed indices on Danish macrobenthos data by examining changes in some of these indices along an environmental gradient of oxygen deficiency.

2 Development of reference conditions for phytoplankton in Danish waters

By Peter Henriksen

2.1 Introduction

Phytoplankton is one of the biological quality elements to be used in the Water Framework Directive. Classification of water quality by means of phytoplankton should be based on phytoplankton biomass, composition and abundance and, in addition, phytoplankton bloom frequency and intensity. For all of these phytoplankton parameters the deviation from reference or undisturbed conditions is a measure of the water quality.

Danish waters have been heavily impacted by human activity for a long time. Thus, it is not possible to obtain reference conditions from data from the present marine monitoring programmes, generally initiated during the 1970s or later. Some early records of phytoplankton from Danish waters are available (eg. Petersen 1903, Ostenfeld 1913). However, the phytoplankton data presented is qualitative, or at best semi-quantitative, and collected using plankton net samples rather than modern techniques precluding a direct comparison with recent data.

In contrast to data on phytoplankton parameters, a number of approximately 100 years old measurements of Secchi depth are available for Danish waters. Secchi depth, or water clarity, is dependent on the abundance or biomass of phytoplankton, commonly expressed by the concentration of chlorophyll *a* (Chl *a*), present in the water. However, there is not a linear relationship between phytoplankton chlorophyll *a* and Secchi depth due to the influence of e.g. suspended particles and dissolved organic matter (DOM) on the water transparency.

The present work aimed at establishing preliminary reference conditions for phytoplankton biomass, expressed as chlorophyll *a*, using historical Secchi depth measurements and relationships between Secchi depth and Chl *a* obtained from recent monitoring data from the geographical areas selected as intercalibration sites for the comming European WFD intercalibration.

2.2 Methods

Data on Secchi depth, Chl *a* concentrations (in 1m depth) and total nitrogen (TN, in 1m depth) collected from the intercalibration sites (Figure 2.1) during the VMP and NOVA monitoring programmes were obtained from the Danish national marine database (MADS).

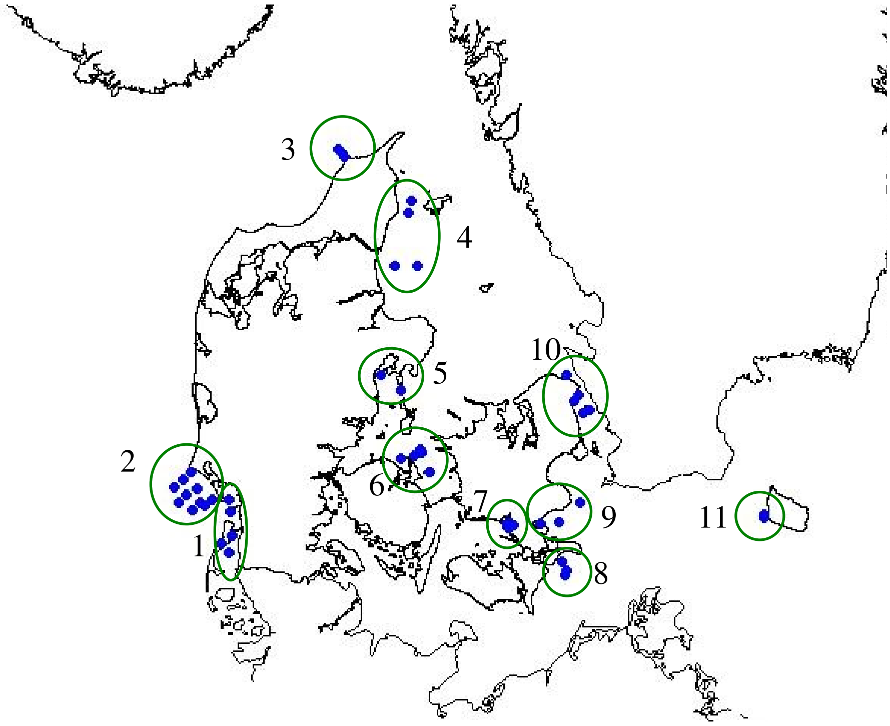

Figure 2.1. WFD intercalibration sites in Danish waters where phytoplankton data is available. 1: Inner Wadden Sea, 2: Outer Wadden Sea, 3: Hirtshals, 4: Northern Kattegat, 5: Århus Bay, 6: North of Funen, 7: Dybsø Fjord, 8: Hjelm Bay, 9: Fakse Bay, 10: Northern and central part of the Sound, 11: West of Bornholm. Each intercalibration site includes several monitoring stations.

For each intercalibration site data from several monitoring stations were included.

Secchi depth measurements taken from Danish waters during the early 1900s (Aarup 2002, data available from the ICES data base) were used for comparison with the relationships found in recent monitoring data. The geographical distribution of these measurements is shown in Figure 2.2. In addition, series of Secchi depth measurements taken from lightships in northern Kattegat and in the Great Belt during the 1960s and 1970s were available for comparison with recent data.

For all intercalibration sites, recent monitoring data were used to establish Secchi depth–Chl *a* and Secchi depth–TN relationships (Figure 2.3). In addition to Chl *a*, suspended particles and DOM will affect Secchi depth. Therefore, relationships between Secchi depth and Chl *a* showed a very large scatter with, in particular at the numerically low Secchi depths, a range of Chl *a* values corresponding to each Secchi depth. To compensate for the lack of complementary data on other factors than Chl *a* influencing Secchi depth, relationships were established using boundary functions describing the upper bounds of the distributions (Blackburn et al. 1992, Krause-Jensen et al. 2000). The rationale behind this approach was that the higher the Chl *a* values at individual Secchi depth, the higher the contribution from Chl *a* to the total light attenuation and thereby influence on Secchi depth. Furthermore, analyses were only performed on summer samples

(May-September) to reduce the likeliness of strong wind events, which potentially lead to heavy resuspension of sediment. During this time period growth of phytoplankton is predicted to be limited by the availability of nutrients rather than light, and Chl *a* is expected to be a major contributor to light attenuation in the water column.

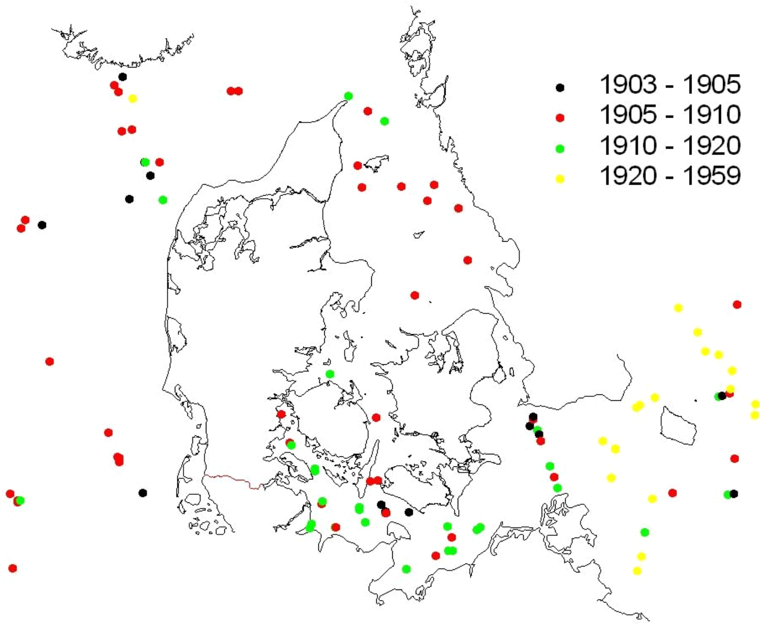

Figure 2.2. Historical Secchi depth measurements from Danish waters (Aarup 2002). Each marker may represent several measurements.

In shallow areas where the cover and depth limit of benthic vegetation has changed from the time of the historical Secchi depth measurements until now, the contribution from resuspended particles to total water column light attenuation may have changed. Such potential changes have not been included in the present work.

Data were grouped into classes representing 1m Secchi depth intervals and for each interval the maximum and 90th percentile of the concurrent Chl *a* measurements were calculated. Subsequently correlations between Secchi depth and Chl *a* were established from regression analyses assuming exponential relationships (Figure 2.3).

2.3 Results

2.3.1 Chl *a*

For all intercalibration sites a significant correlation was found between the Secchi depth and the 90th percentiles of Chl *a* concentrations found within each Secchi depth interval (Figure 2.4). Correlations based on the maximal Chl *a* values were also significant but generally with a lower R^2 than those based on the 90th percentiles (Figure 2.4).

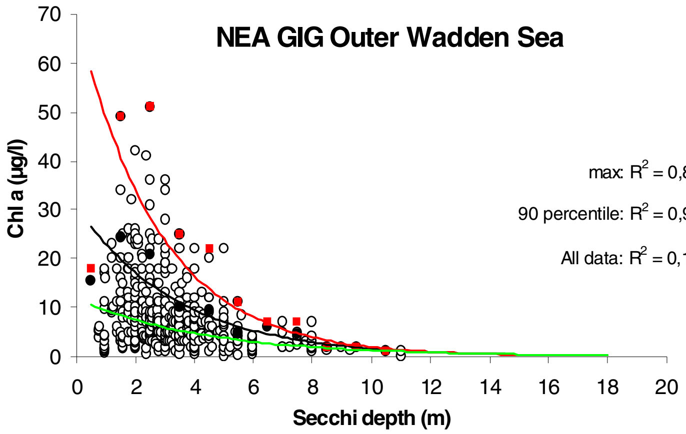

Figure 0.3. Relationship between Secchi depth and Chl *a* for stations included in the Outer Wadden Sea intercalibration site. Open circles represent ungrouped data points while black and red markers represent 90th percentiles and maximum values, respectively, of grouped classes. Green line = all data, black line = 90th percentile and red line = maximum.

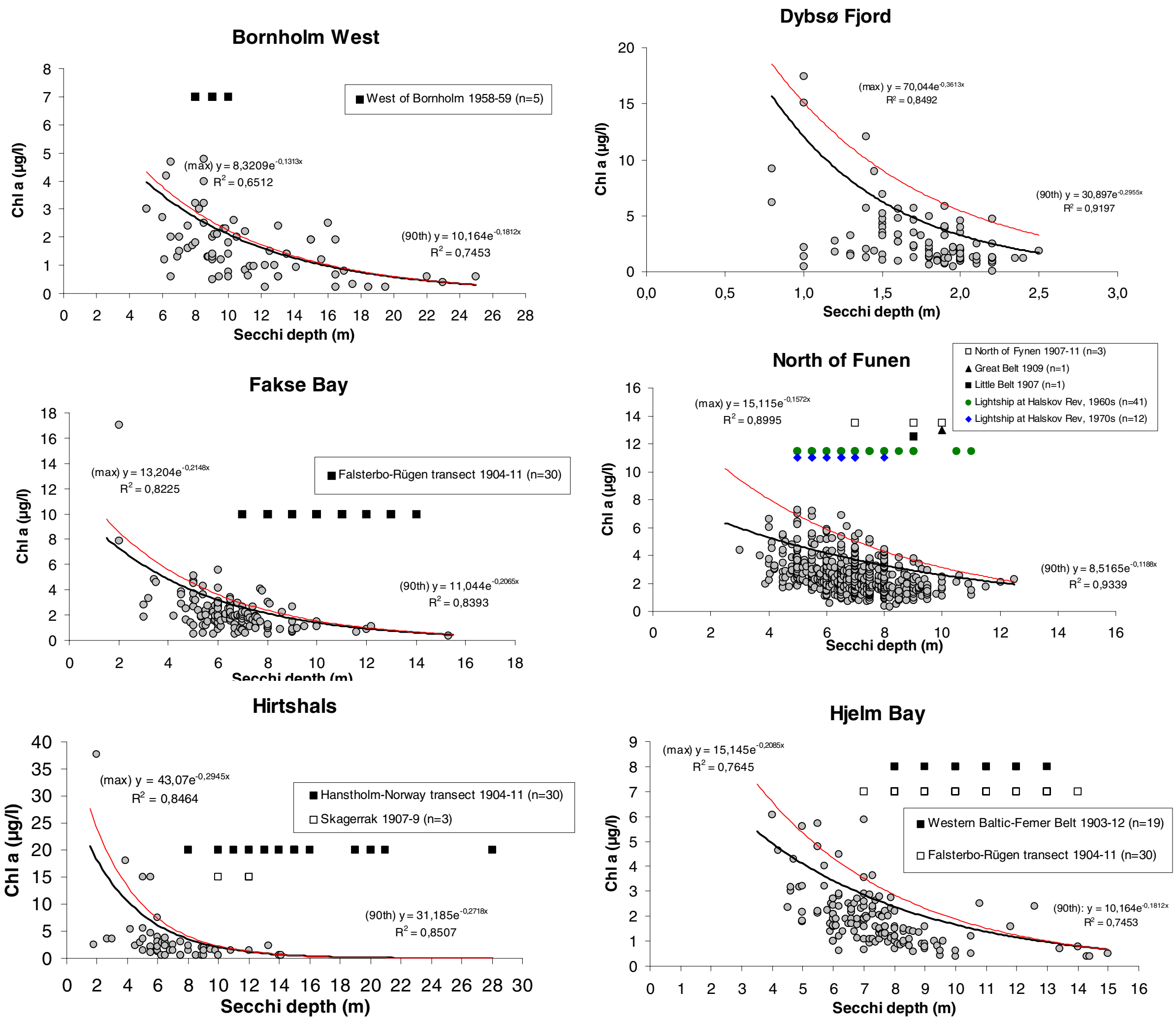

Figure 2.4. Corresponding summer (May-September) values of Secchi depth and Chl *a* concentration in EU WFD intercalibration sites. Additional markers indicate historical Secchi depth measurements available in MADS. Markers of historical Secchi depths are placed arbitrarily along the y-axis (no corresponding Chl *a* values available). Regression models using 90th percentiles or maximum values are shown.

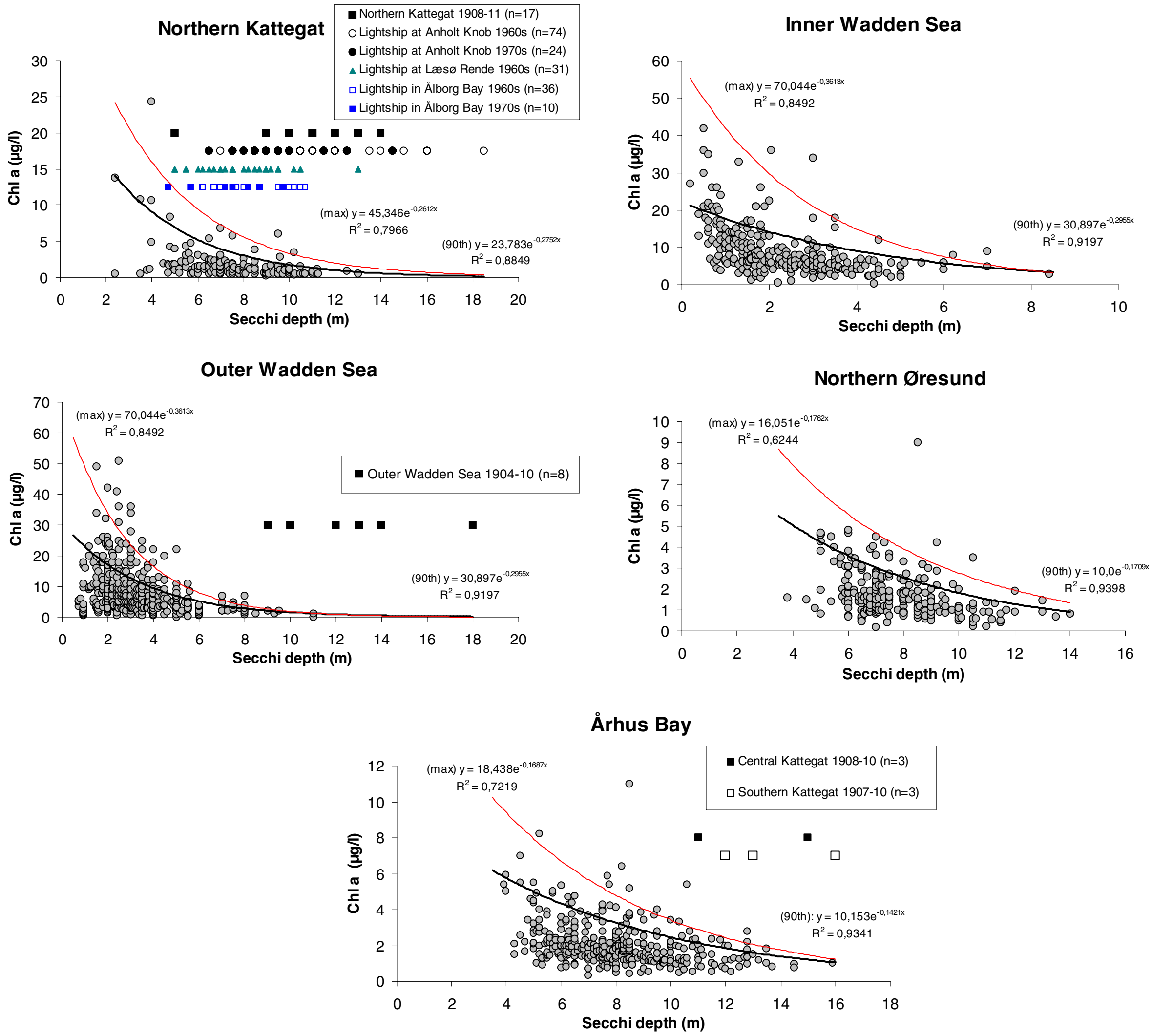

Figure 2.4 (continued). Corresponding summer (May-September) values of Secchi depth and Chl *a* concentration in EU WFD intercalibration sites. Additional markers indicate historical Secchi depth measurements available in MADS. Markers of historical Secchi depths are placed arbitrarily along the y-axis (no corresponding Chl *a* values available). Regression models using 90th percentiles or maximum values are shown.

For areas with historical Secchi depth data "historical Chl *a* values" were calculated using the 90th percentile correlations and averages of the historical Secchi depth measurements (Table 2.1). Historical Secchi depths used for the Hirtshals and outer Wadden Sea intercalibration sites originated further off shore than the intercalibration sites.

Based on Aarup (2002) historical Secchi depths from these two sites were assumed to have been approximately 75 and 65%, respectively, of the off shore values. In Figure 2.6 the reference Chl *a* values, representing the Chl *a* concentration lower than which 90% of the summer Chl *a* concentrations were expected to be approximately 100 years ago, are plotted onto the ranges of summer Chl *a* concentrations found in the recent data.

For all areas the calculated 90th percentiles of historical Chl *a* concentrations were lower than recent ones. Thus the 90th percentiles of recent Chl *a* measurements were from 1.4-fold (North of Funen) to 7-fold (outer Wadden Sea) higher than the calculated historical values.

The historical Secchi depth measurements available for the area north of Fynen ranged from 7 to 10 m. However, data on the depth limit of eelgrass, *Zostera marina*, from the same area and time period showed growth of eelgrass down to approximately 10.4 m depth (Ostenfeld 1908). Thus the few (five) historical Secchi depth measurements available for that area seem to underestimate the Secchi depth resulting in too high a Chl *a* reference condition for that area. This is further supported by the much lower calculated "historical" Chl *a* concentration in Århus Bay located close to the area north of Funen and a generally similarity in the calculated Chl *a* reference conditions for the other intercalibration sites showing similar Secchi depth – Chl *a* relationships (northern Kattegat, northern part of the Sound, Hjelm Bay and Fakse Bay, Figure 2.5 and Table 2.2). Using the relation between depth limit of eelgrass and Secchi depth given in Nielsen et al. (2002) a depth limit of 10.4 m would correspond to a Secchi depth of 12.8 m and subsequently a "historical" Chl *a* concentration (90th percentile) of 1.9 µg l^{-1}. This value is in better agreement with reference values from the other sites in the Kattegat area and is therefore suggested as a better estimate of Chl *a* reference conditions north of Fynen (Table 2.1, 2.2).

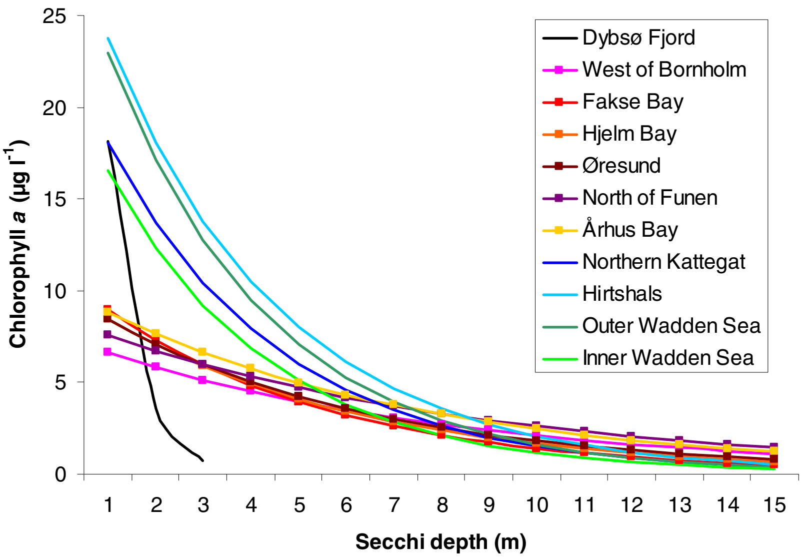

Figure 2.5. Relationship between summer (May-September) values of Secchi depths and Chl *a* concentrations in the Danish EU WFD intercalibration sites. Relationships are based on regression models using 90th percentiles. For some stations the range of Secchi depths plotted exceeds the range found in the monitoring data.

Secchi depths measurements obtained from lightships in the northern Kattegat and the Great Belt during the 1960s and 1970s illustrate a reduction in water clarity from about 1900 until the 1960s and a further reduction from the 1960s till the 1970s (Tables 2.1 and 2.2). Data from the intercalibration sites show that water clarity has improved since the 1980s and present conditions in Ålborg Bay, a major data contribution station from the northern Kattegat intercalibration site, are comparable to those in the 1960s (Figure 2.7, Table 2.2). Eutrophication leading to increased biomass of phytoplankton is expected to be a major cause for the changed Secchi depth regimes shown in Figure 2.7. While Chl *a* measurements are available only from the 1980s and onwards the early measurements of primary production made by Steemann Nielsen in the Kattegat during the 1950s support the hypothesis that the reduced water clarity is coupled to eutrophication. Thus Richardson & Heilmann (1995) calculated a two to three fold increase in primary production in the Kattegat from the 1950s until the period 1984-1993.

Historical Secchi depth measurements were not available for the northern part of the Sound. A modelled "pristine" scenario for the Sound (Øresundsvandsamarbejdet, 2004) suggested that recent summer (June-August) Chl *a* concentrations are approximately 75-90% higher than those 150 years ago. Assuming similar seasonal distributions of Chl *a* during these two time periods and an increase in Chl *a* of 85% from pristine to recent conditions, an estimate of historical Chl *a* for the northern part of the Sound would be that 90% of summer Chl *a* measurements were below 1.7 $\mu g\ l^{-1}$ (Figure 2.6). While not directly comparable these values are in reasonable agreement with a reference Chl *a* concentration (mean of June-August) of 1.4 $\mu g\ l^{-1}$ for the coastal areas of the northern part of the Sound estimated by Samuelsson et al. (2004).

Estimates of historical Chl *a* concentrations are summarised in Table 2.3. In addition to the "historical" 90th percentiles of Chl *a*, estimates of "historical" summer (May-September) average Chl *a* values were calculated (Table 2.4). "Historical" summer averages were calculated from the "historical" 90th percentiles assuming a similar ratio between averages and 90th percentiles in historical and recent (monitoring) data.

When in lack of any historical data it has been suggested that reference conditions, or the boundary between high and good ecological status according to the Water Framework Directive, may be estimated as the 10th or the 20th percentile of recent monitoring data (Andersen et al. 2005). For comparison the 10th and 20th percentiles of the Chl *a* measurements from the monitoring programmes have been included in Table 2.4.

For most of the Danish intercalibration sites the 10th or 20th percentile of monitoring data were in good agreement with estimated reference conditions. At Hirtshals and west of Bornholm the 10th and 20th percentiles were much lower than the estimated reference conditions and in the outer Wadden Sea the 10th and 20th percentiles were approximately 2- and 3-fold higher than the estimated reference Chl *a* concentration (Table 2.4). It should be emphasised that even though the 10th or 20th percentile method showed reasonable agreement with estimated reference Chl *a* concentrations in several of the Danish intercalibration sites this approach should not be taken in more enclosed and heavily eutrofied areas where all recent Chl *a* measurements will be expected to exceed reference conditions.

Table 2.1. Reference summer (May-September) Chl *a* concentrations calculated for the Danish WFD intercalibration sites.

Area	Recent data series [1]	Historical data	Historical Secchi depths					Calculated historical Chl a concentrations			
			n	Min	Max	Avg	Stdev	90th percentile model	Max	Min	Average
West of Bornholm	1988-2003	West of Bornholm (1958-59)	5	8	10	8,8	0,8	Chl a = 7,501e-0,128*Secchi	2,7	2,1	2,4
Fakse Bay	1972-2003	Falsterbo-Rügen transect (1904-11)	30	7	14	10,1	1,8	Chl a = 11,04e-0,207*Secchi	2,6	0,6	1,4
North of Funen	1974-2003	North of Funen (1907-11)	3	7	10	8,7	1,5	Chl a = 8,517e-0,119*Secchi	3,7	2,6	3,0
		North of Funen (Ostenfeld 1908)				12,8		Chl a = 8,517e-0,119*Secchi			1,9
		Great Belt (1909)	1	10	10	10,0		Chl a = 8,517e-0,119*Secchi	2,6	2,6	2,6
		Little Belt (1907)	1	9	9	9,0		Chl a = 8,517e-0,119*Secchi	2,9	2,9	2,9
		I/v Halskov Rev (1960s)	41	5	11	7,1	1,6	Chl a = 8,517e-0,119*Secchi	4,7	2,3	3,7
		I/v Halskov Rev (1970-71)	12	5	8	6,3	0,9	Chl a = 8,517e-0,119*Secchi	4,7	3,3	4,1
Hirtshals	1983-2003	Skagerrak (1907-9)	3	10	12	11,3	1,2	Chl a = 31,19e-0,272*Secchi	2,1	1,2	1,4
		Hanstholm-Norway transect (1904-11)	30	8	28	14,8	4,2	Chl a = 31,19e-0,272*Secchi	3,5	0,0	0,6
		Estimated historical Secchi depth [2]				10,9		Chl a = 31,19e-0,272*Secchi			1,6
Hjelm Bay	1974-2004	Western Baltic - Femern (1903-12)	19	8	13	10,4	1,6	Chl a = 10,16e-0,181*Secchi	2,4	1,0	1,6
		Falsterbo-Rügen transect (1904-11)	30	7	14	10,1	1,8	Chl a = 10,16e-0,181*Secchi	2,9	0,8	1,6
Northern Kattegat	1972-2004	Northern Kattegat (1908-11)	17	5	14	10,5	2,0	Chl a = 23,78e-0,275*Secchi	6,0	0,5	1,3
		I/v Anholt Knob (1960s)	74	6,5	18,5	10,2	2,4	Chl a = 23,78e-0,275*Secchi	4,0	0,1	1,4
		I/v Laesoe Rende (1960s)	31	5	13	8,0	2,1	Chl a = 23,78e-0,275*Secchi	6,0	0,7	2,7
		I/v Aalborg Bay (1960s)	36	4,7	10,7	7,8	1,4	Chl a = 23,78e-0,275*Secchi	6,5	1,3	2,8
		I/v Anholt Knob (1970s)	24	6,5	14,5	9,4	2,0	Chl a = 23,78e-0,275*Secchi	4,0	0,4	1,8
		I/v Aalborg Bay (1970s)	10	4,7	9,7	6,8	1,7	Chl a = 23,78e-0,275*Secchi	6,5	1,6	3,7
Outer Wadden Sea	1982-2003	Outer Wadden Sea (1904-10)	8	9	18	13,0	2,8	Chl a = 30,90e-0,296*Secchi	2,2	0,2	0,7
		Estimated historical Secchi depth [3]				8,5					2,5
Århus Bay	1971-2003	Central Kattegat (1908-10)	3	11	15	12,3	2,3	Chl a = 10,15e-0,142*Secchi	2,1	1,2	1,8
		Southern Kattegat (1907-10)	3	12	16	13,7	2,1	Chl a = 10,15e-0,142*Secchi	1,8	1,0	1,5

1 : Some years missing from the data series

2 : Historical Secchi depths (Figure 2.2) originate further off shore than the Hirtshals intercalibration site. From Aarup (2002) it is estimated that Secchi depths at the Hirtshals site will be approx. 75% of those further off shore.

3 : Historical Secchi depths (Figure 2.2) originate further off shore than the Outer Wadden Sea intercalibration site. From Aarup (2002) it is estimated that Secchi depths at the Outer Wadden Sea site will be approx. 65% of those further off shore.

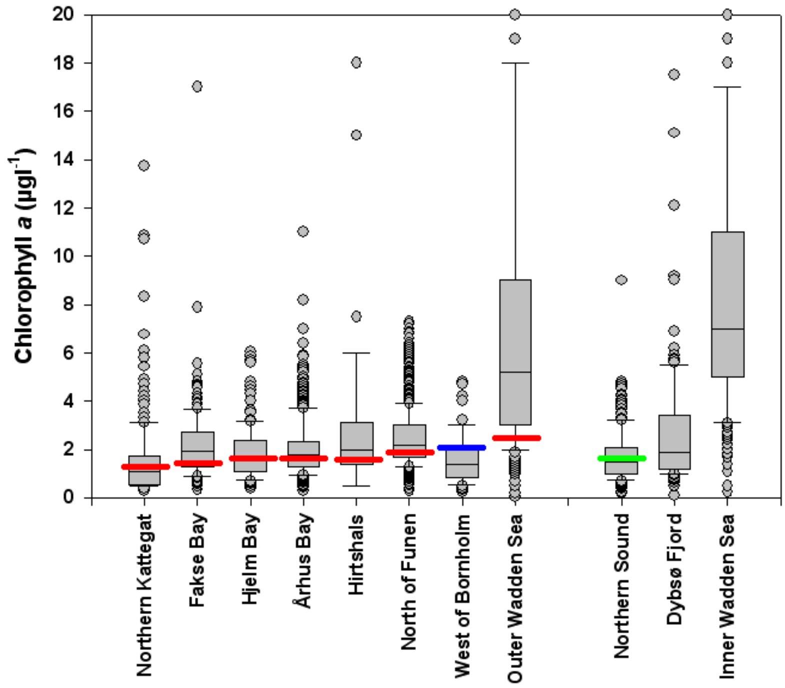

Figure 2.6. Ranges of monitoring data on summer (May-September) concentrations of Chl *a* in the Danish WFD intercalibration sites. Y-axis has been reduced to less that the whole data range for better resolution. Boxes represent 25-75 percentiles and bars 10th and 90th percentiles, respectively. Thin black lines across boxes are median values while thick coloured (red, blue or green) lines represent the calculated 90th percentile of historical Chl *a* concentrations. Red lines are based on historical Secchi depth from about 1900 (see table 2.1) while blue line (west of Bornholm) is based on data from 1958-59. Green line is derived from modelling of pristine conditions in the Sound (see text). Historical Chl *a* concentrations calculated for the area north of Funen are based on observations of depth limit of eelgrass (see text).

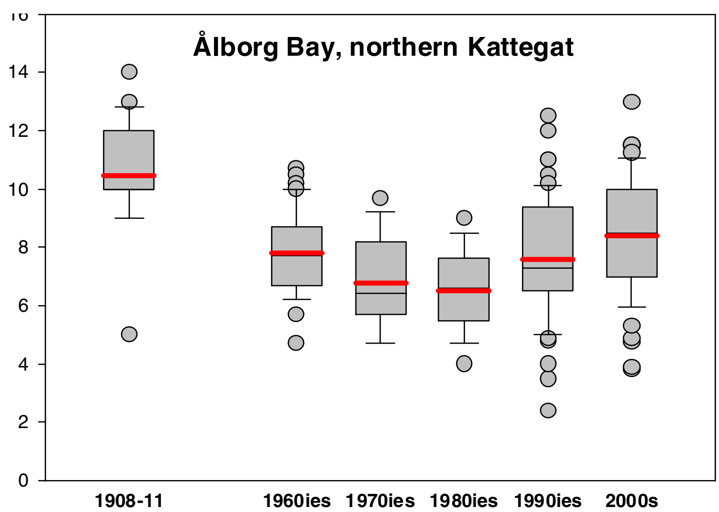

Figure 2.7. Ranges of summer (May-September) Secchi depths at station 409, Ålborg Bay, in the northern Kattegat. Data from 1908-11 are from several positions in the northern Kattegat. Boxes represent 25-75 percentiles and bars 10th and 90th percentiles, respectively. Thin black lines across boxes are median values while thick red lines represent mean values.

Table 2.2. Recent and historical summer (May-September) averages of Secchi depth at the Danish EU WFD intercalibration sites. Historical measurements are taken from Aarup (2002) while recent values originate from the modern monitoring data at the individual site.

	Secchi depth (m)						
	Historical average	Average of 1960s data	Average of 1970s data	Average of 1980s data	Average of 1990s data	Average of 2000s data	2000s data/historical concentrations
Northern Kattegat	10.5	9.1	8.7	6.7	7.6	8.5	0.8
Fakse Bay [a]	10.1			6.7	6.9	6.7	0.7
Hjelm Bay	10.4			5.9	6.8	6.7	0.6
Hirtshals [1 b]	10.9				7.6	7.8	0.7
Århus Bay [c]	13.0		6.7	6.8	8.2	8.9	0.7
West of Bornholm [2]	8.8			7.4	11.4	11.4	1.3
Outer Wadden Sea [3]	8.5			3.0	3.4	3.4	0.4
North of Funen	12.8 [4]		7.0	6.4	6.7	7.7	0.6
Northern part of the Sound				7.2	7.3	8.1	
Dybsø Fjord [d]				1.6	1.8	2.2	
Inner Wadden Sea					1.7	1.8	

1: Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 75% of those off shore.
2: Derived from data collected during 1958-59.
3: Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 65% of those off shore.
4: Based on observations of depth limit of eelgrass (Ostenfeld 1908) converted to Secchi depth according to Nielsen et al. (2002).
a: Station 441 only
b: Station 7715 and 7725
c: Station 170006 only
d: Station 0103011 excluded due to few measurements and only during the 1980s

2.3.2 TN

Relationships between Secchi depths and TN concentrations from the recent monitoring programmes were significant for only four of the WFD intercalibration sites: west of Bornholm, Fakse Bay, Hirtshals and Århus Bay. From relationship between 90th percentiles of TN within 1m Secchi depth intervals and Secchi depth, historical TN concentrations were estimated for these four areas (Table 2.2). These estimates suggest that recent TN concentrations range from approximately similar to those calculated for the late 1950s (west of Bornholm) to 1.3-fold those originating from the early 1900s estimated increases in TN concentrations from "historical" to recent levels were much lower than the increases calculated for Chl *a*.

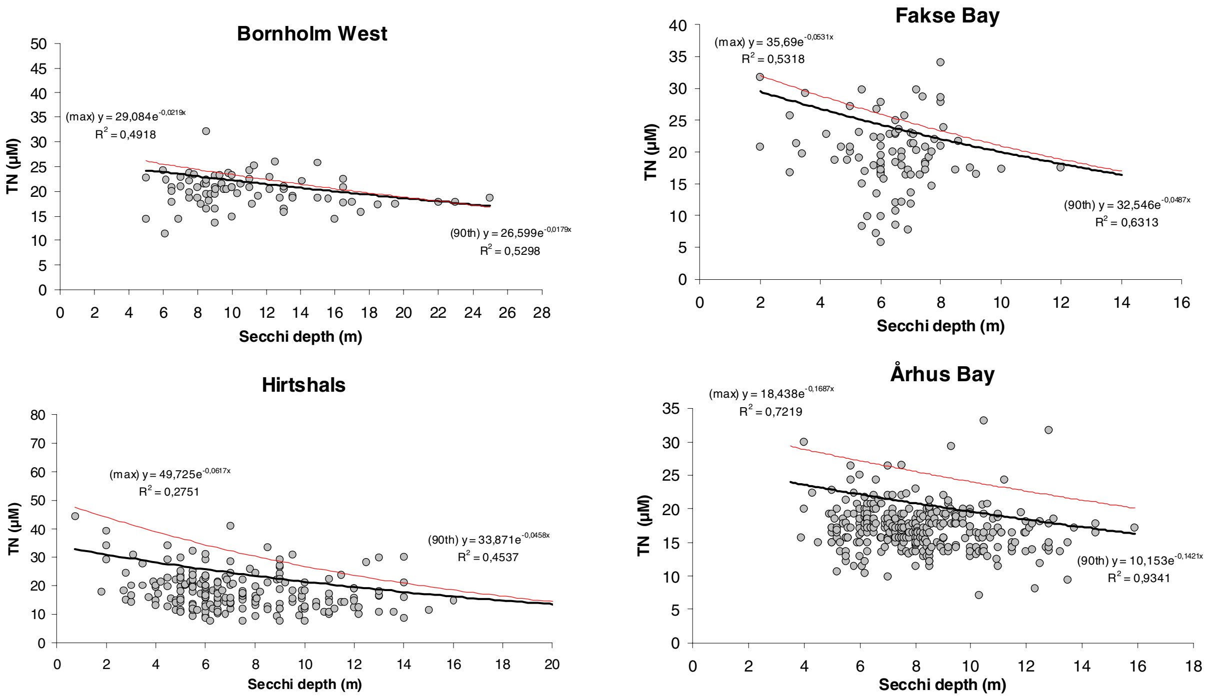

Figure 2.8. Corresponding summer (May-September) values of Secchi depth and TN concentration in WFD intercalibration sites. Only areas with significant correlations are shown. Regression models using 90th percentiles or maximum values are shown.

Table 2.3. Recent and calculated "historical" 90th percentiles of chlorophyll *a* and TN concentrations at the Danish WFD intercalibration sites.

	Chlorophyll *a*			TN		
	Calculated historical 90th percentiles (µg l^{-1})	90th percentiles of recent data (µg l^{-1})	Recent/ historical concentrations	Calculated historical 90th percentiles (µM)	90th percentiles of recent data (µM)	Recent/ historical concentrations
Northern Kattegat	1.3	3.1	2.3		23.7	
Fakse Bay	1.4	3.7	2.7	20.9	27.5	1.3
Hjelm Bay	1.6	3.1	2.0		25.3	
Hirtshals [1]	1.6	5.6	3.5	20.6	27.5	1.3
Århus Bay	1.6	3.1	1.9	17.9	20.7	1.2
West of Bornholm [2]	(2.1)	3.0	(1.5)	(22.7)	23.5	(1.0)
Outer Wadden Sea [3]	2.5	18.0	7.2		50.7	
North of Funen [4]	1.9	3.9	2.1		26.0	
Northern part of the Sound [5]	1.7	3.2	1.9		22.1	
Dybsø Fjord		5.3			55.0	
Inner Wadden Sea		17.0			70.7	

[1]: Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 75% of those off shore.
[2]: Derived from data collected during 1958-59. Not comparable to other "historical" values.
[3]: Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 65% of those off shore.
[4]: Based on observations of depth limit of eelgrass (Ostenfeld 1908) converted to Secchi depth according to Nielsen et al. (2002)
[5]: Based on a modelled "pristine" scenario (Øresundsvandsamarbejdet 2004).

Table 2.4. Recent and calculated "historical" summer (May-September) averages of chlorophyll *a* concentrations and reference conditions for summer Chl *a* derived from the 10th and 20th percentiles of monitoring data at the Danish EU WFD intercalibration sites. "Historical" averages were derived from calculated "historical" 90th percentiles by multiplying the 90th percentiles with the ratio "average" to "90th percentile" of the modern monitoring data at the individual site.

	Chlorophyll *a* ($\mu g\ l^{-1}$)							
	Calculated historical average	Reference conditions based on 10th percentile of monitoring data	Reference conditions based on 20th percentile of monitoring data	Average of 1970s data	Average of 1980s data	Average of 1990s data	Average of 2000s data	2000s data/ historical concentrations
Northern Kattegat	0.7	0.5	0.5	1.2	2.4	2.0	1.1	1.6
Fakse Bay	0.8	0.9	1.1	2.2	1.9	2.4	1.9	2.4
Hjelm Bay	1.0	0.8	1.0			2.2	1.6	1.6
Hirtshals [1]	0.9	0.5	0.5				3.1	3.4
Århus Bay	0.9	0.9	1.2	1.3	2.4	2.2	1.8	2.0
West of Bornholm [2]	(1.3)	0.6	0.6		1.6	1.8	1.4	1.1
Outer Wadden Sea [3]	1.1	2.0	3.0		4.8	8.5	5.9	5.4
North of Funen	1.2 [4]	1.3	1.6	2.0	3.9	2.5	2.2	1.2
Northern part of the Sound [4]	0.9	0.7	0.9	1.8	2.3	2.1	1.5	1.7
Dybsø Fjord		0.6	0.9		2.5	2.3	1.3	
Inner Wadden Sea		3.3	4.5		6.2	9.6	8.4	

[1] : Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 75% of those off shore.
[2] : Derived from data collected during 1958-59. Not comparable to other "historical" values.
[3] : Derived from historical off shore Secchi depths and the assumption that Secchi depths at the intercalibration site are 65% of those off shore.
[4] : Based on observations of depth limit of eelgrass (Ostenfeld 1908) converted to Secchi depth according to Nielsen et al. (2002).
[5]: Based on a modelled "pristine" scenario (Øresundsvandsamarbejdet 2004)

2.4 Conclusion

The present work provides a first step in establishing "reference conditions" for phytoplankton in Danish waters. Chl *a* is the most commonly used proxy for phytoplankton biomass and it has been chosen as the first phytoplankton metric for the WFD intercalibration process. However, in addition to Chl *a*, phytoplankton species composition, abundance and bloom frequency/intensity should be included in the future assessment of water quality. At present, reference conditions are not available for these indicators.

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3 Development of macroalgal indicators of water quality

By Jacob Carstensen, Dorte Krause-Jensen, Karsten Dahl & Anne Lise Middelboe

3.1 Introduction

There is a general need for good biological indicators of marine water quality. Identification of suitable indicators has gained an even higher priority with the adoption of the Water Framework Directive in Europe. According to this directive, the conservation status of marine habitats and the ecological quality of coastal surface waters must be assessed on the basis of quantitative biological indicators. Marine vegetation is generally known to respond to changes in light and nutrient levels and it should therefore be possible to identify macroalgal indicators of water quality.

A major effect of eutrophication on submerged vegetation in coastal waters is shading. Increased nutrient richness stimulates the growth of epiphytic algae (Borum 1985) as well as phytoplankton growth and thereby reduces water clarity (e.g. Nielsen et al. 2002a). Reduced water clarity and increased epiphytic biomass force depth limits of seagrasses and macroalgae towards shallower depths (Duarte 1991, Nielsen et al 2002b) and also reduces the abundance of the vegetation in deeper waters (Dahl et al. 2001, Krause-Jensen et al. 2003). Macroalgal depth limits have some limitations as indicators of water quality, however, because lack of suitable substratum in deep areas may prevent macroalgae from colonising as deep as water clarity allows and in shallow water areas the maximum depth of the area defines colonisation depths. Depth-related cover of macroalgae is therefore likely to be a better indicator of water clarity in Danish waters.

Total cover of erect macroalgae along depth gradients has proven to be a useful indicator of water quality on stone reefs in the Kattegat, Denmark (Dahl et al. 2001). Erect macroalgae generally cover the stone reefs completely down to water depths of 8-10 m and algal cover then declines gradually towards deeper water along with the reduction in light. Algal cover at specific water depths varies among years depending on changes in nutrient level and light climate, the cover being highest in years characterised by low nutrient input and high water transparency. In Danish coastal waters, macroalgal cover along depth gradients has also been shown to reflect differences in water quality between areas (Krause-Jensen et al. 2005). However, coastal algal cover is still not a sensitive indicator as it responds only weakly to changes in water quality. Macroalgal cover thus increased only 4% on average when Secchi depth increased 1 m and did not respond to interannual changes in water quality (Krause-Jensen et al. 2005). Algal cover is therefore not as good an indicator of water quality in Danish coastal areas as it is on stone reefs.

One reason for the limited response of coastal macroalgae to changes in water quality could be that physical exposure plays a major regulating role in these communities. Wind- and wave exposure, desiccation and ice-scour may thus reduce macroalgal cover in shallow coastal waters, while water clarity is likely only to control algal cover at deeper, light-limited water depths. The result of the combined control by physical exposure and water clarity should be a bell-shaped pattern of depth distribution with maximum algal cover at intermediate water depths where physical disturbance is moderate and light levels sufficient. Macroalgal cover is therefore unlikely to respond to changes in light and nutrient levels in shallow water while the algal response in deeper water is likely to be more directly regulated by light and nutrients. As a consequence we expect a higher sensitivity of macroalgae as indicators of eutrophication when focusing exclusively on the depth range from the depth of maximum algal cover to the lower depth limit.

Another reason why coastal macroalgae are less sensitive indicators of water quality than macroalgae on stone reefs could be that diver effects are more prominent in the Danish coastal monitoring programme where many different divers are involved while only few divers are involved in the monitoring of stone reef algae. A third reason could be that the substratum of shallow coastal waters typically is more unstable than the stone reef substratum. Algae growing on small stones or mussel shells are likely to have lower cover due to frequent disturbance of the substratum, and if such data are included in the analysis they are likely to blur relations between algal cover and water quality.

Eutrophication may also cause changes in cover of algal groups having specific demands to nutrient- or light levels. Increased nutrient richness stimulates the group of opportunistic algal species, which have potentially high growth rates (Littler & Littler 1980; Steneck & Dethiers 1994; Duarte 1995; Pedersen 1995). The abundance of opportunistic algae is therefore likely to increase at the expense of perennial algae as a function of increased nutrient input. This pattern of response has been used to develop a macroalgal indicator of water quality in Greek coastal waters (Orfanidis et al. 2001 and 2003). Previous attempts to apply this indicator to Danish coastal waters have failed, however, because the relative abundance of opportunistic algae in a given water body did not relate to water quality but instead responded only to differences in salinity – being highest in the most brackish areas (Krause-Jensen et al. 2005). A recent mesocosm experiment also found no clear response of the entire group of opportunistic algae to increased nutrient level but instead found that green opportunists responded (Karez et al. 2004). Green opportunistic macroalgae are commonly known to respond to nutrient enrichment (Pedersen 1995) and may be more sensitive to changes in water quality than is the entire group of opportunists.

3.2 Aim

This study aims to identify appropriate macroalgal indicators to be used in the assessment of water quality according to the Water Framework Directive. More specifically, we aim to test the following hypotheses: 1) The cover of the algal community in deeper water increases when water quality improves. 2) The composition of the algal community changes towards reduced cover of opportunistic species as water quality improves. 3) Macroalgal cover on unstable substratum is lower than on firm substratum. 4) Diver effects are an important source of variation. We test our hypotheses using a large monitoring data set from Danish coastal waters. We expect that this detailed analysis of regulating factors leads to a refinement of previously developed models (Krause-Jensen et al. 2005).

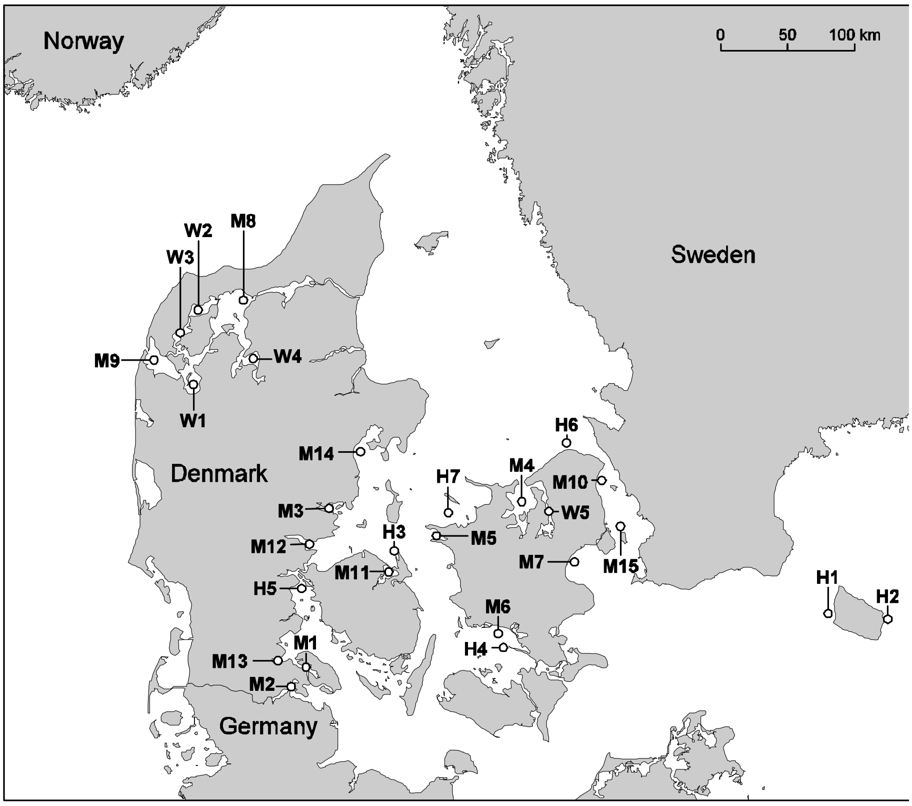

Figure 3.1. Map showing the location of sampling areas. Numbers refer to the areas listed in table 3.1.

3.3 Methods

3.3.1 Algal data

We used data from the Danish National Monitoring and Assessment Programme and regional monitoring activities collected by the Danish counties and stored centrally in the National Environmental Research Institute's (NERI's) database. Data represent a total of 1415-1419 observations, depending on the specific indicator, distributed along 1-11 depth gradients/sites in each of 27 coastal areas (Table 3.1, Figure 3.1). Algal data were collected during summer (May-September) of 2001 and 2003. We chose to use the most recent data set from 2001 and 2003 rather than the entire data set dating back to 1989 because the recent data set is more uniform and better integrated with the pelagic monitoring program. It was collected according to new common guidelines (Krause-Jensen et al. 2001), where divers visually recorded the percent cover of individual erect algal species and of the total erect macroalgal community (excluding the crust-forming algae). Algal cover was estimated in percent of the hard substratum within 3 sub-areas of 25 m^2 in each 2-m depth interval along the depth gradients/sites. Data sets where the summed cover of algal species constituted <80% of the estimated total algal cover were excluded, because we suspected that species registration in these data sets might be incomplete.

Table 3.1. Overview of sampling areas, depth range and number of sites and observations of the macroalgal variables included in the analyses. Sampling years: 2001 and 2003. Area numbers (No.) refer to the numbers in Figure 3.1.

(No.) Area	Depth range (m)	No. of sites (No. of obs.)
Weakly exposed areas		
(W1) Limfjorden, Venø Bay	1-5	2 (31)
(W2) Limfjorden, Mors NW	1-7	3 (52)
(W3) Limfjorden, Mors W	1-5	3 (46)
(W4) Limfjorden, Skive Fjord	1-7	4 (70)
(W5) Roskilde Fjord	1-7	7 (62-63)
Moderately exposed areas		
(M1) Augustenborg Fjord	3-9	6 (32)
(M2) Flensborg Fjord	3-13	10 (78)
(M3) Horsens Fjord	3-7	5 (14)
(M4) Isefjord	3-7	6 (34)
(M5) Kalundborg Fjord	3-11	9 (113)
(M6) Karrebæksminde Bay	3-7	4 (16)
(M7) Køge Bay	3-9	6 (47-51)
(M8) Limfjorden, Løgstør Broad	3-7	4 (47)
(M9) Limfjorden, Nissum Broad	3-7	3 (37)
(M10) Nivå Bay	3-7	2 (15)
(M11) Odense Fjord	3-5	1 (18)
(M12) Vejle Fjord	3-13	5 (35)
(M13) Åbenrå Fjord	3-9	8 (55)
(M14) Århus Bay	3-13	10 (165)
(M15) Øresund	3-11	11 (132)
Highly exposed areas		
(H1) Bornholm W	5-13	3 (70)
(H2) Bornholm E	5-13	3 (56)
(H3) Fynshoved	5-11	4 (51)
(H4) Kirkegrund/ Knudshoved	5-13	5 (70)
(H5) Lillebælt	5-13	6 (126)
(H6) Sealand N	5-13	5 (95)
(H7) Sejerø Bay	5-11	4 (36)
Total		139(1603-1608)

All species were allocated to a functional group, using the system of Steneck & Dethiers (1994, Table 3.2). The functional groups 1-3: microalgae, filamentous algae and single-layered foliose algae are dominated by opportunistic algal species with thin thalli, fast growth rates and ephemeral life forms, while the remaining groups primarily include perennial species with thick, corticated, leathery or calcareous thalli and relatively slow growth rates. In the following we therefore refer to groups 1-3 as 'opportunistic algae' and to groups 4-7 as late-successional species. Microalgae (functional group 1) and crustose algae (functional group 7) were not consistently recorded in the entire dataset and were therefore excluded from analysis.

Table 3.2. Overview of functional groups (Steneck & Dethiers 1994). *Microalgae and crustose algae are not represented in this investigation.

Functional group	Examples of algal genus
1. Microalgae (single cell)*	Cyanobacteria and diatoms
2. Filamentous algae (uniseriate)	Cladophora, Bangia
2.5 Filamentous algae (polysiphonous or thinly corticated)	Polysiphonia, Ceramium, Sphacelaria
3. Foliose algae (single layer)	Monostroma, Ulva, Porphyra
3.5 Foliose algae (corticated)	Dictyota, Padina
4. Corticated macrophytes	Chondrus, Gigartina
5. Leathery macrophytes	Laminaria, Fucus, Halidrys
6. Articulated calcareous algae	Corallina, Halimeda
7. Crustose algae*	Lithothamnion, Peyssonnelia, Ralfsia

We analysed six algal variables: Cumulated cover of erect macroalgae was calculated by summing the cover values of all individual species. Cumulated cover values could surpass 100%, because algae can grow in several layers. Total cover represented the diver estimates of total erect macroalgal cover for each subsample, which represented values in the range 0-100%. The remaining algal variables to be analysed were related to the composition of the macroalgal community. Cumulated cover of opportunistic algae was calculated as the summed cover of all algal species belonging to functional groups 1-3, cumulated cover of opportunistic green algae was calculated as the summed cover of all green algae belonging to functional groups 1-3, and cumulated cover of late-successional algae was calculated as the summed cover of algae belonging to algal groups 4-6. Relative cover of opportunistic algae was finally calculated by dividing the cumulated cover of opportunists by the cumulated cover of all species and therefore provided data in the range 0-100%.

3.3.2 Substratum

Composition of substratum was registered along with the collection of algal data. Divers visually recorded the total cover of suitable hard substratum as well as the cover of various substratum classes: size classes of stones, sand, mud and shells. Data on cover of suitable hard substratum as well as the cover of stones >10 cm were extracted from the database together with each algal dataset.

3.3.3 Physicochemical variables

Spatial variations in algal variables were related to physicochemical variables salinity, nutrient concentrations, chlorophyll concentrations and Secchi depths. These data were sampled at sites situated in the vicinity of vegetation sites. The water chemistry sites were typically located centrally in the investigated coastal areas or subareas, and generally 2 or more algal sites/depth gradients were related to the same water chemistry site.

We assumed that mean values from the various algal sites would represent the algae of a given coastal area and that the centrally located water chemistry site would represent physico-chemistry of the same coastal area in spite of some distance between macroalgal- and water chemistry sites.

A total number of 41 water chemistry sites were used in the analysis. Data were collected by the Danish counties and stored in NERI's database. Sampling and chemical analysis were performed according to common guidelines (Kaas & Markager 1999) and typically represented a sampling frequency between weekly and monthly sampling.

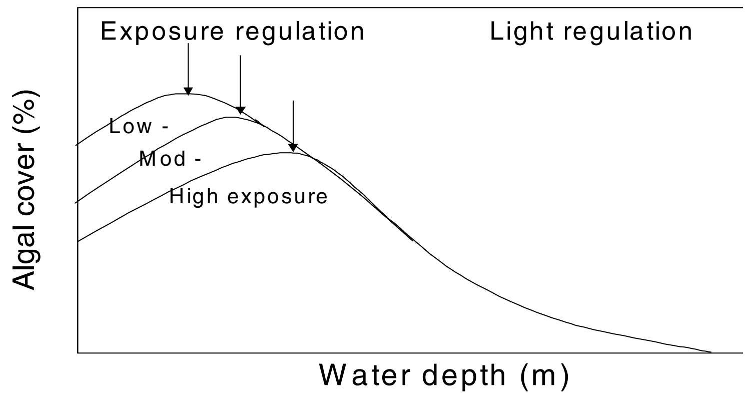

Figure 3.2. Illustration of the hypothesis that algal cover in shallow water is reduced due to physical exposure while from intermediate water depth towards deeper water algal cover is reduced in parallel to reductions in available irradiance. As a consequence maximum algal cover is found at intermediate water depths and we hypothesize that maximum algal cover is located deeper in more exposed areas.

3.3.4 Statistical analyses of algal variables

We wanted to focus the analysis exclusively on algae from the depth range where disturbance was no longer a major controlling factor for cover. We expected that the depth range influenced by physical exposure would increase from weakly towards highly exposed areas (Figure 3.2). The coastward end of this depth range was estimated as the water depth with highest algal cover using non-parametric adjustment (LOESS, Cleveland 1979). This adjustment was made separately for each area and showed that the areas could be categorised in weakly exposed areas where maximum cover was located at water depths of ~1 m, moderately exposed areas with maximum cover at water depths of ~3 m and highly exposed areas with maximum cover at water depths of ~5 m (Figure 3.3). As a consequence we restricted the analysis to water depths >1 m in weakly exposed areas, >3 m in moderately exposed areas and >5 m in highly exposed areas. Only few (79) observations represented water depths >13 m at 4 specific localities (Bornholm West and East, North of Zealand and Little Belt) and we therefore restricted the analysis to water depths <13 m.

Algal cover was estimated as substratum-specific cover, which should imply that cover levels were independent of substratum composition at the sampling sites. A possible dependence on the amount of hard substratum was tested initially using a non-parametric adjustment (LOESS, Cleveland 1979) of each of the potential algal indicators to the amount of hard substratum. This analysis led to the formulation of a model, in which the relation between algal cover and hard substratum differed for levels of hard substratum of below and above 50%.

Algal data representing cumulated cover levels were ln transformed before analysis. By contrast, raw values of the algal variables 'total cover' and 'fraction of opportunists' were in the range 0-100% and greater variation was expected around 50% than at 0% and 100%, so for use in the statistical analyses we employed the following transformation of these data (p, Sokal & Rohlf 1981):

$$x = \arcsin\sqrt{p} \qquad (1)$$

Variations in algal variables (representing either ln transformed or arc sin transformed data, x) were described by the following generic model:

x = area + subarea (area) + site (subarea) + year + month + % hard substratum (0-50%) x depth interval + % hard substratum (50-100%) * depth interval (2

The model is based on the assumption that the observed level of each algal variable depends on coastal area, subarea (inner or outer parts of estuaries), site/depth gradient within the area, water depth, sampling year and month, and substratum composition within depth intervals. The latter is expressed by a linear relation that differs between depth intervals as well as between levels of hard substratum below and above 50%. The model calculates the marginal distributions for the area-specific and depth-specific variations as well as for the year-specific and month-specific variation in algal variables. Marginal distributions describe the variation in a specific factor of the model when variations of all other factors are taken into account. Thus, mean values of each algal variable were calculated for each area, taking into account that monitored depth intervals, substratum composition and sampling year could vary among areas. Thereby, the model provided comparable values of algal variables between areas. These marginal means represented expected values corresponding to a water depth of 7 m (average of the depth range 1-13 m included in analysis), averaged over the two sampling years (2001 and 2003), averaged over the months used in the analysis (May-September), and for a substratum composed of 50% hard bottom. Similarly, the model provided comparable values of algal variables for different water depths and for different sampling years or months. The variation shown by the marginal means should be interpreted as relative variation and not actual levels as some areas, for instance may be shallower than 7 m. However, site-, depth-, time- and substratum-specific values can also be computed by means of the model.

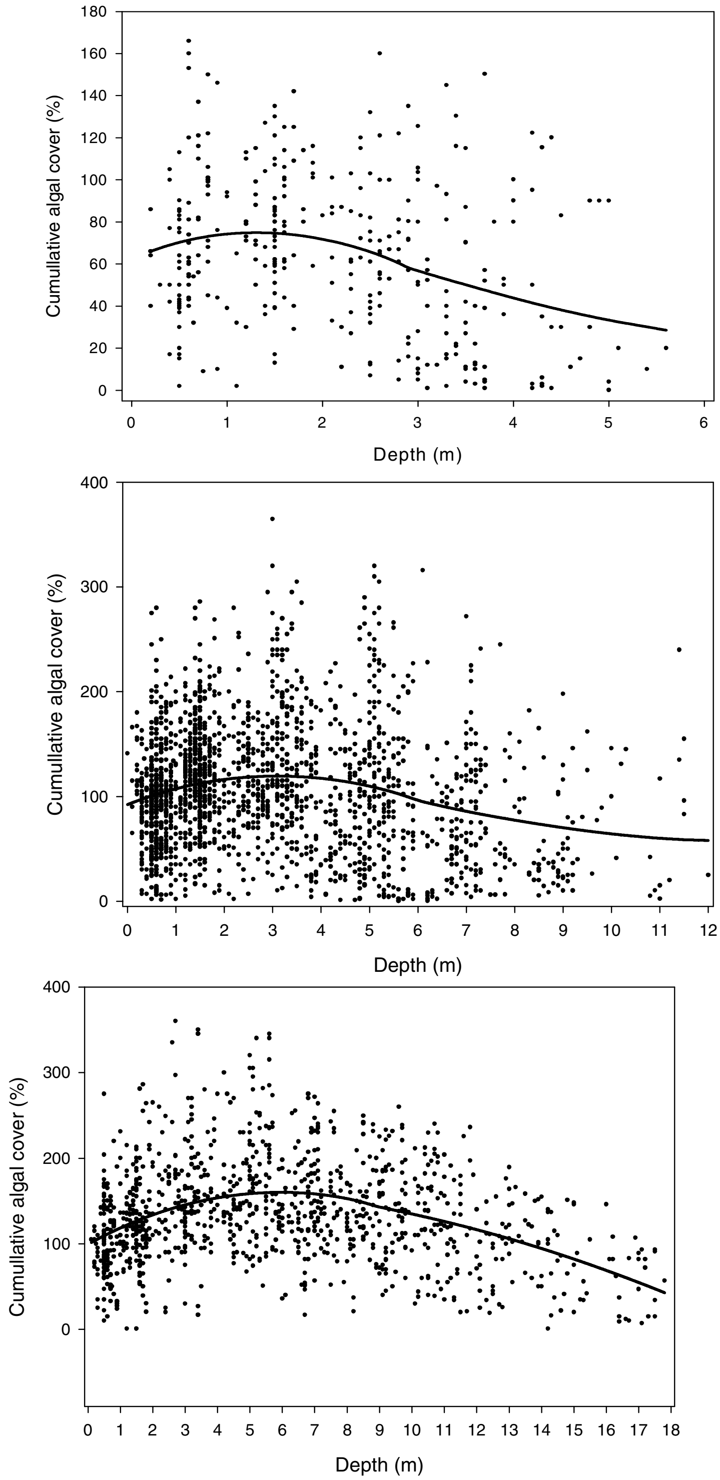

Figure 3.3. Cumulated algal cover as a function of water depth in weakly exposed areas (upper panel), moderately exposed areas (central panel) and highly exposed areas (lower panel).

3.3.5 Testing possible effects of unstable substratum

We tested possible effects of stone size on algal cover through correlation analysis between cumulated algal cover and the fraction of stones >10 cm.

3.3.6 Testing diver effects

The effect of diver, e.g. the possibility that cover estimates differ systematically between divers, was tested using subsets of data. Diver effects can only be exactly compared if different divers make observations at the same site at the same time, excluding spatial and temporal variances. The data did not fulfil this requirement since no site was investigated by more than one diver on each sampling date. However, there was an overlap between divers who had measured cover in the same area(s) either on different sites and/or on different sampling dates/years. It was possible to group such overlapping data sets and compare the average cover levels obtained by different divers. This procedure provided an estimate of the maximum diver effects within each group of data.

3.3.7 Coupling algal variables to water quality

The variation in water quality variables was initially analysed using a model similar to the general model described for algal variables. The model describes water quality variables with respect to area-specific variation, site-specific variation, seasonal variation and year-to-year variation among hydrological years, i.e. July-June. For each water quality variable we calculated area-specific marginal means.

Algal variables were related to physicochemical variables using multiple regression analysis. First we identified the variable with the highest explanatory power and then analysed the residuals of this regression against the remaining water chemistry variables. Supplementary variables were included iteratively as long as the new variables significantly improved the model (forward selection). The analyses were conducted on a spatial basis to explain differences in algal parameters between various coastal areas/subareas.

3.4 Results

3.4.1 Descriptive analyses of the algal community

Data on the various algal variables were modelled based on variation between areas, subareas and sites within each subarea as well as on variation between depth intervals, substrate composition in depth intervals, seasonal variation and year-to-year variation (Table 3.3). The next paragraphs describe the different components of variation for each of the analysed algal variables with a main focus on the variable 'cumulated algal cover'.

Table 3.3. Levels of significance for each model component, coefficients of determination (R^2) and number of observations (n) for the overall model. Models were generated for each of the algal variables: Cumulated algal cover (Cum. cov.), Total cover (Tot. cov.), Cumulated cover of opportunists (Cum. opp. cov.), Cumulated cover of green opportunists (Cum. green cov.), Cumulated cover of late-successional species (Cum. late cov.), and fraction of opportunists (Frac. opp.). P-values for each model component are shown in addition to the coefficient of determination (R^2) and number of obs (n) for the overall model.

Model component	Cum. cov.	Tot. cov.	Cum_late _cov	Cum_opp.	Cum_green _opp	Frac_ opp.
Area	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
- Type (I, O, C)	<0.0001	<0.0001	<0.0001	0.1783	<0.0001	<0.0001
- Site	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Depth interval	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0001
% hard substratum (0-50) x depth	<0.0001	<0.0001	<0.0001	<0.0001	0.0001	0.0296
% hard substratum (50-100) x depth	0.0002	<0.0001	0.0186	0.1275	0.5145	0.0238
Month	0.0022	0.8258	<0.0001	<0.0001	0.0.029	<0.0001
Year	0.0062	0.0082	0.8455	<0.0001	<0.0001	<0.0001
R^2 (%)	65.5	70.0	64.3	54.1	46.6	61.5
n	1419	1419	1419	1419	1419	1415

Variation between areas

Modelled levels of all analysed algal variables differed significantly between areas and except for the cumulated cover of opportunistic algae all variables also differed significantly between subareas (Table 3.3). The modelled levels of mean cumulated algal cover varied markedly between areas, with the lowest levels (down to 9%) in the Limfjord basins and some inner estuaries and the highest levels (up to 347%) along open coasts (Figure 3.4A). Of the areas subdivided into inner and outer areas many showed a tendency towards higher cumulated cover in outer than in inner areas, but this trend was significant only for Flensborg Fjord and Roskilde Fjord (Table 3.4).

Table 3.4. Cumulated cover in inner parts of estuaries (I) relative to outer parts of estuaries (O) or open water coasts outside estuaries (C). Differences in cover between sub-areas are tested for significance using t-test; p-values are given.

Area	Percentages cumulated cover in inner relative to outer estuaries or open coasts (95% C.L.)	P-value
Augustenborg Fjord, I/O	144 (60-339)	0.41
Flensborg Fjord, I/O	13 (7-21)	<0.0001
Horsens Fjord, I/O	55 (22-132)	0.18
Isefjord, I/O	53 (25-113)	0.10
Kalundborg Fjord, I/O	88 (67-115)	0.35
Roskilde Fjord, I/O	17 (10-27)	<0.0001
Skive Fjord, I/O	100 (70-146)	0.97
Vejle Fjord, I/O	91 (34-244)	0.85
Åbenrå Fjord, I/O	127 (60-273)	0.53
Århus Bay, I/O	94 (67-131)	0.72
Århus Bay, O/C	92 (68-123)	0.56
Århus Bay, I/C	86 (66-111)	0.26

Modelled levels of total algal cover showed the same trend as that of cumulated cover with lowest levels (down to 7%) in the basins of Limfjorden and some inner estuaries and highest levels (up to 100%)

along open coasts and outer parts of some estuaries (Figure 3.4B). However, data on total cover differed from those of cumulated cover in that the range of variation was much smaller and the majority of areas had quite similar and high total cover values (>75%). Modelled cumulated cover of late-successional species was also lowest (down to <1%) in the Limfjord basins and some inner fjords and highest along open coasts (up to 489%, Figure 3.4C).

Modelled levels of cumulated cover of opportunistic algae also showed a minimum in the Limfjord basins (down to <1%) while highest values occurred to the south east in e.g. Nivå Bay and along the coasts of Bornholm (up to 90%, Figure 3.4D). Modelled cover of green opportunistic algae showed a very low span, ranging from <0.05% in Venø Bay, Odense Fjord and Roskilde Fjord to a maximum of only 3% in Augustenborg outer fjord (Figure 3.4E). The modelled fraction of opportunists ranged from <1% in Augustenborg inner fjord to almost 100% in Roskilde inner fjord (Figure 3.4F).

Variation along depth gradients

Modelled levels of all tested algal variables differed significantly between water depths (Table 3.3). As data from the most exposed inner depth intervals, having low algal cover were excluded in the data analyses, the modelled levels of all algal variables, except the fraction of opportunists declined exponentially with water depth. On average, total cover and cumulated cover of all algae, late-successionals, opportunists and green opportunists at 7-9 m depth was reduced to only 1-7% of the level at 1-3 m depth (Figure 3.5A-E). Levels of total cover (Figure 3.5B) tended to decline less markedly than cumulated cover levels (Figure 3.5A, 3.5C-E). The fraction of opportunists showed a relative minimum in shallow water and a maximum at 3-7 m depth (Figure 3.5F).

Temporal variation

The average cumulated algal cover was about 11% (95% confidence interval: 3.0%-19.6%) higher in 2001 than in 2003. On average, cumulated algal cover increased from May to July and stayed at the same level from July to September (Figure 3.6A).

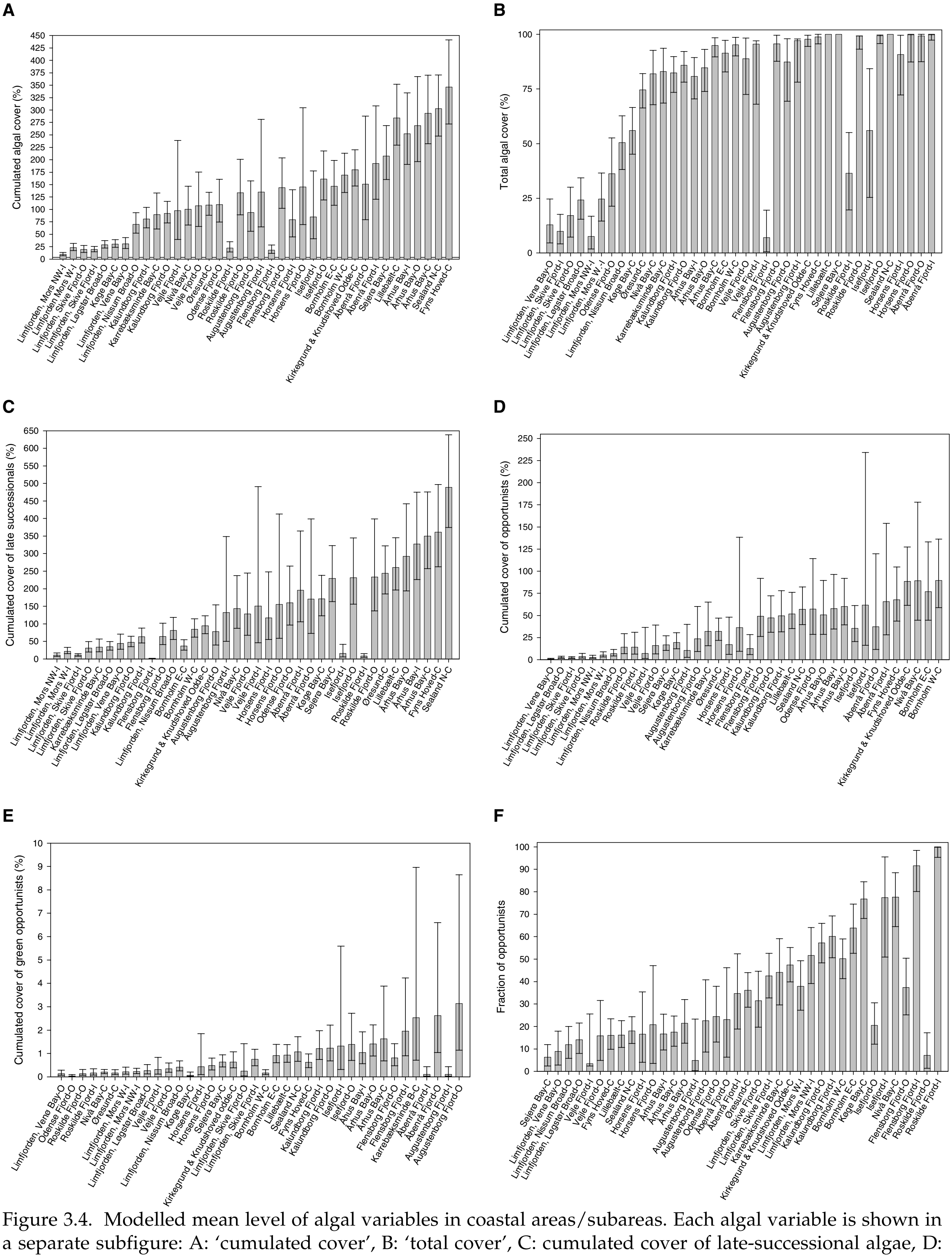

Figure 3.4. Modelled mean level of algal variables in coastal areas/subareas. Each algal variable is shown in a separate subfigure: A: 'cumulated cover', B: 'total cover', C: cumulated cover of late-successional algae, D: cumulated cover of opportunistic algae, E: cumulated cover of green opportunistic algae, F: fraction of opportunists. Areas are ranged according to their mean cover level. In cases where areas are subdivided into inner parts (I), outer parts (O) and coastal areas (C) these are shown side by side and located in the figure according to the average for the area. Data are from 2001 and 2003. Error bars represent confidence intervals.

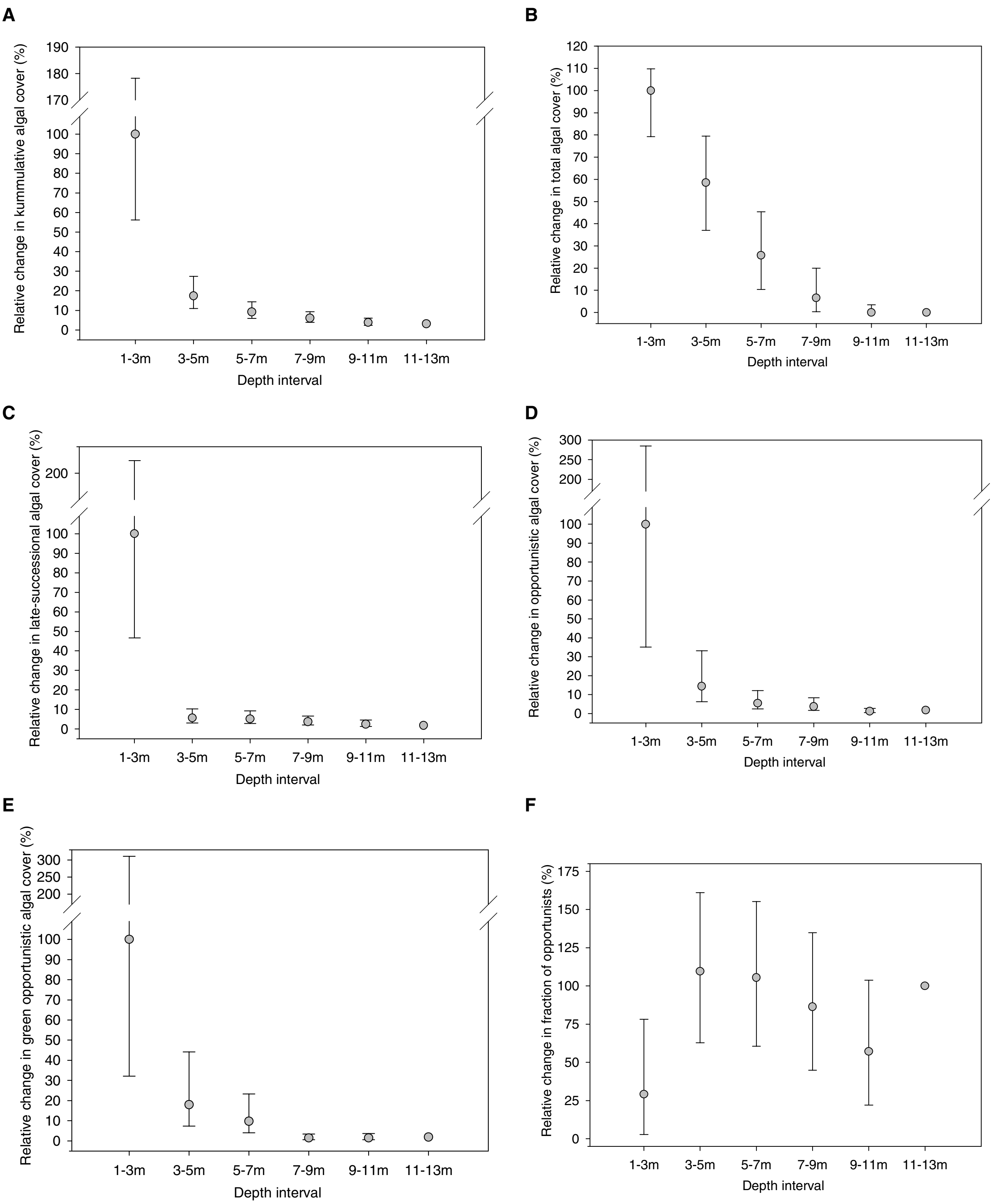

Figure 3.5. Modelled levels of algal variables as a function of water depth relative to the level at 1-3 m depth. Each algal variable is shown in a separate subfigure: A: 'cumulated cover', B: 'total cover', C: cumulated cover of late successional species, D: cumulated cover of opportunistic algae, E: cumulated cover of green opportunistic algae, F: fraction of opportunists. Data are from 2001 and 2003. Error bars represent confidence intervals.

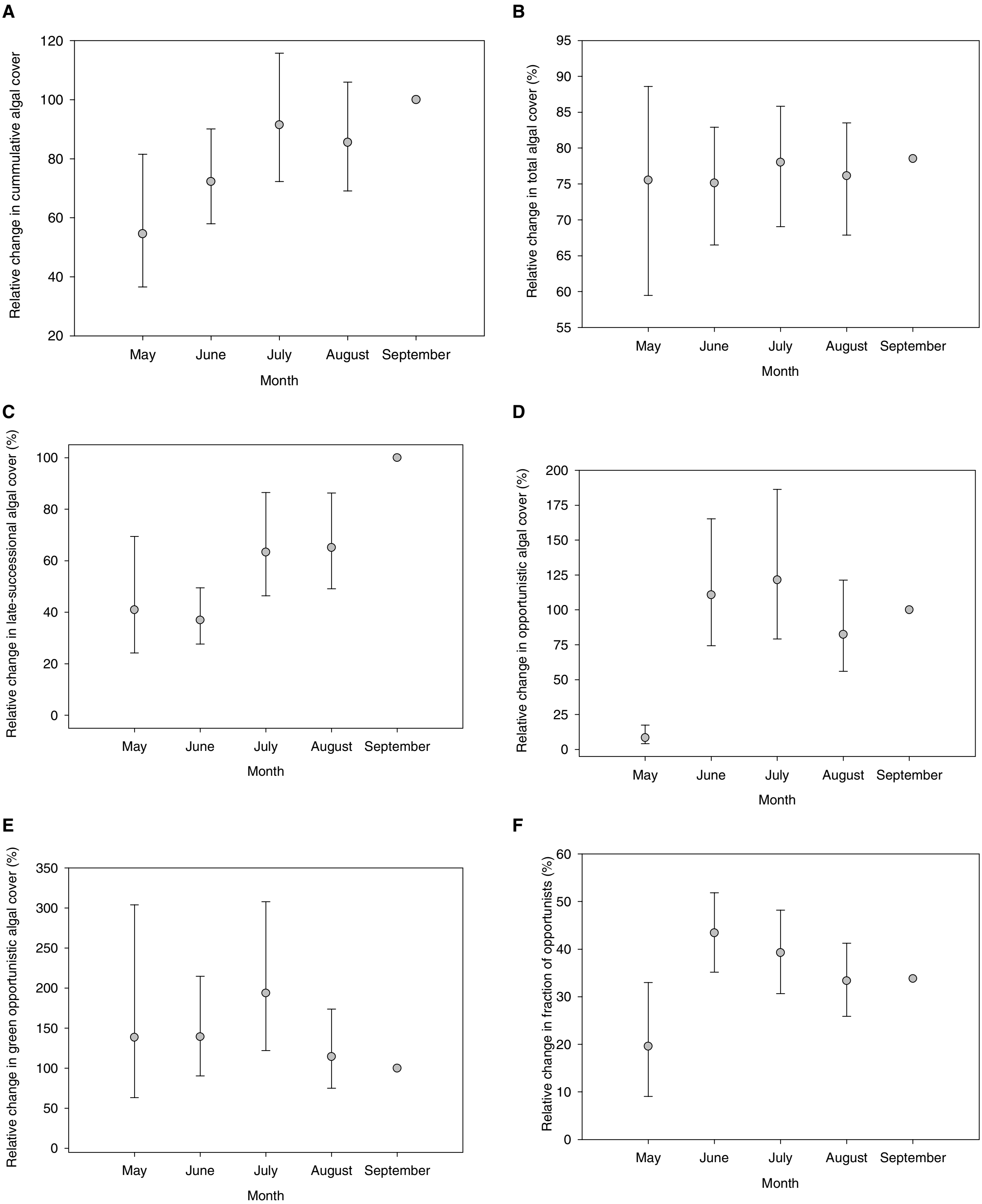

Figure 3.6. Seasonal modelled levels of algal variables relative to the level in September. A: ‘cumulated cover’, B: ‘total cover’, C: cumulated cover of late successional species, D: cumulated cover of opportunistic algae, E: cumulated cover of green opportunistic algae, F: fraction of opportunists. Based on data from 2001 and 2003. Error bars represent confidence intervals.

Differences between years were significant for the other tested algal variables as well, except for the cumulated cover of late successionals. Seasonal variations were also significant for all variables, except for the total cover of the algal community (Table 3.3). Total cover was relatively uniform from May to September while cumulated cover of late-successional algae increased over the entire period. Cumulated cover of opportunists and green opportunists showed a maximum in July while the fraction of opportunists peaked in June (Figures 3.6B-F).

Dependence on substratum composition

Modelling of algal variables was improved by taking into account that algal cover varied with the level of hard substratum (Table 3.3). As an example cumulated algal cover generally increased as a function of increasing levels of hard substratum up to hard substratum levels of 50%; but the increase was significant only in the depth interval 5-13 m (t-test, $p<0.05$). Further increases in the level of hard substratum had no significant effect on algal cover except in the depth intervals 3-5 m and 7-9 m (t-test, $p<0.05$; Figure 3.7).

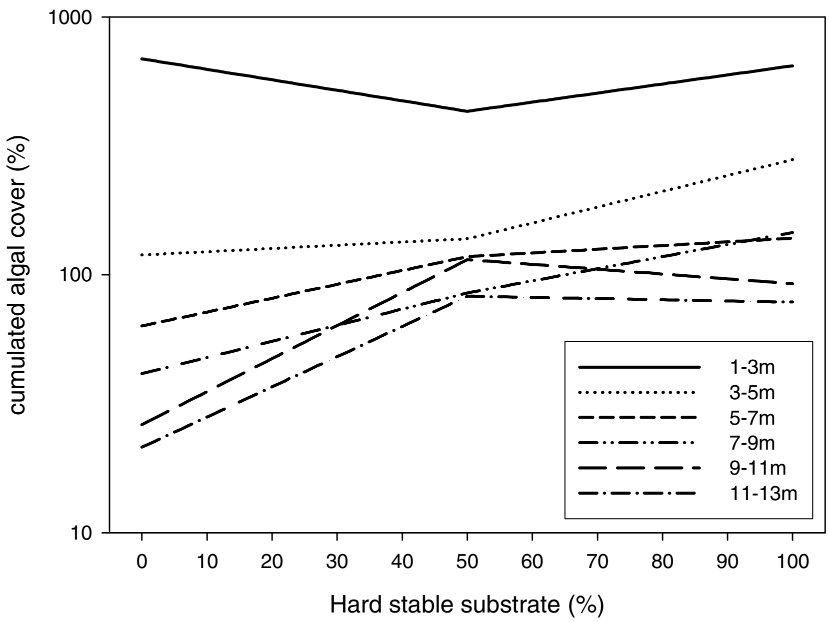

Figure 3.7. Non-parametric curve-fitting (LOESS) of the modelled level of cumulated algal cover as a function of the fraction of hard substratum in different depth intervals.

Possible effects of unstable substratum

Possible effects of unstable substratum were tested only for the variable 'cumulated algal cover'. Unstable substratum had a small but significantly negative effect on cumulated algal cover (t-test, $p<0.05$, Figure 3.8). In order to investigate this relation closer, we focused on the sites having large fractions of unstable substratum/small stones. The majority of these sites were found in Limfjorden. However, a closer look on the relation between algal cover and the fraction of unstable substratum/small stones at these sites revealed that the lowest cover levels occurred at intermediate levels of unstable substratum rather than at the most unstable substratum. These cases of low algal cover were especially prominent in deeper waters and occurred in specific areas and years so that a given site could experience a markedly reduced algal cover in deep water in 2003 relative to 2001 while sites located in other areas in Limfjorden could show the opposite trend. Due to these difficulties in interpreting the effects of unstable substratum, this variable was not included in the general model. We suspect that the sudden decrease in algal cover from one year to another may be due to the exploitation of mussels in the Limfjord.

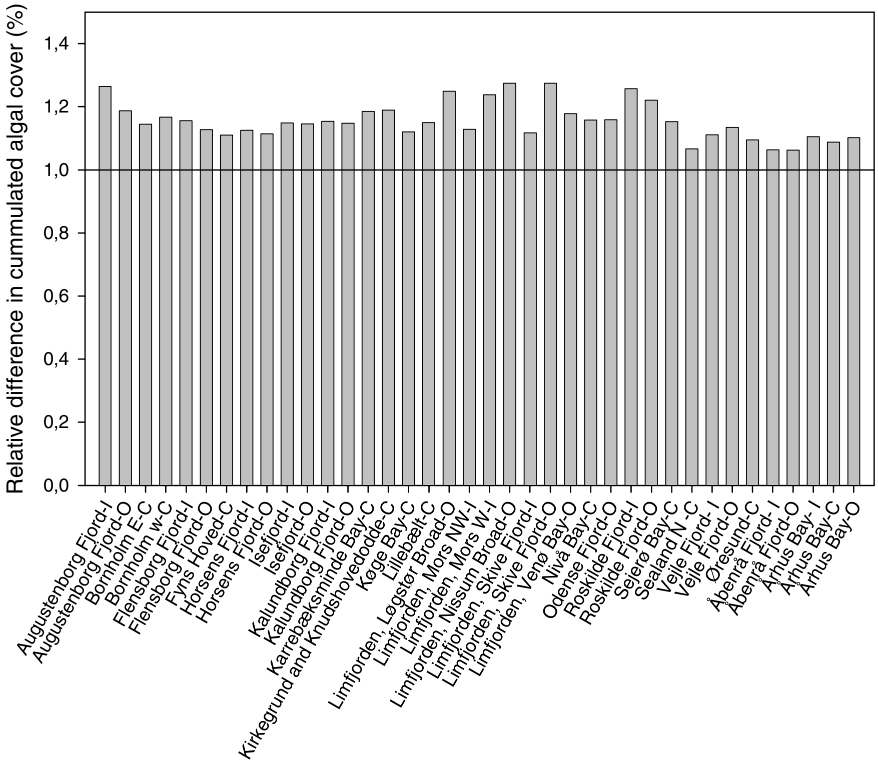

Figure 3.8. Effects of unstable substratum illustrated as the ratio between levels of cumulated cover adjusted and non-adjusted for effects of unstable substratum. Substratum stability is measured as the cover of stones>10cm divided by the total cover of hard substratum.

Diver effects

Possible effects of divers were tested only for the variable 'cumulated algal cover'. Among the fourteen divers involved in macroalgal investigations in 2001 and 2003 we identified four groups of divers who had made observations in the same areas (though on different sites and/or different sampling dates/years). Each of two groups including eight and four divers, respectively, was tested for systematic differences between the divers' cover estimates. The remaining two groups included only one diver each and could consequently not be tested for diver effects.

Cover estimates differed significantly between divers in both of the tested groups (F-test, $p<0.0001$ and $p=0.0160$ for the two groups). When taking our estimates of diver effects into account, cover levels often changed markedly (Figure 3.9). However, as different divers never investigated the same sites at the same time, the differences between the divers' cover estimates cannot be purely attributed to diver effects but are also due to real differences in levels of algal cover between the sites, water depths and sampling dates/years investigated by the various divers. When our test identifies that a diver provides relatively high cover levels, this can either be due to a tendency of that diver to produce high cover estimates relative to the other divers, or it can be because the sites or sampling dates/years investigated by that diver generally had high cover levels. Differences between divers' estimates of cumulated cover can be due to tendencies to under- or overestimate cover levels but can also be influenced by different taxonomic skills of the divers since longer species lists may tend to create higher levels of cumulated cover.

It was not possible to take diver effects into account in the general model, because we could not quantify systematic differences between all divers involved in the investigations. However, the analyses suggest that diver effects can be considerable. If we consider the diver effect not to be systematic, i.e. divers are not expected to give persistent systematically different cover estimates, but to be random we can estimate that the random diver variation has the same magnitude as the residual variation.

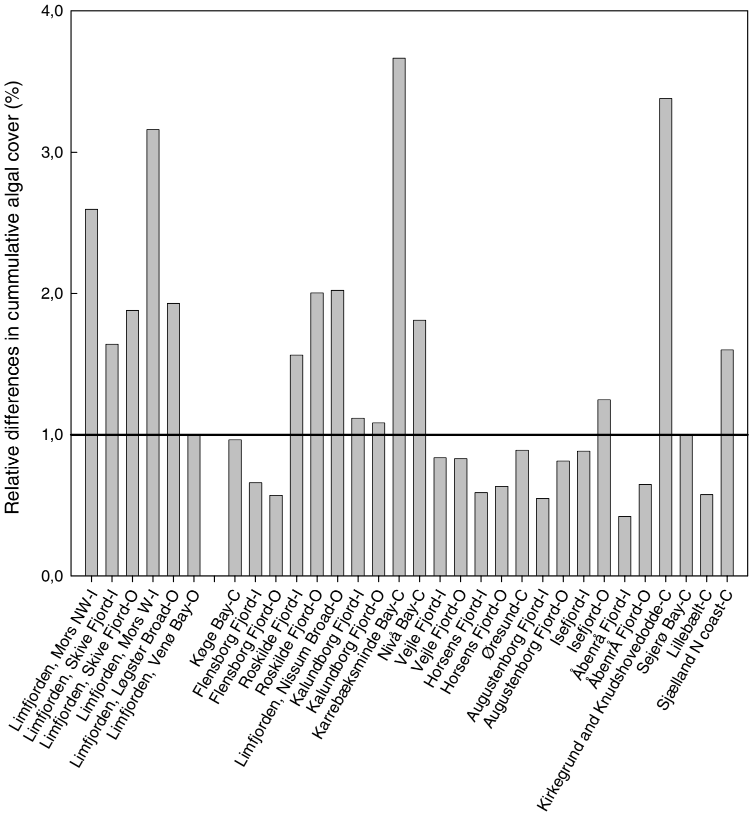

Figure 3.9. Potential diver effects shown for the separate two groups of data where diver effects could be tested. The bars represent the ratio between levels of cumulated cover adjusted and non-adjusted for potential diver effects.

3.4.2 Physicochemical variables

Physico-chemical variables were also successfully modelled using the same model as for algal variables. Annual mean levels of physico-chemical variables varied markedly between areas (Figure 3.10). Nutrient concentrations were generally highest in inner estuaries and

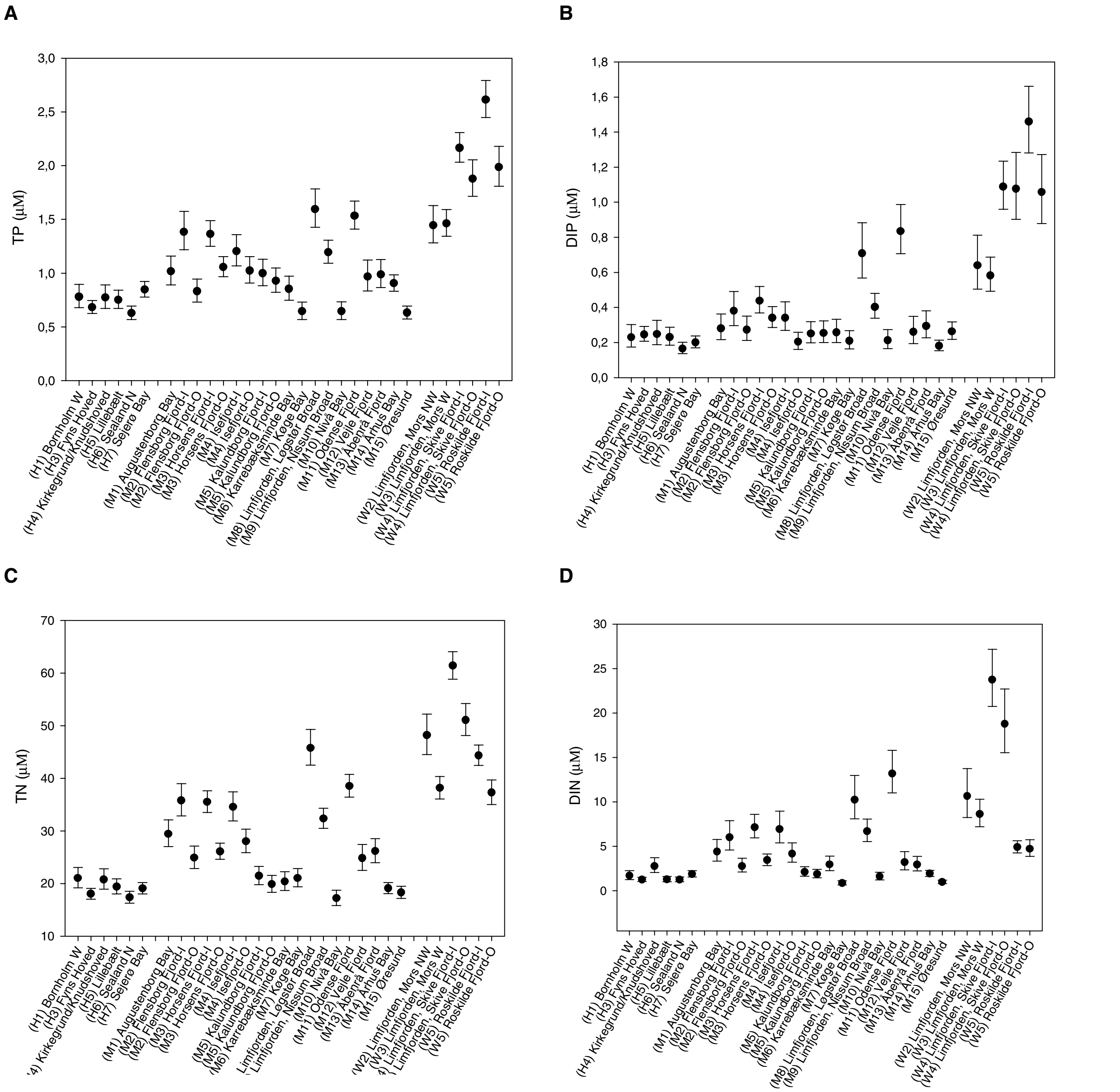

Figure 3.10. Mean modelled levels of physico-chemical variables in the various coastal waters included in the analysis: A: conc. of of TP, B: conc. of DIP, C: conc. of TN, D: conc. of DIN (figure continued on next page).

lowest along open coasts. Modelled mean concentrations of total phosphorus ranged from 0.63 µM TP in Øresund to 2.61 µM TP in inner parts of Roskilde Fjord, while concentrations of inorganic phosphorus ranged from 0.17 µM DIP north of Sealand to 1.46 µM DIP in inner parts of Roskilde Fjord (Figure 3.10A). Modelled mean concentrations of total nitrogen ranged from 17.23 µM TN in Nivå Bay to 61.42 µM TN in inner parts of Skive Fjord, while concentrations of inorganic nitrogen ranged from 0.87 µM DIN in Køge Bay to 23.73 µM DIN in inner parts of Skive Fjord (Figure 3.10B).

Inner estuaries also generally had the most turbid waters with low Secchi depths and high concentrations of chlorophyll while open coastal waters had high water clarity and low concentrations of chlorophyll. Area-specific mean secchi depths ranged from an average of 3.2 m in Horsens Inner Fjord to 14.1 m around Bornholm while area-specific mean chlorophyll concentrations ranged from 1.3 µg l^{-1} in Køge Bay to 7.2 µg l^{-1} in Skive Fjord (Figure 3.10C).

Area-specific mean salinities declined markedly from water bodies in the north-west towards those in the south-east, reflecting the mixing between North Sea water of high salinity and Baltic Sea water of low salinity. Salinity means ranged from an average of 30.7 in Nissum Broad to an average of 7.5 around Bornholm (Figure 3.10D).

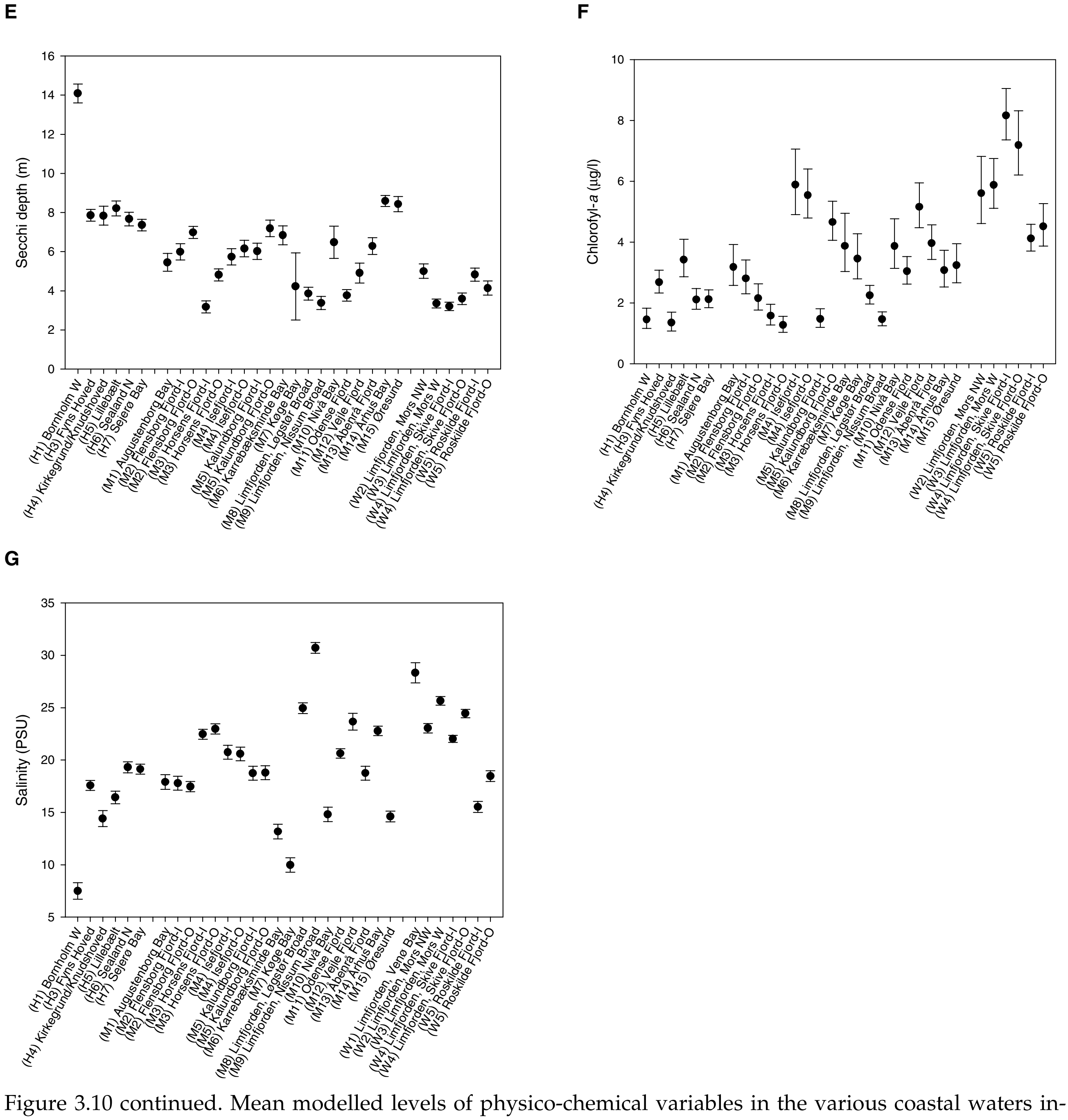

Figure 3.10 continued. Mean modelled levels of physico-chemical variables in the various coastal waters included in the analysis: E: Secchi depth, F: conc. of chl. , and G: salinity. The areas are organised alphabetically within groups of exposure levels (H: high, M: mean, W: weak).

3.4.3 Algal variables in relation to physicochemical variables

The area-specific modelled marginal means of cumulated algal cover were related to the area-specific modelled means of the physico-chemical variables through multiple regression analysis. In this analyses mean cover levels for each coastal area/subarea represented an average water depth of 7 m, a substratum composed of 50% hard bottom and July as the sampling month. The results are presented below for each of the tested algal variables.

Cumulated cover of the algal community

The cumulated cover of the algal community was positively related to salinity and Secchi depth and negatively related to TN (Multiple linear regression, R^2= 0.73, Figure 3.11, Table 3.5). Algal cover generally increased 6.0% per increase in salinity units, -4.3% per 1 µM increase in TN and 30% per 1 m increase in Secchi depth. TN and Secchi depth were, however, correlated, so in order to estimate the direct effect of TN concentration, we fitted a non-linear model for the relationship between Secchi depth and TN (Figure 3.12).

Table 3.5. Significant parameter estimates, intercepts, coefficients of determination (R^2) and levels of significance for relationships between algal variables and physicochemical factors modelled by linear regression analysis. The following algal variables were analysed: Cumulated algal cover (Cum. cov.), Total cover (Tot. cov.), Cumulated cover of opportunists (Cum. opp. cov.), Cumulated cover of green opportunists (Cum. green cov.), Cumulated cover of late-successional species (Cum. late cov.), and fraction of opportunists (Frac. opp.). The table shows significant parameter estimates, intercept, coefficients of determination (R^2) and p-values.

Variable	TN	DIP	Secchi	Salinity	intercept	R^2	p
Cum. cov. (log)	-0.0432		2.264	0.060	2.7633	0.73	<0.0001
Tot. cov.	-0.0226				1.3472	0.73	<0.0001
Cum. late cov. (log)	-0.0732				6.2236	0.53	<0.0001
Cum. opp. cov. (log)	-0.0624		0.1591		3.2620	0.53	<0.0001
Cum. green cov. (log)		-1.4667			-0.8970	0.25	0.0045
Frac. opp.				-0.0354	1.0713	0.56	<0.0001

The explanatory variables included in the model all represented annual mean values. Mean summer values of physicochemical variables did not provide models with higher explanatory power. Data from Bornholm deviated from the overall relationships found for the other coastal areas and were therefore not included in the general model (Figure 3.11).

Though the developed linear model was highly significant, the response of macroalgal cover to increasing concentrations of TN seemed to describe a marked change (threshold) at TN values of 35-40 µM (illustrated by grey marking on Figure 3.11) which would be better described by a non-linear model (see later).

Total cover of the algal community

Total algal cover showed a strong negative relation to TN (R^2=0.73, Figure 3.13, Table 3.5). It generally decreased 2.2% per 1 µM increase in TN. Inclusion of salinity as an explanatory variable in addition to TN did not improve the model.

Total algal cover also tended to describe a threshold at TN values in the range 35-40 µM (shown by grey marking on Figure 3.13) which would be better described by a non-linear model (see later).

Cumulated cover of late-successional algae

The cumulated cover of late successional species was negatively related to concentrations of total nitrogen (R^2=0.53, Figure 3.14, Table 3.5). It generally decreased 7.3% per 1 µM increase in TN. It also tended to increase with increasing salinity, but this tendency was non-significant.

Cumulated cover of opportunistic algae

The cumulated cover of opportunistic algae was positively related to Secchi depths and negatively related to concentrations of total-nitrogen (R^2=0.53, Figure 15, Table 2.5). The cumulated cover of opportunists thereby showed the same relations to water quality as did the cumulated cover and the total cover of the entire algal community. The cumulated cover of opportunists also tended to decline as salinity increased, but this tendency was non-significant.

Cumulated cover of green opportunistic algae

The model that best explained variations in cumulated cover of green opportunistic algae included the concentration of inorganic phosphorus as the only independent variable. Cover of the green opportunists declined as concentrations of inorganic phosphorus increased (R^2=0.25, Figure 3.16, Table 3.5)

Fraction of opportunistic algae

The fraction of opportunistic algae was negatively related to salinity (R^2=0.56, Figure 3.17, Table 3.5) and generally declined by 4% per 1 psu increase in salinity. However, the fraction of opportunists showed no relation to water quality as expressed by nutrient concentration or water clarity.

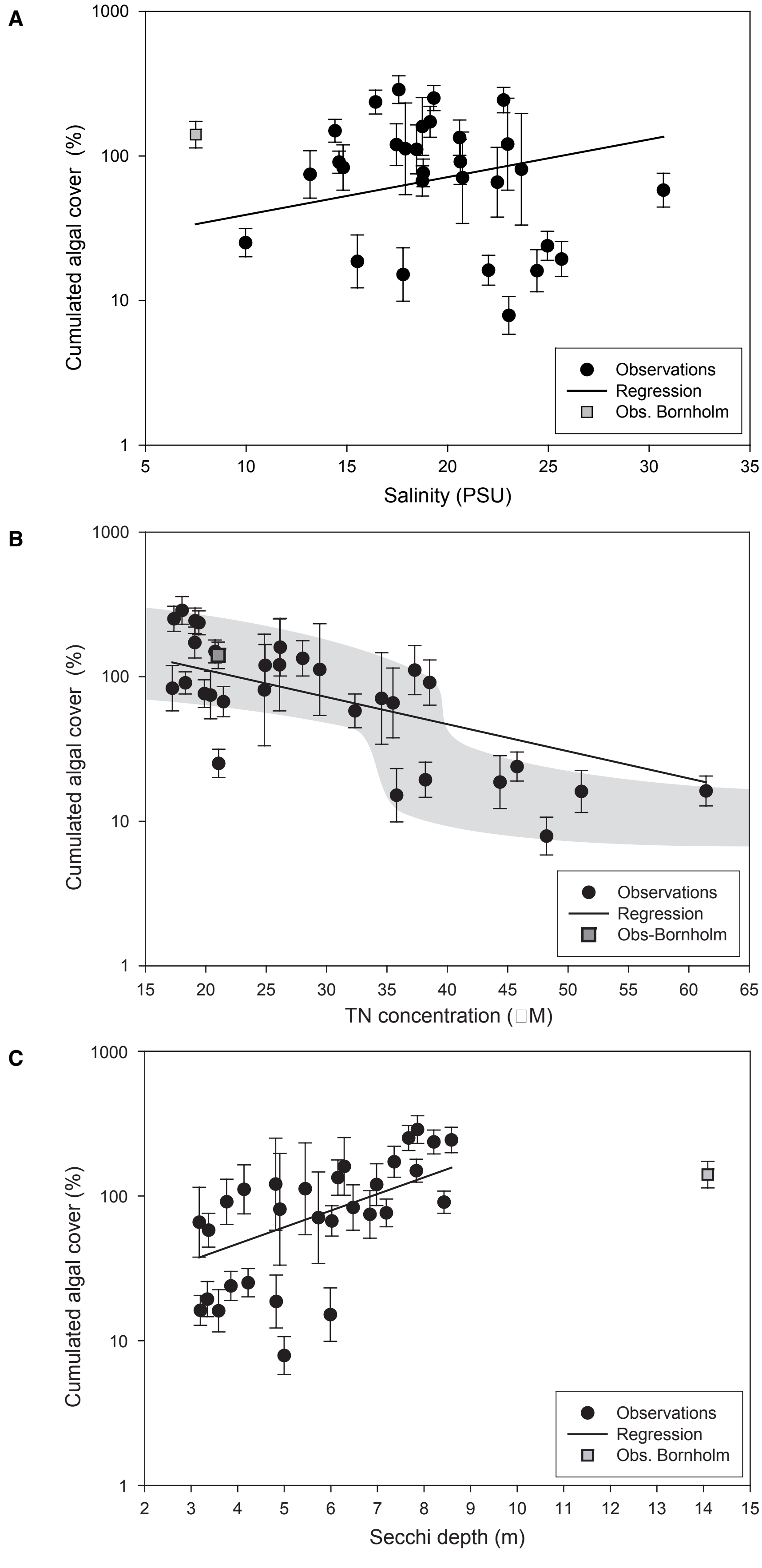

Figure 3.11. Cumulated algal cover in relation to physicochemical variables. A: Algal cover versus salinity. B. Algal cover versus TN. A grey marking is added to illustrate the abrupt change/threshold at TN concentrations of 35-40 µM. C. Algal cover versus Secchi depth. Data in annex 1.

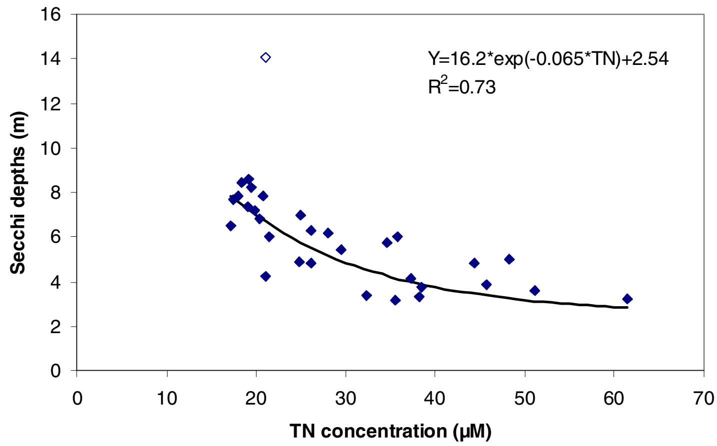

Figure 3.12. Relationship between Secchi depth and TN fitted by a non-linear model.

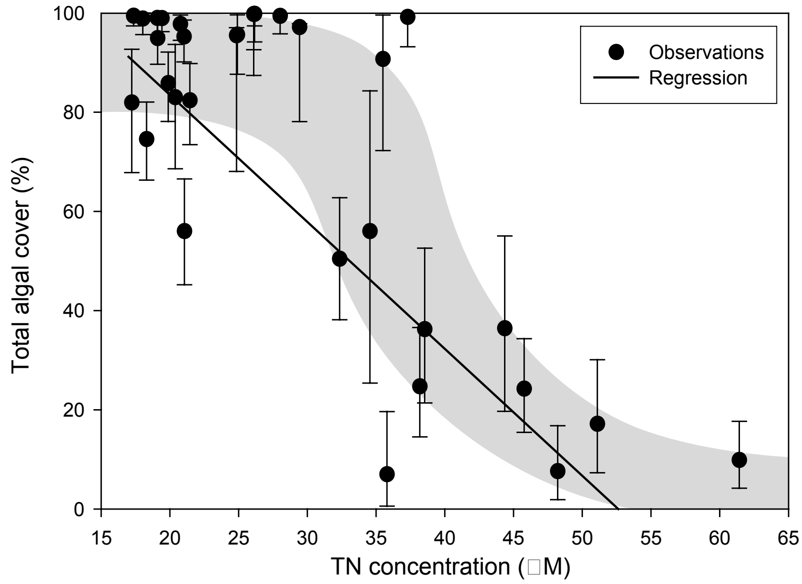

Figure 3.13. Total algal cover in relation to concentrations of total nitrogen (TN). A grey marking is added to illustrate the abrupt change/threshold at TN concentrations of 35-40 µM. Data in annex 1.

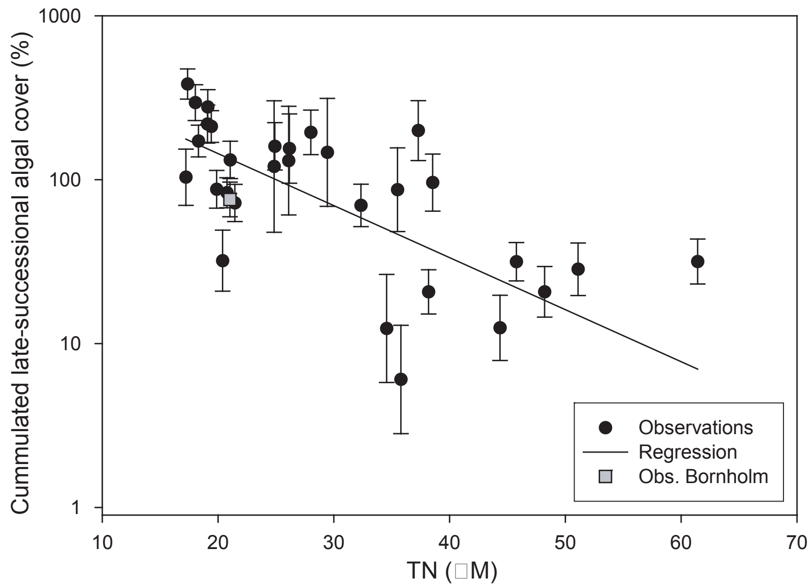

Figure 3.14. Cumulated cover of late-successional algae in relation to concentrations of total nitrogen (TN).

3.5 Discussion

The study demonstrated significant relationships between several algal variables and water quality and the relationships typically had high explanatory power. These results indicate that our strategy with focus on algae from deeper, light-limited waters and exclusion of algae from shallow exposed waters was useful and rendered the algal indicators sensitive to changes in water quality.

3.5.1 Algal variables as indicators of water quality

Cumulated cover and total cover of the algal community increased as water quality improved. When concentrations of total nitrogen decreased by 1 µM, the modelled cumulated cover generally increased 4.2% and total cover increased 2.2%. These results confirmed our hypothesis and proved that these algal variables are useful indicators of water quality. The generated models could explain almost 75% of the variation in both of these algal variables and most of the variation was explained by water quality. Regarding total algal cover, 73% of the variation among sites was explained by the concentration of total nitrogen. This makes the indicator much more sensitive than identified in our previous analyses (Krause-Jensen et al. 2005) where the coupling between total cover and TN was much weaker (correlation analysis, R=0.69). The increased sensitivity is most likely due to the exclusion of data from shallow water, the subdivision of some data sets into inner and outer estuaries and that the analysis was based on the most recent data sets where more uniform and well-defined sampling methods were applied.

Regarding the composition of the algal community, the study showed that the cumulated cover of late-successional species also increased as water quality improved and the models could explain >50% of the variation in cover levels between areas. This algal variable can thereby also be considered a useful indicator of water quality though the available explanatory models are not as strong as for the cumulated and total cover of the algal community.

In contrary to our hypothesis, the cover of opportunistic and green opportunistic algae also increased as water quality improved. Thus they followed the same trend as the cumulated and total cover of the algal community and as the cumulated cover of late-successional species. This is in contrast to many earlier findings that have demonstrated a relative stimulation of opportunistic species at high nutrient levels both through nutrient addition experiments (e.g. Pedersen 1995), in large-scale comparisons (Duarte 1995) and over long timescales (Middelboe & Sand-Jensen 2000). Recently the cover of opportunists versus the cover of late-successional species has also been suggested as a useful indicator of water quality under the Water Framework Directive (Orfanidis et al. 2001 & 2003). Most of such studies have, however, concentrated on algae growing under light saturated conditions that allow a full exploitation of the large growth potential of opportunistic algae. In contrast, our study focused on algae growing in deeper, light limited waters. This may be the reason why we see a positive effect of improved water quality and clarity on the cover of opportunists. When light levels do not saturate growth, the opportunistic algae cannot realise their potentially high growth rates. As these algae are subject to a high grazing pressure because of their high nutrient content, they risk grazing control (Geertz-Hansen et al. 1993) and this risk increases when growth rates are reduced. The positive effect of improved water quality on cover of opportunistic macroalgae in deeper water can thus be explained by the better light conditions for growth as nutrient concentrations decrease.

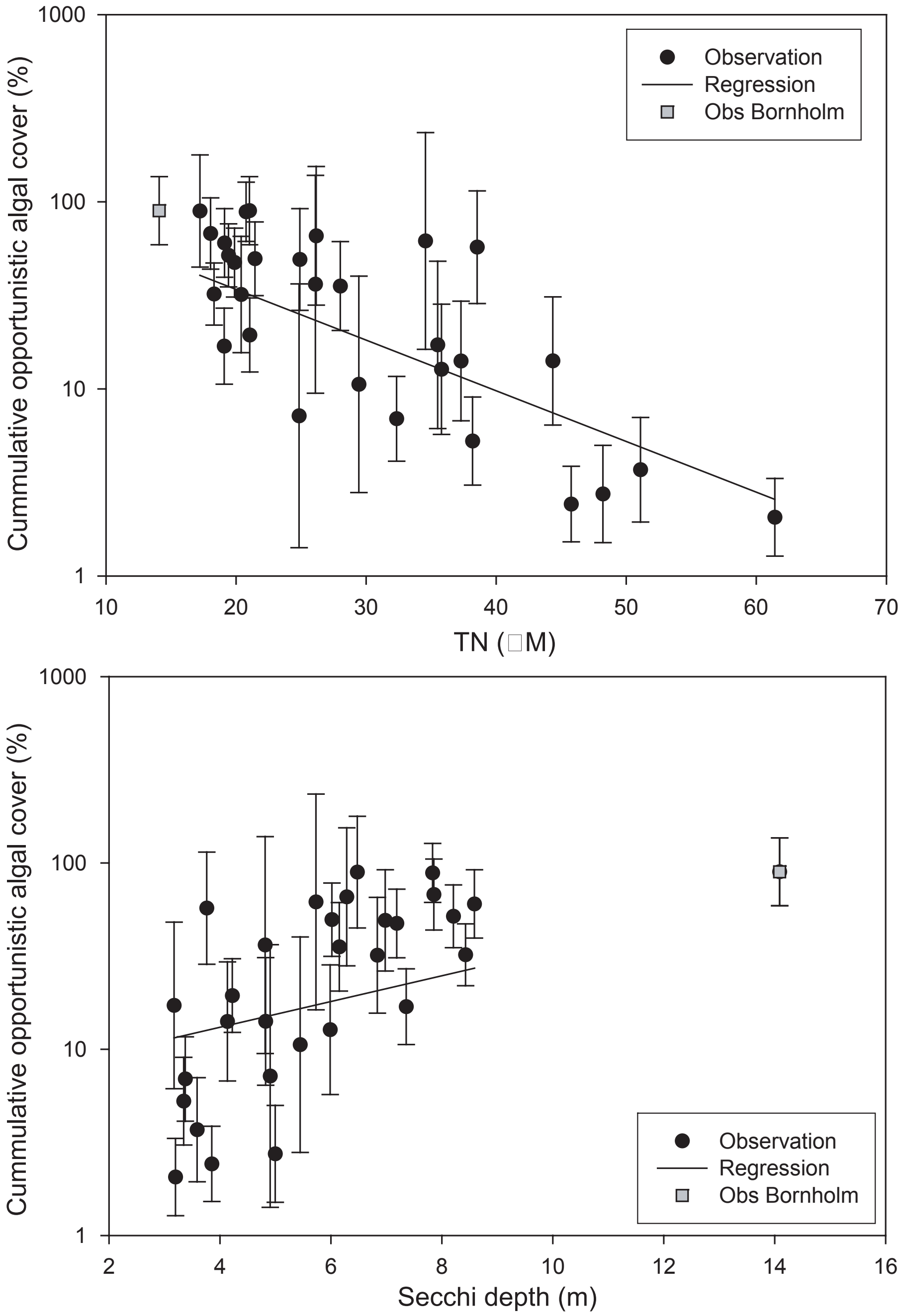

Figure 3.15. Cumulated cover of opportunistic algae in relation to physico-chemical variables: A) Algal cover versus TN, B) Algal cover versus Secchi depth.

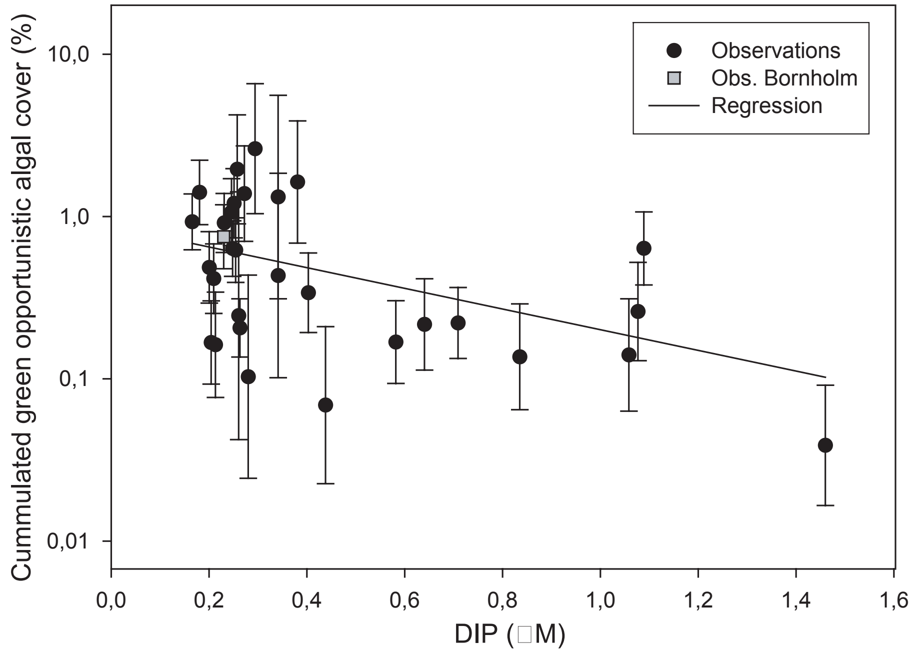

Figure 3.16. Cumulated cover of green opportunistic algae in relation to concentrations of inorganic phosphorus (DIP).

The fraction of opportunistic algae was unrelated to water quality and depended only on salinity. This independence of water quality arises because the fraction of opportunists is calculated as the ratio of two variables (cumulated cover of opportunists divided by cumulated cover of all species) which both increase with increasing water quality. The fraction of opportunists can therefore not be used as an indicator of water quality for deeper Danish coastal waters in general. In shallow coastal areas where light is not a limiting factor, where physical exposure does not export these algae, and where no strong salinity gradients exist, the cover of opportunists as well as the fraction of opportunists should increase with increasing eutrophication.

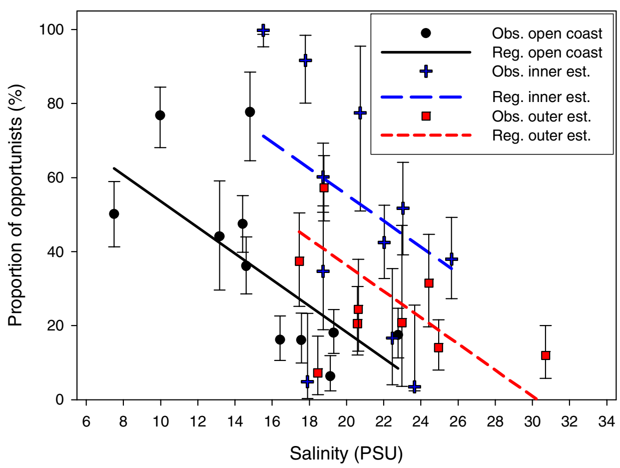

Figure 3.17. Fraction of opportunistic algae in relation to salinity. Data from different area types are fitted individually.

3.5.2 Possible thresholds in the response of indicators to changes in water quality

Our analyses have been conducted using linear methods. However, the decline in total algal cover with increasing TN concentration did suggest a threshold effect because cover tended to drop markedly at concentrations of 35-40 µM (Figure 3.13). This pattern of response could be a consequence of the fact that total cover levels have a lower limit of 0% and an upper limit of 100% which forces the distribution of cover data towards an S-curve. However, cumulated cover levels, which do not have a similarly distinct upper distribution limit, also showed signs of a threshold response to TN concentration (Figure 3.11). In order to verify whether a real threshold effect, in mathematical sense, exists in these data it is necessary to go deeper in the analyses and formulate and test a mathematical non-linearity.

A threshold response of the algae to increasing nutrient concentration could be a consequence of light levels becoming critical to the large, canopy forming and structuring macroalgae which then disappear and cause a reduction in diversity and cover of the community. Threshold effects have also been demonstrated in e.g. lake ecosystems where increases in nutrient concentrations above a certain level cause a sudden shift from macrophyte towards phytoplankton dominance (Scheffer et al. 2001). Information on possible threshold nutrient levels is important from a management point of view as there are clear advantages connected with maintaining nutrient levels below the threshold.

3.5.3 Salinity effects

Salinity also affected several of the tested indicators. Cumulated cover increased significantly with increasing salinity. This pattern may be due to the fact that species number increases with increasing salinity as more species are adapted to marine than to brackish conditions (Nielsen et al. 1995). Larger diversity could have a positive effect on cover of the algal community because the many species representing various life forms and forming a multi-layered community should be able to exploit the incoming light more efficiently and thus be more productive and dense than a less diverse community (Spehn et al. 2000). The methodology used in our study also generates a higher likelihood of obtaining high levels of cumulated cover when more species are present, because even if one species grows in more layers, its maximum possible cover is 100%. But large diversity is not a universal prerequisite for high cover. For example, *Fucus vesiculosus* often has a high cover in the brackish areas of the species-poor Baltic Sea, and in underwater forests of large brown macroalgae like *Laminaria sp.* or *Macrocystis sp.* the thalli of these genera often obtains a cover of several 100% (Lüning 1990 and references therein), which may, of course, be further increased by other species in these communities.

Salinity also tended to have a positive effect on the cover of late-successional species and a negative effect on the cover of opportunistic algae. These opposing, but non-significant trends, resulted in a significantly negative effect of increasing salinity on the fraction of opportunistic macroalgae. This pattern is most likely due to an increased diversity of late successional species and a decreased diversity of opportunistic algae as salinity increases (Nielsen et al. 1995).

3.5.4 Assessing water quality according to the Water Framework Directive

The identification of biological indicators that respond to changes in water quality is a first important step towards assessing water quality according to the Water Framework Directive (WFD, see box 1). The most useful macroalgal indicators identified in this study were 'the cumulated cover of the macroalgal community' and 'the total cover of the macroalgal community'. In order to use these indicators under the WFD, it is necessary to identify 'reference conditions' and 'ecological quality classes' for each of the indicators and identify whether the level of the indicators differs between water body types.

Box 3.1. Assessing water quality according to the Water Framework Directive (WFD)

> The WFD aims to achieve at least a good ecological status in all European rivers, lakes and coastal waters and demands that the ecological status is quantified based primarily on biological indicators, i.e. phytoplankton and benthic flora and fauna. The WFD thereby challenges us to identify useful biological indicators that respond predictably to human impact and can be quantified with sufficient precision. The WFD demands that ecological status is quantified and expressed as a so-called 'Ecological Quality Ratio' (EQR), defined as the ratio between the actual level of a biological indicator and the reference level of the indicator. The reference level or reference condition is defined as the level of the indicator in an 'undisturbed' ecosystem with 'no or only very minor' anthropogenic influence. Ideally, reference levels should be defined based on information on existing, undisturbed water bodies, but widespread eutrophication is typically a hindrance to this approach and makes it necessary to define reference levels based on historical data, modelling or expert judgement instead. According to the WFD reference levels must be defined for so-called water body types, defined by physical characteristics of the water bodies, and the classification thereby becomes 'type-specific.'
>
> Depending on the degree of deviation from reference levels, the WFD defines five 'ecological status classes': 'high status,' 'good status,' 'moderate status,' 'poor status' and 'bad status'. 'High status' is obtained when the biological indicators meet reference levels and have EQR values close to 1. 'Good status' is achieved when the biological indicators differ only slightly from their reference levels. At moderate, poor and bad status the biological indicators show moderate, major and severe deviation from reference levels, respectively. As the WFD requires that all European surface waters must reach at least 'good status,' definition of the boundary value between good and moderate status is of utmost importance. Regarding coastal benthic flora, the WFD defines ecological status based on species composition, cover and abundance of seagrasses and macroalgae.

The most correct way of assessing reference levels would be to use historical data representing algal cover during a period with low nutrient loads. However, such data do not seem to exist. Instead, the actual level of the algal indicators in the most oligotrophic/least eutrophic areas could be used as a minimum estimate of reference conditions taking into account that reference levels should be graduated according to salinity. Another way of assessing reference levels for the algal indicators is using our developed model to hindcast reference conditions. Reference levels of nutrient concentrations or Secchi depths can be entered in the generated models and corresponding reference levels of algal cover for the specific areas included in the model can then be calculated. Reference nutrient or light levels can be entered in the model as general values or, better, as specific values for given areas. Time series of nutrient inputs to Danish coastal waters going back to the beginning of the 20th century have recently been estimated (Conley et al. subm.) and reference nutrient concentrations may be derived from this in combination with other reference material. Estimates of reference Secchi depths are also available from Ostenfelds (1908) occasional measurements around year 1900 which ranged from 3.7-5 m in estuaries/smaller sounds to ~10 m in Kattegat (Table 3.6). Observations of eelgrass depth limits down to almost 11 m in Kattegat do however suggest that even larger Secchi depths (>13 m) are likely to have prevailed at the time (based on model by Nielsen et al. 2002 describing eelgrass depth limits as a function of Secchi depth). See also chapter 2 of this report which presents a larger compilation of historic Secchi depths.

Table 3.6. Secchi depth recorded in the summer of 1901 (Ostenfeld 1908, p. 18 and appendix). Secchi depths were measured using a white plate of the dimension 20 cm x 15 cm which was lowered down through the water column to the largest depth where it was still visible. The historic Secchi depths were measured in 'favne' and converted to meter using the same conversion factor as Ostenfeld: 1 favn=1.83 m.

Site	Date	Secchi (m)
Kattegat		
- North of Randers Fjord (Sødringholm skov)	31.07.01	10.1
- Kattegat, Als church	31.07.01	9.15
Baltic Sea		
- Fakse Bay (Central)	06.08.01	8.2
- at Falster	05.08.01	7.32
- at Vigersløse church	05.08.01	7.3
Archipelagoes and larger sounds		
- Smålandshavet (Helleholm Lighthouse)	07.08.01	9.15
- Langelandsbælt	1/8.01	9.15
Estuaries and smaller sounds		
- Limfjorden, Nissum Broad	29.07.01	5
- Limfjorden, Løgstør Broad (Livø)	30.07.01	3.66
- Guldborgsund, Skjelby Kirke, Gedser Havne Fyr	05.08.01	4.8
- Guldborgsund	05.08.01	5.49

Once reference levels of the selected indicators are identified it is possible to define quality classes for the indicators and thus develop a classification scheme where boundaries between quality classes are defined as % deviances from reference levels. As the WFD requires that all European surface waters must reach at least 'good status,' definition of the boundary between good and moderate status is of utmost importance. A deviance of 15%, 20% or 25% from reference levels has been suggested for other indicators (e.g. Dahl et al. 2005) and may also be applicable for the macroalgal indicators.

When using the developed macroalgal indicators in practice, total algal cover and cover of individual algal species are measured in 2-m depth intervals from the coast towards deeper water. Levels of hard substratum are recorded simultaneously. Our analyses underline the importance of minimising diver effects and focus data collection on the most stable substratum. In data analysis, the depth representing maximum algal cover is defined and algal data from shallower water depths are discarded. The actual cover levels at specific water depths are then compared with the modelled reference levels for the area in question and compared with the classification scheme.

3.6 Conclusion and perspectives

In conclusion, the total cover and the cumulated cover of coastal macroalgal communities have been identified as suitable indicators of water quality. If appropriate reference levels for these indicators are defined, the indicators can be used to assess water quality according to the Water Framework Directive given that quality classes can be defined with sufficient accuracy.

The indicators can be further explored through temporal analyses of relations between indicator levels and water quality on the large data set from the Danish monitoring programme using the technique of omitting the shallow-water data and focusing on the depth range where the algae are light limited. Such analyses would reveal whether the indicators reflect the smaller-scale changes in water quality, which have occurred over the monitoring period from 1989 till now. Analyses of possible threshold levels would also be highly relevant from a management – as well as from a scientific perspective.

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4 Development of tools for assessment of environmental quality using macrobenthic fauna

By Alf B. Josefson & Jørgen L.S. Hansen

4.1 Introduction

Macrobenthic fauna is one of the quality elements to be used in the implementation of the Water Framework Directive (WFD). Benthic fauna responds to changes in pressure variables both natural and man made. A major challenge for environmentalists is to separate effect of natural and effects of man induced change in the environment. This study is a part of a wider project with the aim to facilitate implementation of the WFD in Danish waters. A first step is to identify suitable metrics of benthic fauna to describe impacts from pressure variables. One way to do this is to test existing metrics in gradients of pressure variable (like oxygen deficiency). If a metric responds as expected to a pressure variable, the dose-response relationship should be identified. Having such a relation the faunal response should be divided into 5 classes of environmental quality from High to Bad, with High corresponding to, or being close to, a reference condition. Establishing a reference condition, however, is not a trivial task. Since it probably is difficult to find comparable areas in present time unaffected by man, the reference condition should ideally be established using historical data; that is data sampled before significant human impact. In Danish waters there are historical benthic fauna data starting from the beginning of the previous century taken by the Danish Biological Station. Revisits of historical sites in recent time have for instance been made in the Kattegat (e.g. Pearson et al. 1985; Josefson and Jensen 1992), the Skagerrak (Rosenberg et al. 1987), Swedish fjords (Josefson and Rosenberg 1988), the Øresund (Göransson 2002), and areas around the island of Funen (E. Glob pers. comm.). In the Limfjord there are data in a virtually unbroken time series from 1909 until now (Christensen et al. 2005). These studies tell us that species composition has changed to some extent and that biomass often has increased over time. While it is possible to use biomass and possibly sensitivity of some larger animals for a reference, it is not possible for abundance and diversity measures due to the methodological differences.

Many attempts have been made over the years to describe environmental quality/status using various numerical expressions - so called indices or metrics. Several of these indices incorporates diversity aspects and two of the earliest ones were the Shannon wiener index (Shannon & Weaver 1963) and the Margalefs richness index. In recent years indices based on the expected number of species in a random sample of individuals (Hurburt's ES, Hurlburt 1971) have been proposed. Here the sensitivity of the species is judged from how often species occur in a low or high diversity environment (Rygg 2004;

Rosenberg et al. 2004). Another type of index was proposed by Borja et al. (2000), a development of index by Grall and Glemarec (1997), the AMBI index which is based on sensitivity classification of species from occurrence in relation to pollution sources. Still other indices combine both diversity and functional aspects (Weisberg et al. 1997) into one metric.

The purpose with this work is to evaluate some of the earlier proposed indices on Danish data with the final objective to arrive at a/some useful indices applicable on Danish conditions. The approach is to examine changes in some of these indices along an environmental gradient - specifically a gradient of oxygen deficiency. Seasonal oxygen deficiency is one of the major problems in Danish waters and it therefore seemed natural to choose this impact factor to test these indices. The pressure gradient we use in this report is frequency of low oxygen concentrations in the bottom water in autumn. This is related to nutrient concentrations and consequently eutrophication.

The work is the first test of the AMBI index on Danish monitoring data, and is a part of the work to evaluate several different measures of benthic quality and to establish limits for environmental quality in Danish waters.

We used data from 11 of the areas monitored in the NOVA programme (Danish National Monitoring and Assessment Programme). The areas (Figure 4.1), all situated in polyhaline to euhaline waters (> 18 psu), were ranked from severe to benign with respect to frequency of low oxygen in the bottom water (< 2ml/l). Data used for the ranking were from the 1990s and presented in Josefson & Hansen (2004). Areas ranged from those hit by severe seasonal hypoxia nearly each year to those who never experience hypoxia. As an example the Lillebælt N area about 25% of the oxygen measurements in August-October were less than 2 ml/l.

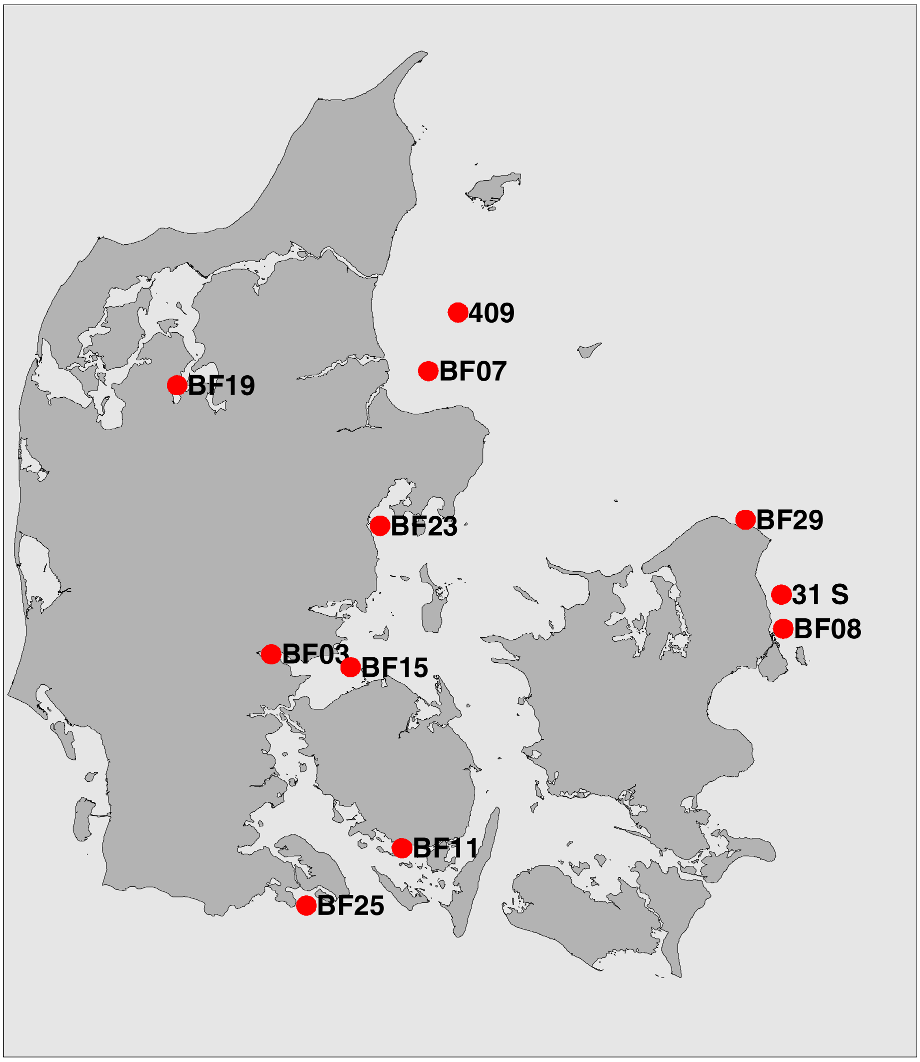

Figure 4.1. Map showing positions of the sampled areas/stations.

From these sites we used annual benthic fauna data from the period 1999-2003 and all sites except Århus Bay were sampled in spring months (April – June). Århus Bay data were from autumn (September, October).

For each sample (year and site) we calculated the following indices:

1. The average number of species per sample unit (i.e. ca 0.0125 m^2),
2. The Margalef's richness,
3. The Shannon wiener index with log e,
4. The number of species in 20 sample units (haps) determined from randomised species area curves,
5. The AMBI biotic index as described by Borja et al. (2000).

These indices were regressed against each other and the rank order with respect to oxygen deficiency. If diversity metrics and AMBI reflect environmental quality with respect to oxygen levels we expect a correlation between the two types of indices and furthermore both should correlate with the rank order with respect to oxygen deficiency.

Table 4.1. Physico-chemical parameters for the 11 coastal areas ranked from the most severely affected to the least with respect to oxygen deficiency. Water residence time (TD), Area specific annual N-loading (TN), Mean water depth for the sampling points (Z), Median (MSAL) for the whole year, Median (MOX), coefficient of variation (CVO) and the percentage of measurements < 2 ml l^{-1} (POX) of oxygen concentrations in the months August-October, number of measurements of oxygen concentrations (N oxygen).

Rank	Area	TD (days)	TN (g $m^{-2}yr^{-1}$)	Z (m)	MSAL (psu)	MOX (ml l^{-1})	CVO	POX (%)	n oxygen
1	Lillebælt N		.	19.4	27.48	2.94	39.3	24.8	90
2	Flensborg Fjord	50	2.4	20.5	20.48	2.10	56.1	24.1	2688
3	Skive Fjord	100	33.9	4.4	25.54	5.04	51.2	19.0	409
4	Ringgårdsbassin		3	14.6	19.00	4.55	38.6	15.0	1428
5	Århus Bay	12	5.6	15.2	27.80	3.43	38.9	14.0	385
6	Vejle Fjord	16	36.6	6.3	22.86	5.53	30.2	5.8	1404
7	Hevring Bay			12.1	27.87	4.86	17.7	0	370
8	409					>5		0	
9	Øresund		.	14.1	23.23	5.18	20.4	0	251
10	31S			17		>5		0	
11	Hornbæk		.	27.9	32.00	>5		0	

Table 4.2. Pearson correlation matrix: Rank order = rank number from the most severe (1) to the most benign (11), Grp V = Group V species in the AMBI index, BI = The AMBI biotic coefficient, Abundance= number of individuals in a sample unit (Haps), Species nr/sample = Number of species per sample unit (Haps), Margalef's R = Margalef's species richness index (number of species/10log (nr of individuals), elog H = Shannon wiener index with the base elog, ns = P>0.05 Bonferroni probabilities. Most other correlations are significant at the 1% level (P<0.001).

n=53	Rank order	Grp V	BI	Abundance	Species No./sample	Margalef's R
Rank order						
Grp V	-0.40					
BI	-0.60	0.73				
Abundance	0.25 ns	-0.21 ns	-0.16 ns			
Species No./sample	0.57	-0.50	-0.58	0.73		
Margalef's R	0.60	-0.59	-0.66	0.58	0.97	
elog H	0.55	-0.62	-0.65	0.49	0.91	0.96

4.2 Results

Variation in the AMBI index over some years is shown from two end-points in the hypoxia gradient, one from the most severe conditions BF15 in the Northern Lillebælt (Figure 4.2) and one from areas with no reports of oxygen deficiency, Station 31S in the Øresund (Figure 4.3). There is a clear difference between the two sites with respect to composition of sensitive (blue species) and less sensitive species (for example yellow species. At station BF15 the AMBI index is close to the border between Good and Moderate Ecological quality (Slightly polluted and Meanly polluted in Figure 4.2). At the station 31S in the Øresund the index was close to the border between High and Good ecological quality most of the time (Unpolluted and Slightly polluted in Figure 4.3).

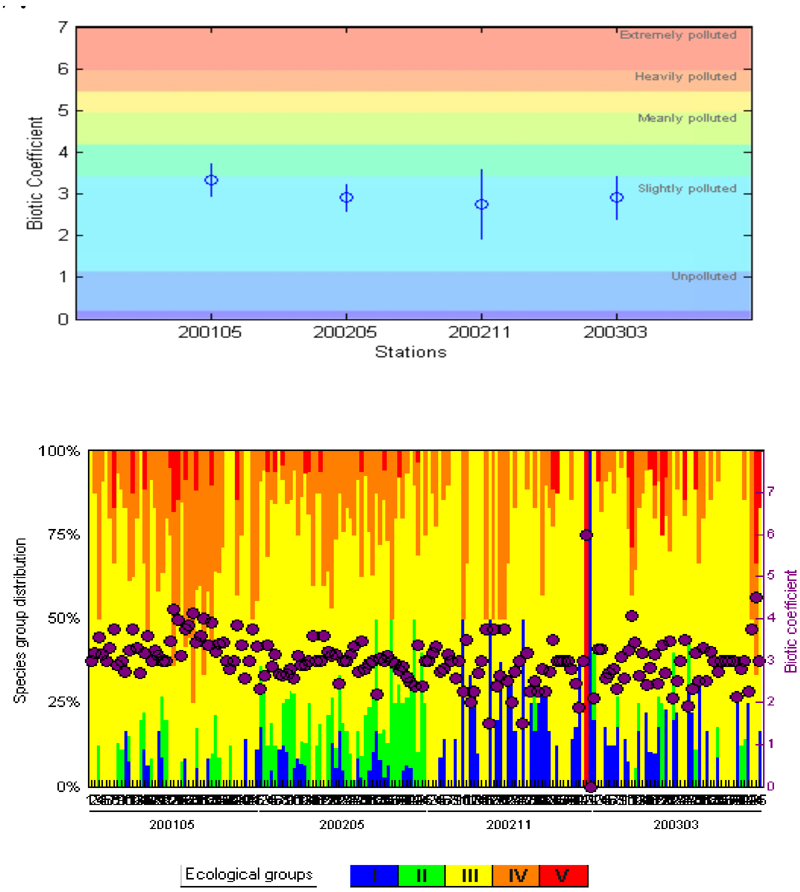

Figure 4.2. Variation in the AMBI biotic coefficient at station BF15 in the Northern Lillebælt. Upper graph shows the temporal variation over 2 years (sample means with SD) and lower graph the variation between individual sample units (haps). The station has Rank number 1 in the gradient of hypoxia and the fauna is highly dominated by more tolerant species (yellow colour) compared to station 31S. The ecological status lies close to the border between Good and Moderate in the actual period.

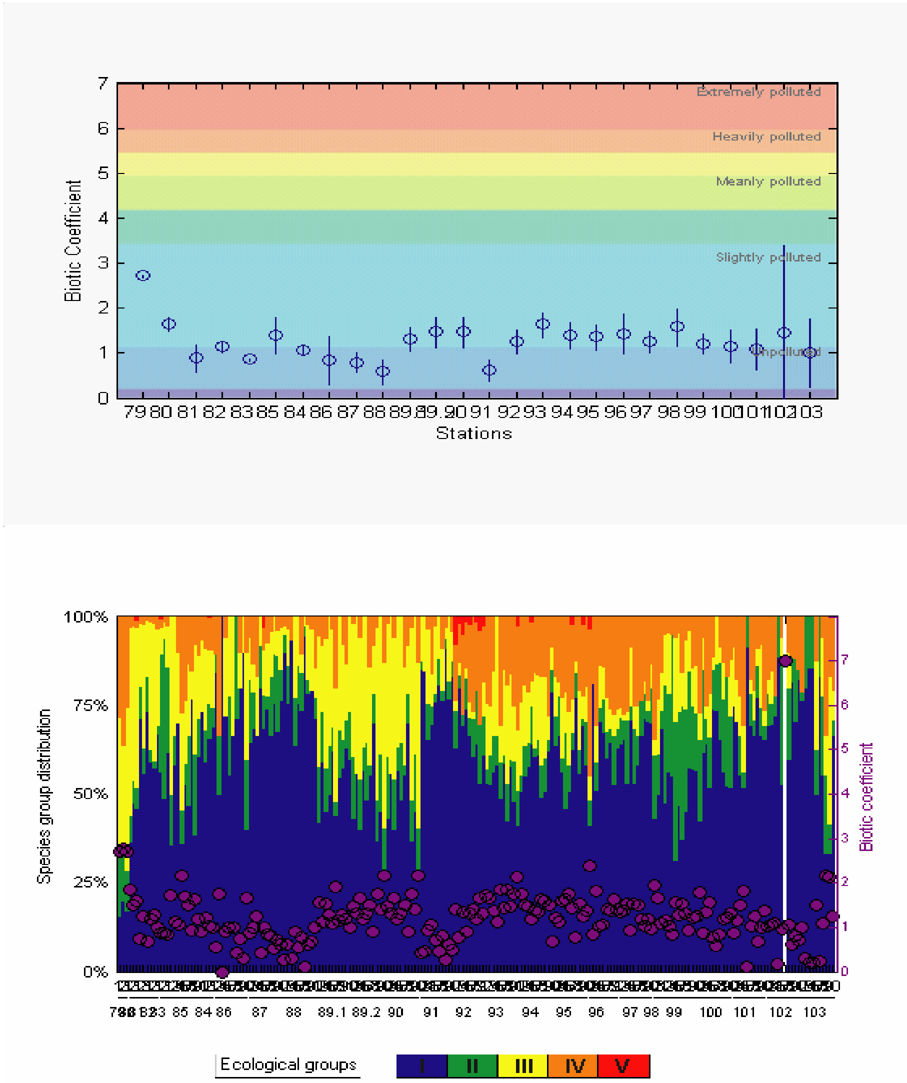

Figure 4.3. Variation in the AMBI biotic coefficient at station 31S in the Øresund. Upper graph shows the temporal varaition over 25 years (annual means with SD) and lower graph the variation between individual sample units (haps). The station has Rank number 10 in the gradient of hypoxia and it is evident that the fauna is highly dominated by sensitive (blue colour) species. The ecological status lies close to the border between High and Good environmental quality most of the time.

The correlation analysis showed, not unexpectedly, that the diversity based metrics, Alpha diversity, Margalef's richness and Shannon's H were strongly positively correlated with each other (Table 4.2). Less expected was the strong negative correlation between the diversity based indices and the AMBI index (Table 4.2, Figure 4.4). This means that high diversity goes together with a low AMBI index indicating high quality of the benthic environment and vice versa. Although not very different, the best correlation was found between AMBI and Margalef's richness, and index where number of species is "normalised" with respect to number of individuals. Using the translation from benthic community health classes determined by AMBI to the WFD Ecological status classes as suggested by Muxika et al. (2005), most of the data fall within the classes High (H), Good (G) and Moderate (M). The great majority of data fall, within the class Good (Figure 4.4).

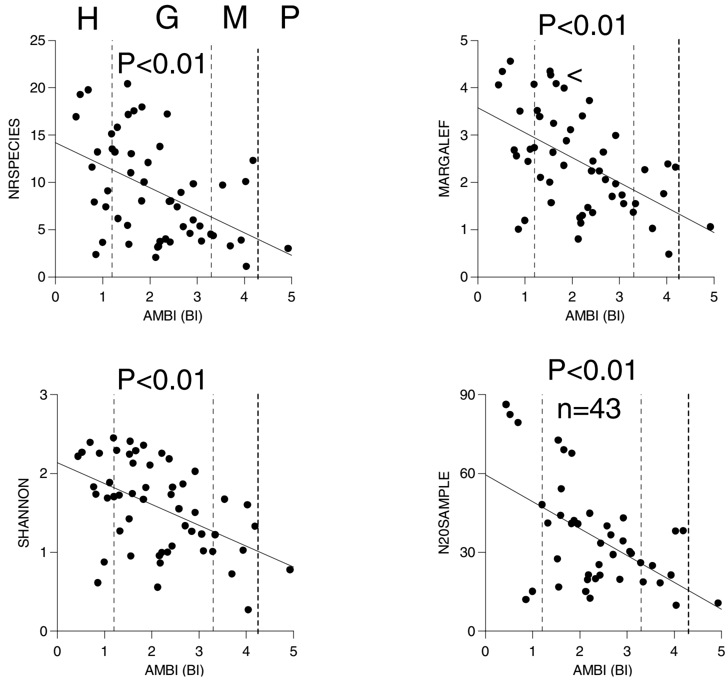

Figure 4.4. Plots with regression lines of the diversity measures Number of species per sample, Maragalef' s richness, Shannon's H (elog base)and The number of species in 20 samples versus the AMBI index (BI). Vertical dashed lines delimit the Ecological Status areas High (H), Good (G), Moderate (M) and Poor (P) as suggested by Muxika et al. (2005). Significance levels are based on Bonferroni probabilities for the Pearson correlation coefficients (7 variables, n= 53).

Now, how do these metrics relate to the pressure variable hypoxia? Both the diversity measures and AMBI where significantly correlated with rank numbers based on hypoxic conditions (Figure 4.5). Diversity was low and AMBI high were oxygen levels were low (e.g. Northern Lillebælt) and diversity was high and AMBI low in areas without hypoxia (e.g. BF29 in the Northern mouth of Øresund). Thus, the metrics behave as expected along the pressure gradient of hypoxia.

4.3 Conclusion

These results indicate that both diversity-based indices like alpha diversity and Margalef's richness or Shannons H, and the sensitivity based index AMBI can be used to evaluate quality status of benthic communities in Danish waters. This applies so far to bottoms in the salinity regime >18 psu i.e. polyhaline and euhaline waters. It is likely that a different classification scheme has to be developed for mesohaline areas (5-18 psu) since diversity naturally is much lower in these areas. Future work will involve attempts to construct a single environmental quality index for Danish bottom fauna. The successful test of AMBI and diversity measures in this report have warranted a combination of AMBI and diversity into one multimetric Danish index - the DKI index. The index, to be described in detail in Borja et al. in prep., is similar to the index used in the UK, and is a summation of AMBI and the Shannon Wiener diversity (H) standardised to obtain values between 0 and 1. Preliminary work shows that the index behaves reasonably in the oxygen deficiency gradient, that is increases with decreasing frequency of oxygen deficiency. Future work will compare this index with other indexes used in Europe and with other measures of quality specifically the depth of the oxidised layer in the sediment.

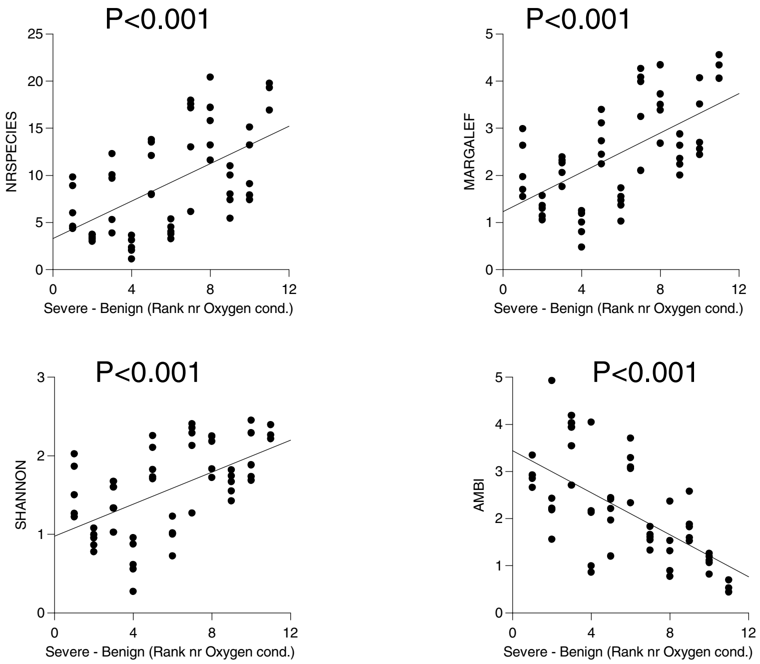

Figure 4.5. Plots with regression lines of the metrics Number of species per sample, Maragalef's richness, Shannon's H (elog base) and the AMBI index (BI) versus the rank number of stations. The stations were ordered from the most severe (No. 1) to the most benign (No. 11) with respect to oxygen conditions in the bottom water. Significance levels are based on Bonferroni probabilities for the Pearson correlation coefficients (7 variables, n= 53).

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5 Overall conclusion and perspectives

According to the WFD the future assessment of water quality, that is ecological classification of coastal water quality, is to a.o. be assessed by the biological quality elements phytoplankton, macroalgae and macrobenthos and more specifically by means of different indicators within the elements. The analysis and identification of biological indicators that respond to changes in water quality, which implies the usefulness of these, is a first important step towards establishing reference conditions and thereby also towards assessing water quality according to the WFD.

The indicators for phytoplankton as presented by the WFD are biomass, species composition, abundance and bloom frequency/intensity. In this report focus has been on biomass represented by chlorophyll *a* and the Secchi depth-Chl *a* relationship has been analysed with regard to present and historical data.

While the present work do not completely fulfill the objectives of establishing reference conditions for phytoplankton biomass the results on the Secchi depth-Chl *a* relationships provide an important first step along the track of achieving reference conditions for phytoplankton in Danish coastal waters.

As a complement to the analysis of biomass and Secchi depth a similar analysis of the relationship between Secchi depth and total nitrogen (TN) was performed. While the rationale behind the analysis is sound, methodological problems i.e. the lack of sufficient TN data was probably the reason why the correlation failed to present reliable data.

With respect to this result it is important to bear in mind that Chl *a* is still the most commonly used proxy for phytoplankton biomass and that it has been chosen as the first phytoplankton metric for the WFD intercalibration process (see appendix 1). However, in addition to Chl *a*, phytoplankton species composition, abundance and bloom frequency/intensity should be included in the future assessment of water quality. At present, reference conditions are not available for these indicators and future work remains.

The ecological status based on the biological quality element macroalgae should be based on species composition, cover and abundance. The analysis in this report identified that the most useful indicators and therefore the most suitable in assessing water quality are 'the cumulated cover of the macroalgal community' and 'the total cover of the macroalgal community'. Assuming that appropriate reference levels for these indicators can be identified, these indicators can be used to assess water quality according to the WFD.

The study on macroalgae also revealed specific points which should be taken into account in future analyses and management strategies. I.e. a specific focus on algae from deeper, light-limited waters and thereby exclusion of algae from shallow exposed waters was useful as it rendered macroalgal indicators sensitive to changes in water quality. Furthermore, the analyses indicated that it is important to try and minimise diver effects and at the same time focus data collection on the most stable substratum. An indication of a possible nutrient level threshold in the correlation between algal cover and TN points at the need for further analyses not least because the knowledge on nutrient threshold limits will be very important from a management point of view.

Macrobenthos indicators according to the WFD are composition and abundance of benthic invertebrate fauna. In this report quality status described by diversity based indices as well as sensitivity based indices has been evaluated with a result that indicates that both types of indices can be used as an evaluation tool on quality status of benthic invertebrate fauna in Danish waters. As the evaluation only concerns macrobenthic communities in salinities >18 the question whether a different classification scheme has to be developed for areas of lower salinity (5-18) remains unanswered. The results provide a tool for the future work that o.a. will include an attempt to construct a single environmental quality index for Danish bottom fauna, which eventually will be calibrated with similar indices (metrics) from other European countries.

As this sum up of the conclusions and perspectives from the work on the three biological quality elements phytoplankton, macroalgae and macrobenthos shows we are in general not yet at the point where it is possible to set reference condition for the biological indicators in Danish waters according to the WFD. Still the results and the tools developed present a useful step in the right direction of assessing values for the boundaries between ecological status classes.

Annex 1

Annex data for Figure 3.11

Coastal area	Fjord type	Secchi depth (m)	salinity (psu)	TN (µM)	Mean cumulated cover (%)	mean - C.L.	mean + C.L.
Augustenborg Fjord	Inner Fjord	5,447490783	17,90075557	29,43595	111,9998989	54,00376509	232,2796815
Augustenborg Fjord	Outer fjord				78,0111003	47,03697313	129,3818748
Bornholm West	Open coast	14,09049084	7,500254672	21,03303	140,4131665	113,6062121	173,545592
Bornholm East	Open coast				121,8089093	92,75594356	159,9618291
Flensborg Fjord	Inner Fjord	5,986819493	17,78912023	35,79788	15,14646347	9,902368635	23,16772524
Flensborg Fjord	Outer fjord	6,980574432	17,45980744	24,91122	119,6476866	85,79415663	166,8594863
Fyns Hoved	Open coast	7,857508611	17,57209529	18,03557	287,6415577	230,7876332	358,5012965
Horsens Fjord	Inner Fjord	3,17409393	22,46132707	35,50386	65,8193822	37,77223256	114,6924812
Horsens Fjord	Outer fjord	4,81440045	22,97548213	26,09787	120,6593963	58,02796876	250,8909103
Isefjord	Inner Fjord	5,731680756	20,73601547	34,55647	70,70614283	34,13887218	146,4418218
Isefjord	Outer fjord	6,151680756	20,58859929	28,02021	133,7513173	101,0098199	177,1057001
Kalundborg Fjord	Inner Fjord	6,019904607	18,7413908	21,45552	67,14856806	52,80282253	85,39184038
Kalundborg Fjord	Outer fjord	7,189119867	18,78812917	19,88273	76,34120756	61,2863983	95,09418292
Karrebæksminde Bay	Open coast	6,837207048	13,17066346	20,39214	74,52790908	51,09147064	108,715
Kirkegrund & Knudshoved	Open coast	7,833322626	14,41270408	20,76797	149,3902408	124,5251853	179,2203238
Køge Bay	Open coast	4,223701531	9,973274448	21,05579	25,1440911	20,07782295	31,48873853
Lillebælt	Open coast	8,210100344	16,42367427	19,42054	235,8837092	195,1431328	285,1298095
Limfjorden, Mors NW	Inner Fjord	4,998255956	23,03722592	48,21126	7,894765179	5,842415221	10,66807388
Limfjorden, Mors W	Inner Fjord	3,347899519	25,64947697	38,17975	19,36494262	14,63458726	25,62429649
Løgstør Broad	Outer fjord	3,852156772	24,94933908	45,76899	23,90275333	18,9838646	30,09617003
Nissum Broad	Outer fjord	3,375682673	30,7053843	32,3473	57,9691076	44,29729877	75,86054972
Nivå Bay	Open coast	6,476724177	14,81081952	17,22742	83,22586239	58,0377107	119,3455787
Sealand N	Open coast	7,668103356	19,30855249	17,36715	251,4748968	206,0378393	306,9320856
Odense Fjord	Outer fjord	3,763774877	20,62942168	38,53951	91,06833282	63,58259516	130,4357147
Roskilde Fjord	Inner Fjord	4,824340824	15,52205143	44,35787	18,64742019	12,23052993	28,43100682
Roskilde Fjord	Outer fjord	4,135028856	18,45833638	37,29434	111,0605297	75,20375141	164,0136433
Sejerø Bay	Open coast	7,358782669	19,12647843	19,09006	172,1942167	134,5859715	220,3115817
Skive Fjord	Inner Fjord	3,198305905	22,02545343	61,41726	16,20194744	12,7730752	20,5512844
Skive Fjord	Outer fjord	3,588091727	24,43356456	51,09679	16,08421099	11,49834168	22,49905686
Vejle Fjord	Inner Fjord	4,904971568	23,6581028	24,84686	80,89779304	33,26365388	196,7448598
Vejle Fjord	Outer fjord				89,0720487	55,0393698	144,1482686
Venø Bay	Outer fjord		28,33194079		25,37359766	18,12360084	35,52381582
Åbenrå Fjord	Inner Fjord	6,286003256	18,74545137	26,15767	159,9530414	101,047391	253,1977836
Åbenrå Fjord	Outer fjord				125,3842457	66,26632392	237,2428128
Århus Bay	Open coast	8,587925459	22,77666095	19,1119	243,4531163	198,7649528	298,1884835
Århus Bay	Inner Fjord				209,6703203	163,8498004	268,3045272
Århus Bay	Outer fjord				222,9905467	166,7155913	298,2611497
Øresund	Open coast	8,427980928	14,60974002	18,30557	90,53678693	75,84991142	108,0674932

Annex data for Figure 3.13

Coastal area	Fjord type	TN (µM)	mean cover	mean - C.L.	mean + C.L.
Augustenborg Fjord	Inner fjord	29,43595441	97,1507662	78,09199119	97,82914533
Augustenborg Fjord	Outer fjord		87,35941257	69,36383905	98,04128983
Bornholm West	Open coast	21,03303455	95,2627103	90,13995357	98,58093703
Bornholm East	Open coast		91,42397395	82,81740587	97,23708996
Flensborg Fjord	Inner fjord	35,79787882	7,012804482	0,592640799	19,62702057
Flensborg Fjord	Outer fjord	24,91121861	95,6501907	87,64598619	99,62770074
Fyns Hoved	Open coast	18,03557112	98,88524659	95,64580587	99,99981876
Horsens Fjord	Inner fjord	35,50386157	90,72900713	72,25791659	99,5900815
Horsens Fjord	Outer fjord	26,09786909	99,81049464	87,39710908	92,5828888
Isefjord	Inner fjord	34,55646739	56,02085136	25,37897317	84,3113263
Isefjord	Outer fjord	28,02021007	99,41978239	95,79876265	99,70932432
Kalundborg Fjord	Inner fjord	21,45552168	82,38805934	73,46066531	89,80685561
Kalundborg Fjord	Outer fjord	19,88272915	85,85862015	78,14087743	92,11893533
Karrebæksminde Bay	Open coast	20,39213971	83,02741458	68,58450568	93,67311791
Kirkegrund & Knudshoved	Open coast	20,76797495	97,77110514	94,49545217	99,60507046
Køge Bay	Open coast	21,05578972	56,01899498	45,19771024	66,55830726
Lillebælt	Open coast	19,42054367	98,95461808	99,98977204	96,25562418
Limfjorden, Mors NW	Inner fjord	48,21126184	7,622696765	1,868856405	16,7997103
Limfjorden, Mors W	Inner fjord	38,17974947	24,72280048	14,54593712	36,57525215
Løgstør Broad	Outer fjord	45,76898503	24,26301741	15,45845263	34,33005202
Nissum Broad	Outer fjord	32,34729737	50,45896233	38,14404884	62,74566167
Nivå Bay	Open coast	17,2274171	81,95751573	67,80950106	92,67325807
Sealand N	Open coast	17,36714939	99,4395817	99,9846986	97,39106844
Odense Fjord	Outer fjord	38,53950888	36,24749344	21,38876637	52,59544811
Roskilde Fjord	Inner fjord	44,35786572	36,42803154	19,70576125	55,04644675
Roskilde Fjord	Outer fjord	37,29434416	99,21657306	93,18882539	99,2487284
Sejerø Bay	Open coast	19,09006158	99,00915071	99,98590367	95,60185215
Skive Fjord	Inner fjord	61,4172648	9,892655867	4,181431184	17,68049855
Skive Fjord	Outer fjord	51,09678776	17,16200714	7,307252366	30,08416507
Vejle Fjord	Inner fjord	24,84686221	95,49751122	68,05859045	97,03671212
Vejle Fjord	Outer fjord		88,87246539	72,51211303	98,36250704
Venø Bay	Outer fjord		12,92198735	4,602508156	24,61899413
Åbenrå Fjord	Inner fjord	26,15766993	99,83376961	97,38399918	94,16270246
Åbenrå Fjord	Outer fjord		99,31309628	87,50418901	96,23197812
Århus Bay	Open coast	19,11189756	94,94209069	89,65042357	98,41718524
Århus Bay	Inner fjord		80,8274598	70,44273089	89,39851972
Århus Bay	Outer fjord		84,73643038	73,85581766	93,08763718
Øresund	Open coast	18,30557221	74,57948057	66,30713729	82,04261935

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563 Scientific and technical background for intercalibration of Danish coastal waters

This report contributes to the development of tools that can be applied to assess the five classes of ecological status of the Water Framework Directive based on the biological quality elements phytoplankton, macroalgae and macrobenthos. The work on phytoplankton biomass (Chl a) based on monitoring data and historical Secchi depth measurements provides a first step in establishing reference conditions for phytoplankton in Danish waters. The identification of appropriate macroalgal indicators provides the results that total cover and cumulated cover of coastal macroalgal communities are suitable indicators of water quality. The results also indicate that a strategy with focus on algae from deeper, light-limited waters and exclusion of algae from shallow exposed waters renderes algal indicators sensitive to changes in water quality. The macrobenthos project evaluates some of the earlier proposed indices on Danish data along the environmental gradient of oxygen deficiency. Results from this evaluation indicate that both diversity-based indices and the sensitivity based index AMBI can be used to evaluate quality status of benthic communities in Danish waters. The result applies to bottoms in the salinity regime > 18 psu.

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